Atmospheric pollution

# **BIOMONITORING URBAN AIR POLLUTION BY USING LICHENS IN THE GREEN SPACE OF THE UNIVERSITY CAMPUS IN OSIJEK (CROATIA)**

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**Abstract**. The city of Osijek is the administrative, educational and cultural centre of the Eastern Croatia. University of Osijek was founded in 1975, and the academic community consists of 17 337 students and 1134 teaching staff in 2015. University obliged to promote and implement principles and practice of environmental protection and sustainable development at all levels. University campus covers area of 23.2 ha, where construction of new buildings and urban planning started in 2002. In the green space of campus an inventory of trees was carried out in 2012, combined with survey of epiphytic lichen flora. A total of 365 trees classified into 21species were recorded. Planting of landscape trees and other woody plants is considered as an effective measure for increasing annual rate of carbon storage and sequestering in the campus. Epiphytic lichen flora recorded on trees consists of eight species. Foliose lichens dominates (75%), followed by crustose and fruticose lichen with equal shares of 12.5%. By applying the lichens as bio-indicators of air quality, the air in the university campus in the City of Osijek has been assessed as moderately polluted. Due to intake of dust and increased on-road emissions of nitrogen compounds, the trees are covered by nitrophilous lichens, resistant to higher eutrophication and tolerant to air pollution.

Keywords: lichen, air pollution, university, Osijek.

# AIMS AND BACKGROUND

The aim of the study was to make an inventory of trees growing in the green space of the university campus, and to assess the air quality using biomonitoring with epiphytic lichens.

Lichens are symbiotic organisms composed of a fungal partner (mycobiont) and one or more photosynthetic partners (photobiont), which is either a green alga or cyanobacterium. Some 95% of lichens are formed by fungi from the phylum Ascomycota<sup>1</sup>.

Lichens are known to be sensitive to air pollution, due to their ability to absorb chemicals from the air and rainwater<sup>2</sup>. Therefore, the diversity of epiphytic lichens is commonly used as a sensitivity indicator of the biological effects of various air pollutants, such as: sulphur dioxide, nitrogen dioxide, fluorides, ozone,

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hydrocarbons, heavy metals<sup>3</sup> and particulate matter<sup>4</sup>, in and around an urban or industrial area.

University Josip Juraj Strossmayer in Osijek was founded in 1975, and comprises 11 faculties, 5 departments and an Art Academy. The academic community consists of 17 042 students and 1151 teaching staff in 2015. University obliged to promote and implement principles and practice of environmental protection and sustainable development at all levels. In January 2015, the University Senate passed Action Plan for Environmental Protection, a guidance document with list of measures and activities which can be implemented today, or planned for the long-term period. The University campus covers area of 23.2 ha, where construction of new buildings and infrastructure for education and daily life of students, as well as urban planning started in 2002.

## EXPERIMENTAL

*Study area*. City of Osijek is the fourth largest city in Croatia, and administrative, economic and cultural centre of the Eastern Croatia. It is situated in a plain on the right bank of the Drava River, at altitude of 90 m above sea level (Fig. 1). Urban area covers 169 km<sup>2</sup>, and the population is 108 048 in 2011.

The climate is moderately warm with warm summer. Mean annual air temperature is 11°C; minimum in January (-0.2°C), maximum in July (21.3°C).

Annual rainfall is 655 mm; maximum (82 mm) in June, and minimum (35 mm) in March.

City of Osijek is abundant with urban green spaces. There are 17 parks inside the city area, with a total surface of 40 ha.

Sampling and analyses. Lichens found growing as epiphytes on trees in the University campus in Osijek was surveyed during April and May 2012. Identification of lichen specimens was made in the field with a hand lens (magnification  $10\times$ ), and in the laboratory using a dissecting microscope LEICA MZ6 (magnification  $6.3 - 40\times$ ), and comprehensive determination books<sup>5,6</sup>. Tree species according to their rank in botanical systematics were identified in the field. Data on air quality in the city of Osijek were taken from the Annual Report on Air Quality Monitoring in the Republic of Croatia for 2011 year (Ref. 7).

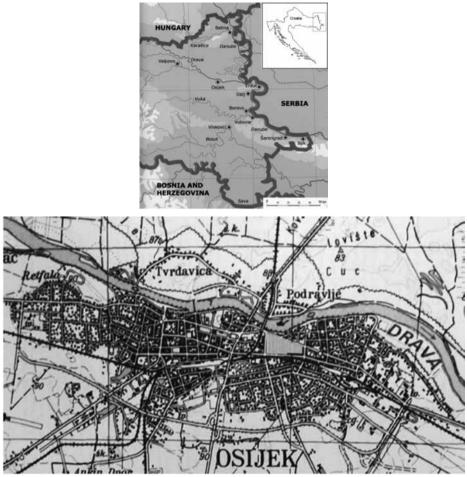


Fig. 1. Geographical location of the City of Osijek (top) and the University Campus (bottom)

# **RESULTS AND DISCUSSION**

Total of 365 trees has been recorded in the green space of the University campus in Osijek, classified into 21 species, 16 of which were deciduous trees (76%) and 5 were conifers (24%). The most frequent tree species were: London Plane tree (*Platanus x acerifolia*), Lime trees (*Tilia* spp.) and Horse Chestnut (*Aesculus hippocastanum*). Species composition, number of recorded trees and their frequency are presented in Table 1.

Scientific name	Number of trees	Frequency (%)
Platanus x acerifolia	61	16.71
<i>Tilia</i> spp.	57	15.62
Aesculus hippocastanum	50	13.70
Picea abies	32	8.77
Thuja occidentalis	30	8.22
Gleditchia triacanthos	27	7.40
Acer spp.	18	4.93
Catalpa bignonioides	18	4.93
Betula pendula	16	4.38
Chamaecyparis lawsoniana	15	4.11
Robinia pseudacacia	9	2.47
Pinus nigra	8	2.19
Populus nigra var. pyramidalis	6	1.64
Fraxinus excelsior	5	1.37
Populus nigra	3	0.82
Populus alba	2	0.55
Pinus strobus	2	0.55
Juglans regia	2	0.55
Corylus colurna	2	0.55
Quercus robur	1	0.27
<i>Koelreutreia paniculata</i>	1	0.27

Table 1. Composition of tree species recorded in the University campus in Osijek

A total of eight epiphytic lichen species, classified into six genera of lichenised fungi, were recorded on trees, as it is listed:

- Candelariella reflexa (Nyl.) Lettau
- Evernia prunastri (L.) Ach.
- Flavoparmelia caperata (L.) Hale
- Parmelia sulcata Taylor
- Physcia adscendens (Fr.) H.Olivier
- Physcia caesia (Hoffm.) Fürnr.
- Physcia tenella(Scop.) DC.
- Xanthoria parietina (L.) Th.Fr.

According to frequency of occurrence, the most frequent were *Xanthoria* parietina and *Physcia adscendens* (recorded on 72% of all trees), followed by *Parmelia sulcata* (24%), *Physcia caesia* (16%), and *Physcia tenella*, *Flavoparmelia caperata*, *Evernia prunastri*, *Candelariella reflexa*, with frequency of 4% each. Foliose lichens dominate (75%), followed by fruticose and crustose lichens with 12.5% each.

The lichens were found growing on 21 tree species. Distribution of number of lichens according to substrate is shown in Fig. 2. In the parks and tree alleys of

the City of Osijek, 16 species of epiphytic lichens were recorded<sup>8</sup>, and the most frequent trees on which lichens are growing were: lime trees (*Tilia* spp.), maple-trees (*Acer* spp.), locusttree (*Robina pseudacacia*), and horse chestnut (*Aesculus hippocastanum*).

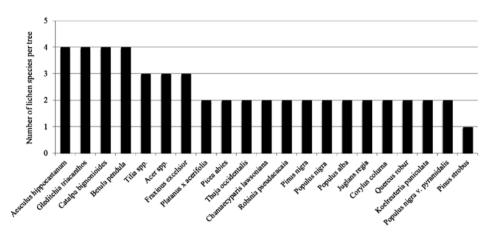


Fig. 2. Distribution of epiphytic lichen species according to host trees

Environmental conditions for lichens are optimum in mountainous, Dinaric region of Croatia<sup>9</sup>. In the area of the Risnjak National Park, 80 lichen species, including some rare old-forest indicator species, were recorded on 16 various substrates, among which dominate Sycamore Maple (*Acer pseudoplatanus*) and European Beech (*Fagus sylvatica*)<sup>10</sup>.

According to composition of the lichen flora and distribution of species, the air in the City of Osijek is estimated as moderately polluted. Noticeably lower number of lichens was observed in tree alleys along the frequent city roads. The area of the University campus is bounded by four very frequent roads, and nearby is corridor of the railway line. Due to intake of dust and increased on-road emissions of nitrogen compounds the trees are covered by nitrophilous lichens, resistent to high ereutrophication and tolerant to air pollution. Street dust contains heavy metals that can influence environment and human health. Data from the air quality measuring station in Osijek during the study period are shown in Table 2.

		Month	
Pollutant	March	April	May
Nitrogen dioxide NO <sub>2</sub> ( $\mu$ g m <sup>-3</sup> )	26.04	19.66	17.95
Sulphur dioxide SO <sub>2</sub> ( $\mu$ g m <sup>-3</sup> )	9.79	3.06	1.78
Carbon monoxide CO (mg m <sup>-3</sup> )	0.48	0.46	0.38
Particulate matter $PM_{10}(\mu g m^{-3})$	34.31	21.73	20.41

Table 2. Mean daily concentrations of air pollutants in Osijek (March, April and May 2012)

Nitrogen doixide concentration in the period March–May 2012, did not exceed limited value of 80  $\mu$ g m<sup>-3</sup> for this air pollutant, and concentrations of sulphur dioxide, carbom monoxide and particulate matter PM<sub>10</sub> were under the limited value, too. Air quality in the City of Osijek has been assessed as the first category (clean or slightly polluted air) related to NO<sub>2</sub>, SO<sub>2</sub> and CO, and as the second category (moderately polluted air) related to PM<sub>10</sub>. Measurements of air quality parameters in the City of Smederevo in Serbia indicate low concentrations of SO<sub>2</sub> and NO<sub>2</sub>, and higher concentration of particulate matter PM<sub>10</sub> in the city centre<sup>11</sup>. In the urban area of Bucharest in Romania, high concentrations of CO, SO<sub>2</sub> and NO<sub>2</sub> in the air varies proportionally with the level of noise pollution<sup>12</sup>.

## CONCLUSIONS

The composition of the epiphytic lichen flora on trees in the University campus in Osijek indicating that air in the City of Osijek is polluted mostly by emissions of dust particles and nitrogen dioxide. Occurrence of lichens contributes to the awareness of biodiversity, and planting of landscape trees and other woody plants provide new habitats for the lichens. It is also an effective measure for increasing annual rate of carbon storage and sequestering in the campus green landscape, so the University can contribute effectively to the sustainable development.

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# DETERMINATION OF TRAFFIC NOISE POLLUTION OF THE CITY OF TEKIRDAG

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**Abstract**. Noise is a phenomenon that has physiological and psychological effects on human. It is a significant environmental problem in many urban areas. There are different kinds of noise sources. Especially, traffic noise is very effective in the center of cities. This problem has not been properly recognised despite the fact that it is steadily growing in developing countries Today there are several studies to decrease the negative effects of noise on human health. Noise maps are one of these studies. These maps are a cartographic representation of the noise level distribution in a determined area and period of time. The aim of the study is to determine the traffic noise in Tekirdag and to investigate possible solutions to reduce the traffic noise. Noise levels e measured on 54 different points between mornings (8:00–9:00), noon (12:00–13:00) and evening (17:00–18:00) hours and the results will be analysed statistically and also these values will be entered and analysed via Geographical Information System (GIS).

Keywords: noise, Tekirdag, traffic noise, GIS, statistical analyses, IDW.

## AIMS AND BACKGROUND

Noise, commonly defined as an unwanted sound, is an environmental problem to which human are exposed throughout their life.

The transportation facilities that participate at the road service, the railroad traffic, and the air traffic are important sources of noise and vibrations. The noise and vibrations generated by them are extremely harmful both for travelers and drivers, and for the environment<sup>1,2</sup>. In contrast to many other environmental problems, noise pollution continues to increase due to the industrial, social and transport development. Different transportation systems modes have different scales of noise emissions. The intensity of noise is directly linked with the magnitude of traffic and travel speed. In urban areas the ambient noise is principally caused by transportation<sup>3</sup>.

According to the WHO, noise pollution is nowadays the third most hazardous environmental type of pollution, preceded only by air and water pollution<sup>4</sup>. Pollution in large cities is an overgrowing problem due to the fact that the urban

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environment is becoming increasingly crowded, busy and noisy. Since the seventies, 'noise' has been largely considered as a major problem of annoyance in cities.

Many researchers have reported that traffic is the predominant and most generalised noise source in urban areas. Exposure to traffic noise is associated with a wide range of effects on human health and well-being. Traffic noise has a variety of adverse effects on human health. Community noise, including traffic noise, is already recognised as a serious public health problem by the World Health Organisation, WHO (Ref. 5). The growth in noise pollution is unsustainable because it involves direct, as well as cumulative, adverse health effects. It also adversely affects future generations and has sociocultural, aesthetic, and economic effects.

Of all the adverse effects of traffic noise the most widespread is simply annoyance. There is also substantial evidence for traffic noise disturbing sleep patterns, affecting cognitive functioning (especially in children) and contributing to certain cardiovascular diseases. For raised blood pressure, the evidence is increasing. For mental illness, however, the evidence is still only limited. The health effects of noise are not distributed uniformly across society, with vulnerable groups like children, the elderly, the sick and the poor suffering most.

Researchers in many countries have investigated and characterised different traffic noise pollution<sup>6–9</sup>. Noise reduction is possible if adequate technology is used to minimise it. However, solving this problem requires specific infrastructure resources. To reduce noise at different environments, one should certainly use all other possible manners and tools that lead to the protection of the population health<sup>10</sup>.

Noise barriers reduce noise levels by 3 to 6 dB (A), depending on their design and height. Roadside noise barriers are only acceptable for motorways and other bypass roads where there is no need for pedestrians to cross. On busy urban streets, which are crossed by pedestrians along their entire length, noise barriers can not be placed directly on the curbside. It is only in non-urban areas that they can provide a solution, therefore<sup>5</sup>. Noise reduction capacity of planted vegetation can be used to abate noise pollution in town and landscape planning if the plantations are at least 12 m wide. To obtain the best effect the rows of trees have to be planted perpendicular to the direction of the sound field<sup>11</sup>.

For many years, it is thought that only create noise problems related to the hearing system. However, the noise of applied scientific studies has proven to be both physical and psychological effects on human health (Table 1).

Degree	Value dB (A)	Effects
1	30–65	discomfort, anger, sleep and attention disorders
2	65–90	increase of blood pressure, the acceleration of the heartbeat and respiration
3	90-120	headache
4	120-140	permanent damage to the inner ear, balance disorders
5	>140	severe brain damage

Table 1. Negative effects of noise<sup>12</sup>

## EXPERIMENTAL

*Materials*. Tekirdag is situated on the northern coast of the Sea of Marmara, 135 km west of Istanbul. The study area is located at 4,540,208.425 N, 4,533,972.195 S, 550,578.695 W, 539,676.195 E coordinates in UTM projection. Tekirdag spreads over the Ergene and Istiranca sections of the Marmara region. It is surrounded by Istanbul to the east, the Marmara Sea to the southeast, Gallipoli of Dardanelles to the south, Edirne to the west, and Kirklareli to the north. The surface area is 6218 km<sup>2</sup>. In the context of study; noise levels were measured on 54 different points (Fig. 1) which are homogeneously selected in urban protected area, during morning, noon and evening hours. Average 10–13 accounts were made at every point. TROTEC SL 300 brand device was used in measurements and made calibration with the other device of the same brand.



Fig. 1. Locations of 54 noise measuring points

*Methods*. Noise values of morning, noon and evening obtained have been assessed with 'Geostatistical Analyst' module on ArcGIS. In this module, Inverse Distance Weight (IDW) geostatistical interpolation methods were used. IDW interpolation method allows the scientist control over the degree of surface adjustment when crossing polygonal boundaries of additional locational variables. IDW algorithm effectively is a moving average interpolator that is usually applied to highly variable data. For certain data types it is possible to return to the collection site and record a new value that is statistically different from the original reading but within

the general trend for the area. Examples of this type of data include soil chemistry results, environmental monitoring data, and consumer behavior observations. It is not desirable to honor local high/low values but rather to look at a moving average of nearby data points and estimate the local trends<sup>12</sup>. In this method; the generic equation for inverse distance weighted interpolation is:

$$Z_{x,y} = \sum_{i=1}^{n} Z_i w_i / \sum_{i=1}^{n} W_i$$

where  $Z_{x,y}$ , is the point to be estimated,  $Z_i$  represents the control value for the *i*th sample point, and  $w_i$  is a weight that determines the relative importance of individual control point  $Z_i$ , in the interpolation procedure. As a binary switch,  $w_i = 1$  for the *n* control points nearest to the point being interpolated, or for the set of control points within some radius, *r*, of the point to be interpolated;  $w_i = 0$  otherwise (Fig. 2) (Ref. 13).

## **RESULTS AND DISCUSSION**

Using the technique IDW, level of morning (8:00–9:00), noon (12:00–13:00) and evening (17:00–18:00) noise maps have been created. These maps have been used when the values are average values. Noise maps modelled by the IDW method have been shown as an example in Figs 2–4.

Noise level is determined as between 53–76 dB(A) according to measurements that are made during morning in area. High noise levels are measured at points 7, 12, 14, 35 and 39. Point 7 is located at the crossroads it is well located at intercity bus terminal. Point 12 is located at the local minibus in the city center. Other measured points are located in the city center. Low noise levels are measured at points 28, 52 and 44 that are in inner sides of districts and far from the main road (Fig. 2).



Fig. 2. Morning noise map modelled by IDW method

Noise level is determined as between 52–72 dB(A) according to measurements that are made during noon in the study area. The highest noise level is measured at points 12 and 34. Point 12 is located at the local minibus in the city center. Point 34 is located at Cengiz Topel Square and in front of a private hospital. Low noise levels are measured at points 27, 28 and 29 that are far from the main road and the city centre (Fig. 3).

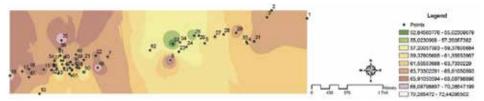


Fig. 3. Noon noise map modelled by IDW method

Noise level is determined as between 54–75 dB(A) according to measurements that are made during evening in the study area. High noise levels are measured at points 3, 31, 7 and 22 is located above the main road and intercity bus terminal. Low noise levels are measured at points 27, 28 and 29 that are far from the main road and city center. Low noise levels are measured at points 24, 31 and 43 that are located at the streets and also are far from the main road (Fig. 4).



Fig. 4. Evening noise map modelled by IDW method

This study was carried out to evaluate the environmental noise pollution of Tekirdag due to the traffic noise, to investigate traffic noise levels using the IDW method and to assess and rate noise exposure in the region. When the measured noise level values are examined, it is seen that the noise level is around morning (53-76) dB(A), noon (52-72) dB(A) and evening (54-75) dB(A). This value shows us the noise pollution in Tekirdag city. In particular, it is seen that these values are very high at some points. The value of the points increased in the main roadsides, intersections, squares, inter-city bus terminals and minibus stops. In other points, the noise levels were less measured. In the study, noise level values were measured for morning, noon and evening and analysed statistically. QQPlot graphics were generated in accordance with these results. Normal OOPlot graphic is relatively obtained with mutual plotting of data of cumulative normal distribution and data of cumulative. Normal QQPlot graphic enables to compare our data with standard normal distribution. As points are near straight line, the distribution of data is also accepted to near-normal distribution. Many of these data are suitable to the normal distribution as shown in Figs 5, 6 and 7.

QQPlot graphic of morning: it is observed as the proximate distribution of data (Fig. 5).

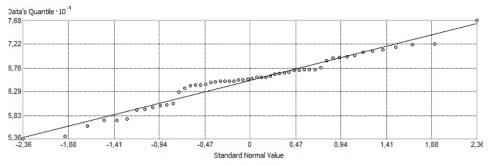


Fig. 5. QQPlot graphic of morning

QQPlot graphic of noon: the data distribution has been observed as a normal distribution (Fig. 6).

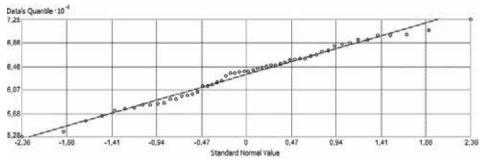


Fig. 6. QQPlot graphic of noon

QQPlot graphic of evening: the data distribution has been observed as a normal distribution (Fig. 7).

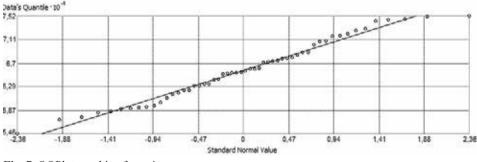


Fig. 7. QQPlot graphic of evening

Histogram tool plots frequency histograms enable us to examine the data distribution for each quality. Data are divided into 10 classes as shown in Figs 8,

9 and 10. Density of data for each class is determined with the height of suitable class. Generally, foremost properties of data distribution are central value, spread and balancing of data. If mean and median values are about the same as quick evaluation, it could be said that the data distribution is normal. Accordingly, histogram shows that the data distribution of morning measurements is not symmetrical but it is bumpy. Due to the fact that values of mean; 65.062 and median; 65.34 are not about the same, the data distribution is normal (Fig. 8).

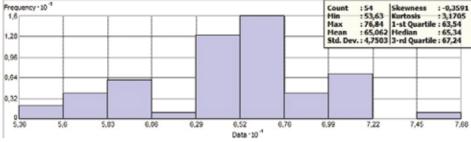


Fig. 8. Histogram of data distribution for noise pollution of morning

Data distribution of morning measurements are symmetrical. Due to the fact that values of mean; 63.26 and median; 63.835 are about the same, the data distribution is normal (Fig. 9).

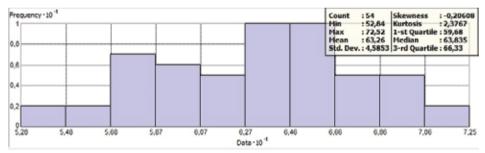


Fig. 9. Histogram of data distribution for noise pollution of noon

Data distributions of morning measurements are symmetrical. Due to the fact that values of mean; 65.59 and median; 65.525 are about the same, the data distribution is normal (Fig. 10).

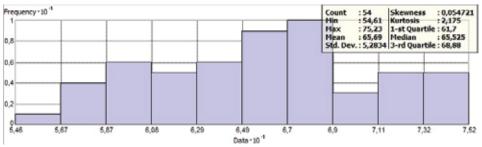


Fig. 10. Histogram of data distribution for noise pollution of evening

# CONCLUSIONS

Noise pollution is a significant environmental problem in many urban areas. This problem has not been properly recognised despite the fact that it is steadily growing in developing countries<sup>14–16</sup>. Tekirdag is one of the fastest growing major city in Turkey. It experienced rapid urbanisation due to the population growth and the increase in vehicle density. Noise, the negative one of these effects on people in Tekirdag province, is considered to be much more real in the coming years. For this purpose it is necessary to establish urban plans related to the transportation plan in order to reduce the noise effect.

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Ecology

# **BIOMONITORING OF HEAVY METAL POLLUTION ON THE LEAVES OF Cupressus arizona AND Albizia julibrissin AND THEIR CONTAMINATION SOURCES IN THESSALONIKI CITY (GREECE)**

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**Abstract**. The research reveals results of heavy metal pollution on leaves of two plant species on the roadside of Thessaloniki city, Greece. Concentrations of copper (Cu), lead (Pb), chromium (Cr), nickel (Ni), cadmium (Cd), manganese (Mn) and zinc (Zn) were determined in washed-unwashed leaves and upper-lower part of tree crown. Acid digestion of leaves was used. Heavy metals concentration was measured by Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES). According to the research results, heavy metals concentration depends on the forest species and the part of tree crown that the leaves where collected. Statistically important differences between the studied species were found for all metals. In addition, not even a single species was found to retain the biggest concentration of all. Correlation analysis shows positive correlations between metals demonstrate that the common origin of above metals associated with traffic.

Keywords: needles, leaves, atmospheric pollutants, pollution sources.

# AIMS AND BACKGROUND

Rapid industrialisation and the uncontrolled development of large cities have seriously contributed to the contamination of air, water and soil. Air pollution has been recognised as a health hazard, since the early decades of the last century, when severe air pollution episodes followed industrialisation in Europe and USA. International standards on air pollution quality have been based mostly on studies that followed these air pollution episodes<sup>1</sup>. Vehicular traffic is widely recognised

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to be a significant and increasing source of atmospheric pollution in urban environments<sup>2-5</sup>. The temporal evolution of the air pollution sources in Greek urban areas is characterised by the continuous increase of the vehicles fleet and mainly the new technology passenger cars equipped with tree-way catalytic converters<sup>6</sup>. Plant biomonitoring is being increasingly used, especially in Europe, as an alternative to the traditional (instrumental) methods of studying regional deposition of natural and anthropogenic substances from the atmosphere to the terrestrial environment<sup>7–10</sup>. Botanical materials such as fungi, lichens, tree bark, tree rings and leaves of higher plants, have been used to detect the deposition, accumulation and distribution of heavy metal pollution<sup>11,12</sup>.

The trees act effectively decreasing the pollutants that are found as gases via their detention mainly by the leaves. Leaves have different forms depending on species<sup>13,14</sup>.

The aim of present study is to look into the pollution levels of copper (Cu), lead (Pb), chromium (Cr), nickel (Ni), cadmium (Cd), manganese (Mn) and zinc (Zn) using *Cupressus semprevirens* and *Albizia julibrissin* leaves in Thessaloniki.

*Cupressus arizonica*, and *Albizia julibrissin* were selected because they are growing widely in both urban and rural areas; and they have a widely geographical range and ecological distribution throughout the world; and also their sampling, identification and cultivation is easy and inexpensive. Also species selection (broadleaves and coniferous, deciduous and evergreen) was made on the basis of different leaves morphology (needle-lamina, simply-compound) and their different silvicultural treatment (*Albizia julibrissin* was being trimmed).

#### EXPERIMENTAL

The city of Thessaloniki, located in the Northern Greece, is the second bigger city in Greece. It is rapidly growing city and its present population is estimated to be above 1 000 000 citizens. The city has Mediterranean climate with dry hot summers and mild winters. The mean annual air temperature is 15.8°C with minimum mean monthly temperature 5.9°C (January) and maximum 25.9°C (July). The mean annual precipitation is 449.3 mm and relative humidity is 66.7% (Refs 13 and 14).

The research was carried out in street trees of one of the main, with heavy traffic, street of municipality according to data of Central Macedonia Prefecture of Thessaloniki that is the K. Karamanlis Avenue (Fig. 1, Table 1). The street trees of the avenue formed mainly with *Albizia julibrissin* and *Cupressus arizonica* (Table 2).



Fig. 1. Part of the studied Karamanlis Avenue (www.google.com).

	Min. (vehicles/h)	Max. (vehicles/h)	Mean (vehicles/h)	Standard deviation
Traffic flow	48137.33	64501.00	56161.27	5190.89

Table 1	Mean	statistics	of traffic	of The	ssalonica	city
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Source: Region Central Macedonia, Adm. Public Work.

Table 2. Number of trees of each forest species of K. Karamanlis Avenue stree	t trees
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Street	Forest species	Number of trees
Karamanlis Avenue	Albizia julibrissin	185
	Cupressus arizonica Green	99
	Liquidambar orientalis Mill.	60
	Populus X euramericana cv. 'I-45/51'	141

Source: Batala, 2006.

K. Karamanlis Avenue is located next to one of the biggest hospitals in town (Ippokratio).

Leaf samples of urban roadside were collected nearby the avenue. Samples were collected during August<sup>15–17</sup>. From each species twenty trees were taken<sup>18,19</sup>. From each tree two samples were collected, one at the 1/3 and the other at the 2/3 of tree crown height.

The leaf samples were then divided into two sub-samples. One sub-sample was thoroughly washed with running distilled water to remove dust particles, and the other remained untreated. All leaf samples were oven-dried at 80°C for 24 h, milled in a micro-hammer cutter and fed through a 0.2-mm sieve. The leaf samples were stored in clean plastic vases. Contamination from the micro-hammer cutter was negligible while grinding, since it was washed with distilled water after each use.

First there was wet digestion of organic samples by the use of concentrated  $HNO_3$ . About, 0.2 g of ground dried plant sample were put in porcelain capsule. After it was heated at 600°C for 5 h, the capsule was removed from the oven and was released reintroducing the sample to environmental temperature. Ten ml of concentrated  $HNO_3$  (2N) were added, and the sample was heated at 80°C on a glow plate in abductor hearth. The solution was cooled and was filtered through Whatman No 42 with diameter 125 mm. The filtrate was diluted to 50 ml of distilled water. Each final solution was analysed for Cu, Pb, Cr, Ni, Cd, Mn and Zn.

Three standards (Culture Labatory Proficiency-ALP program operated by Collaborative Testing Services Inc.) were also analysed. The three standard reference botanical materials were: corn stalk from Connecticut, potato leaves from California and citrus leaves from California. Detection of the metals was carried out with inductively coupled plasma optical emission spectrometry ICP-OES (PS-1000 AT-Seqential Axial Transmittion Model with Autosampler of Leeman Labs). The absorption measurements of the metals were performed under the conditions recommended by the manufacturer.

The values were statistically evaluated with tree-way analyses of variance (ANOVA), in the methodological frame of General Linear Models. Specifically, the data were assessed using linear model that it includes a factor between the experimental units (factor 'Species' with 2 levels: *Cupressus arizonica* and *Albizia julibrissin* and two factors inside the experimental units (factor 'Height' with 2 levels: upper and under, factor 'Treatment' with 2 levels: unwashed and washed). The experimental plot was considered as completely randomised factorial.

The total number of measurements of heavy metals concentrations for each species was 80. The comparisons of means were realised by the Bonferroni multiple range test, for a p = 0.05 confidence level<sup>20</sup>. The cross-correlations between quantitative parameters were evaluated with base the factor of linear cross-correlation of Pearson. The statistical analysis was conducted with the statistical software package of SPSS v. 15.0 (SPSS Inc., Chicago, IL). The level of importance of all statistical controls was predetermined in p = 0.05.

#### **RESULTS AND DISCUSSION**

Forest species are very often used in researches of atmospheric pollution in particularly polluted regions where the lichens and the moss are often absentee<sup>21</sup> but also in urban environments<sup>17</sup>. Both coniferous and broad-leaved tree leaves are used in studies for monitoring metal pollution in urban environment<sup>17,22,23</sup>.

At Napoli were studied copper, lead and iron concentration on *Quercus ilex* leaves surface<sup>24</sup>. Metal concentrations were higher on leaves that were collected at the edge of the road than those that were collected from city blocks and leaves had higher concentrations than those that were collected from park trees. The significant higher concentrations of Cu than Pb and Fe confirm that traffic is the major air pollution source inside cities<sup>25</sup>.

The concentrations of Mn, Zn, Cu, Pb, Cd, Cr and Ni on tree leaves in different tree height and different treatments are given in Fig. 2.

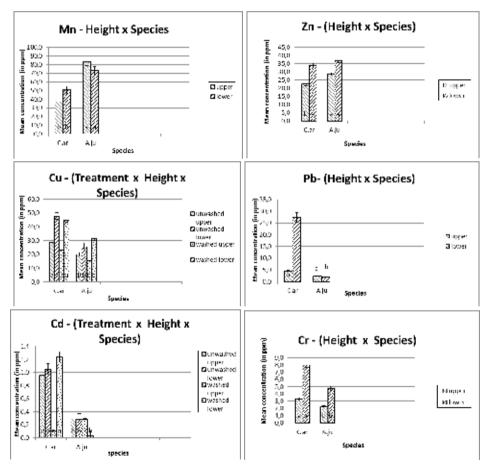


Fig. 2. Mean concentrations of metals in ppm per treatment, height and species, together with standard error (SE) bars (for each species, means that are followed by different letter differ statistically considerably (p < 0.05) (the Bonferroni test))

For all metals that have been studied, there were found statistically important differences among the forest species. A tree species containing the highest concentrations of all metals measured was not observed. Concentrations of copper, lead, chromium, nickel and cadmium were bigger at *Cupressus arizonica*. The other two metals, Mn and Zn were found to have bigger concentration at *Albizia julibrissin*.

Other researchers found that Pb concentration that was measured in the samples of *Nerium oleander* leaves had no difference between the samples that had been washed or no, before the analyses<sup>26</sup>. The present research confirms the above conclusion. There was no statistically important difference of Pb concentration on the leaves that had not been washed with those that had been washed for both species.

Also, for all metals that were studied in the present work, the factor `height' was statistically important (p < 0.001) something that shows the importance of above factor in the measurement of concentration of metals at trees. For *Albizia julibrissin*, bigger concentrations were measured at the upper parts of crowns. In the opposite, for *Albizia julibrissin*, bigger concentrations that were measured at the lower parts of crowns. The bigger concentrations that were measured were Cu – 47.810 ppm (not washed leaves), Pb – 27.470 ppm, Cd – 1.237 ppm (washed leaves), Cr – 7.958 ppm and Ni – 3.296 ppm (washed leaves).

*Cupressus arizonica* retain its needles from two to five years. *Cupressus arizonica* needles are microscopic, abundant, blade shape, aquamarine, with obvious gland in ridge and with white resin<sup>27</sup>. Researchers support that the rough surface of needles and the presence of resin makes surface adhesive and contribute to the increase of heavy metals concentration<sup>28</sup>.

On the leaves of the upper crown part of *Albizia julibrissin* was measured the bigger concentration of manganese and zinc. *Albizia julibrissin* was the only species that was pruned systematically every year. The silvicultural parameters or treatments as crown surface or pruning are crucial parameters for heavy metal concentration on leaves.

According to researchers, exterior leaves of the tree crown have increased faculty to engage elements from atmosphere. Consequently is required further research for the way that pruning influence heavy metals concentration on leaves<sup>22</sup>. Researchers at Thessaloniki<sup>17</sup> found for *Robinia pseudoacacia* as well as for *Albizia julibrissin* that the presence of aphids makes leaves surface more adhesive having as a result the easier detention of heavy metals.

Inter-element relationships provide interesting information on heavy metal sources and pathways. Correlation analysis shows that metals have common origin associated with traffic. Correlation coefficients were higher in Cupressus needles than in Albizia leaves (Tables 3 and 4).

11						
	Zn	Cu	Pb	Cd	Cr	Ni
Mn	0.177	0.460*	0.005	-0.248	0.301	0.163
Zn	1	0.619**	0.708***	0.444*	0.629**	0.882***
Cu	0.619**	1	0.573**	-0.125	0.761***	0.569**
Pb	0.708**	0.573**	1	0.078	0.668**	0.657**
Cd	0.444*	-0.125	0.078	1	-0.144	0.384
Cr	0.629**	0.761***	0.668**	-0.144	1	0.623**

 Table 3. Correlation matrix of the total heavy metals concentration on unwashed Cupressus needles at the upper crown

\*\*\* Indicates significant correlation at a significance level p < 0.001; \*\* indicates significant correlation at a significance level p < 0.01; \* indicates significant correlation at a significance level p < 0.05.

 Table 4. Correlation matrix of the total heavy metals concentration on unwashed Albizia leaves at the upper crown

	Zn	Cu	Pb	Cd	Cr	Ni
Mn	-0.051	-0.289	0.116	0.565**	-0.186	0.159
Zn	1	0.203	-0.236	-0.048	0.164	-0.044
Cu	0.203	1	-0.110	-0.328	0.610**	-0.033
Pb	-0.236	-0.110	1	0.001	0.021	0.305
Cd	-0.048	-0.328	0.001	1	-0.407	0.445*
Cr	0.164	0.610**	0.021	-0.407	1	0.367

\*\*\* Indicates significant correlation at a significance level p < 0.001; \*\* indicates significant correlation at a significance level p < 0.01; \* indicates significant correlation at a significance level p < 0.05.

Moreover, the species *Cupressus arizonica*, except strongly positive correlation between zinc and copper, was found to have high tension positive correlation between zinc to chromium and nickel. Also in this type found very strong degree of positive correlation between the lead and chromium.

For the species *Cupressus arizonica*, correlation analysis (Table 3) shows that Zn and Cu, Zn and Cr and Ni and finally Pb and Cr were positively correlated suggesting that these elements came from same sources. Also for species *Albizia julibrissin* (Table 4), it was found that there is a positive correlation between Mn and Cd. The positive correlations of above metals demonstrate that the common origin of above metals associated with traffic<sup>29,30</sup>. Also the positive correlation between Zn and Cu demonstrates the relationship between the two metals as the main metal pollutants from cars breaks<sup>31</sup>.

#### CONCLUSIONS

The species *Cupressus arizonica* withhold the largest concentrations of copper (Cu), lead (Pb), cadmium (Cd) chromium (Cr) and nickel (Ni) and the species *Albizia jullibrissin* the higher concentrations of manganese (Mn) and zinc (Zn).

The effect of the height to the retention of heavy metals from the leaves of forestry species was found statistically significant. The size factor is significant in the two layers (top and bottom of the crown).

In choosing forestry species suitable for urban use, depending on their behaviour for heavy metals should be taken in account not only the retention percentages of heavy metals of all kinds and the crown surface and the durability of their leaves (deciduous and evergreen).

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# PHYTOCOENOSES WITH *Convolvulus persicus* L. ON THE WESTERN COAST OF THE BLACK SEA

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**Abstract**. Field surveys carried out in Romania and Bulgaria between the years 2012 and 2015 led to the identification of phytocoenoses with *Convolvulus persicus* in some areas of the Danube Delta Biosphere Reserve (Sulina, Sf. Gheorghe, Cardon), in the natural reserve Marine Sand Dunes of Agigea and on Durankulak beach, in Northern Bulgaria. Information regarding floristic composition, ecology and the syntaxonomy of the plant community *Convolvuleum persici* (Borza 1931) Sandra et al. 1998 were presented in the paper. Considerations upon the conservation status and the risk factors which threaten local populations of *Convolvulus persicus* on the western Black Sea coast have been given. Threatened species recorded in the plant association *Convolvuleum persici* in different locations in Romania and Bulgaria, as well as the conservation importance of the plant community, were highlighted in the article.

Keywords: Convolvulus persicus, phytocoenoses, sand dunes, western Black Sea coast.

# AIMS AND BACKGROUND

*Convolvulus persicus* (sand morning glory) is a psammophilic species native on the Caspian Sea littoral, reported in Europe only in Romania, Bulgaria and Turkey<sup>1</sup>. The general distribution of these taxa includes different locations around the Black Sea and on the Caspian Sea<sup>2</sup>: South-East Romania, Eastern Bulgaria, European Turkey, North-West Anatolia, Georgia, Russia (Daghestan), Azerbaijan, Iran and Turkmenistan. Because of its rarity, of the small local populations and of its high vulnerability to the anthropogenic impact, *Convolvulus persicus* is considered a critically endangered species both in 'The Red Book of the Vascular Plants of Romania'<sup>3</sup> and in 'The Red Data Book of the Republic of Bulgaria'<sup>4</sup>.

In Romania, *Convolvulus persicus* has been noticed on the sand dunes in some locations within the Danube Delta Biosphere Reserve – Sulina, Sfantu Gheorghe, Letea, Caraorman<sup>3</sup>, Cardon, C. A. Rosetti, Sfistofca<sup>5</sup>, Sacalin<sup>6</sup>, Perisor<sup>7</sup> and in the natural reserve Marine Sand Dunes of Agigea (Fig. 1).

<sup>\*</sup> For correspondence.

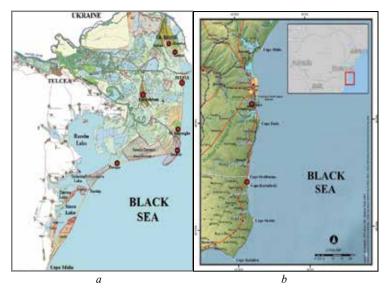


Fig. 1. Distribution area of *Convolvulus persicus* in Romania and Bulgaria: a – the Danube Delta Biosphere Reserve; b – Southern Romanian coast and Northern Bulgarian coast

In Bulgaria, *Convolvulus persicus* was recorded in the last 20 years only on the sandy beach in Durankulak Lake area<sup>4</sup>. Other locations of this taxon in Bulgaria (near the Rezovska river mouth, Silistar bay, and Kamchia river estuary) specified in some old papers<sup>4</sup> were not confirmed after 1995. On the Caspian Sea coast, in the North of Iran, the plant community *Convolvuletum persici* is mentioned in the Miankaleh Biosphere Reserve<sup>8</sup> and in the Boujagh National Park<sup>9</sup>.

In accordance with the bibliographical sources<sup>10</sup>, the plant association *Convolvuletum persici* belongs to the order *Cakiletalia maritimae* R. Tüxen Oberd. 1949 beside other plant communities of the habitat 1210 (Annual vegetation of drift lines).

## EXPERIMENTAL

Observations concerning the phytocoenoses with *Convolvulus persicus* were carried out between the years 2012 and 2015 on the Black Sea coast of Romania and Bulgaria. Field observations and phytosociological releves were achieved according to the methodology of Braun-Blanquet phytosociological school. The syntaxonomic affiliation of this plant community is according to the book 'Phytocoenoses from Romania'<sup>10</sup>. The nomenclature of the species from the floristic composition of the association is in accordance with 'Flora Europaea<sup>1</sup> and 'Vascular plants of Romania'<sup>11</sup>. The conservation status of the species *Convolvulus persicus* was assessed based on a 3-level scale as follows: favourable, unfavourable-inadequate and unfavourable totally inadequate<sup>12</sup>.

## **RESULTS AND DISCUSSION**

Phytocoenoses with *Convolvulus persicus* have a fragmented range along the Black Sea shore and on the marine sandbanks within the Danube Delta Biosphere Reserve. In many locations of the Danube Delta (Letea, Caraorman, C. A. Rosetti, Şfistofca, Perisor),*Convolvulus persicus* has only small local populations. Typical phytocoenoses with *Convolvulus persicus* have been noticed on the sandy beaches of Sulina, Sfantu Gheorghe and Cardon<sup>13</sup>, in Sacalin-Zatoane strictly protected area<sup>6</sup>, in the natural reserve of Agigea and on Durankulak beach, in northern Bulgaria (Fig. 2). On the wide beaches in Sulina and Sfantu Gheorghe, such types of phytocoenoses occur on the mobile and semi-fixed sand dunes, approximately 30–40 m from the shoreline, where the sea waters reach only during storms.



Fig. 2. Phytocoenoses with Convolvulus persicus on the beaches of Sulina (a) and Durankulak (b)

*Convolvulus persicus* reaches in Sulina a good density of 17–20 individuals/m<sup>2</sup>. The flowering period of *Convolvulus persicus* is in June-July. Only approximately 40–50% of individuals have bloomed in the investigated period, the vegetative reproduction being prevalent on the beach of Sulina. The strong attack of the parasitic plant *Cuscuta* sp. could be the reason for the low flowering capacity of the sand morning glory in Sulina.

The flowering rate of *Convolvulus persicus* is higher (approximately 60–70%) on the Sfantu Gheorghe beach than in Sulina and this fact can be linked to the lower density of the local populations (10–12 individuals/m<sup>2</sup>), to the favourable ecological conditions and to a low anthropogenic pressure upon the dune habitats.

In the coastal area of the Danube Delta, phytocoenoses with *Convolvulus persicus* are vulnerable to both anthropogenic and natural pressures such as grazing, tourism, the expansion of recreational areas on the beaches<sup>14</sup>, storms<sup>15</sup>. The conservation status of this species can be considered favourable in Sfantu Gheorghe and unfavourable-inadequate on the beach of Sulina where the human impact upon the dune habitats is higher than in Sfantu Gheorghe. In Sulina, one of the most important touristic gates of the Danube Delta, tourism is better developed than in Sfantu Gheorghe village and this fact has some negative consequences on

the natural vegetation of the beaches. Approximately 3 ha of the beach in Sulina were transformed in 2009 by the local authorities in recreational area. Because of this situation, Romania was subject to the 'infringement procedure' due to the violation of the European legislation regarding the conservation of the habitats from the Council Directive 92/43/EEC.

On the southern coast of Romania, phytocoenoses with *Convolvulus persicus* have been noticed only in the natural reserve Marine Sand Dunes of Agigea, on the mobile sand dunes from the north-western side of the protected area. Currently, there are different harbour facilities between the protected area and the seashore which diminished the positive influences of the sea breezes upon the dune habitats and, consequently, determined significant microclimate changes in the area of the natural reserve. The steppe grasslands and the disturbed habitats from the vicinity of the protected area have facilitated the infiltration of some steppe and ruderal plants, even of some invasive species in the sand dunes complex. *Convolvulus persicus* has at present a favourable conservation status only due to some effective management measures of the dune habitats. The measures consist mainly in removing the invasive species and of some opportunistic steppe plants from the dunes area.

In Bulgaria, only two phytocoenoses with *Convolvulus persicus* have been noticed on Durankulak beach (5 km south from the border with Romania) and these are the only records on the Bulgarian coast. *Convolvulus persicus* occupies here approximately 400 m<sup>2</sup> on the first strips of the shifting sand dunes. The flowering rate of this species is very good on Durankulak beach because more than 70% of the total specimens have bloomed in the period of survey. *Convolvulus persicus* has generally a favourable conservation status in Bulgaria, the risk factors being mainly natural (storms with big waves, strong winds).

The total number of the plant species from the floristic composition of the association is small (45 taxa) due to the restrictive ecological conditions within the dune habitats area. The majority of plant species are psammophilic (57.79%) but steppe species are also numerous (33.33%) (Fig. 3). Most of accompanying species belong to the alliances *Scabiosion ucrainicae* Boscaiu 1975, *Festucion vaginatae* Soó 1929 and *Elymion gigantei* Morariu 1957. *Convolvulus persicus* is the diagnostic taxon of the association *Convolvuletum persici* and it has high values of the abundance-dominance index AD = 3-4 and a high constancy compared the accompanying species of the plant community (Table 1).

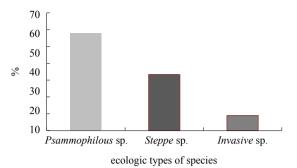


Fig. 3. Comparative values of psammophilic, steppe and invasive species in the plant association *Convolvuletum persici* 

The species characteristic to the alliance *Cakilion maritimae* are well represented only on the beach of Durankulak because here *Convolvulus persicus* reaches up to 10–15 m from the seashore, in the area of the habitat 1210. In the Natural Reserve of Agigea, due to the drier microclimate, the typical species of the alliances *Scabiosion ucrainicae* and *Festucion vaginatae* have a good occurrence and high value of the abundance-dominance index in the releves of the association *Convolvuletum persici* (Table 1).

In the phytocoenoses with *Convolvulus persicus* from the Danube Delta, the species belonging to the class *Ammophiletea* Br.-Bl. et R. Tuxen 1943 are better represented than in Durankulak and Agigea. Here, *Convolvulus persicus* occupies the basis of the sand dunes with *Leymus racemosus* subsp. *sabulosus, Eryngium maritimum* and *Elymus farctus* subsp. *bessarabicus*.

The invasive and potentially invasive plants (*Amorpha fruticosa, Conyza canadensis*), with the exception of *Xanthium italicum* have a low occurrence in the releves of the association *Convolvuletum persici* (Table 1). *Xanthium italicum*, an invasive species in the sand dunes area, represents one of the threats for the population of *Convolvulus persicus* in the long term. Parasitic species from the genus *Cuscuta*are another major threat for the populations of *Convolvulus persicus*, mainly on the beach of Sulina.

Table 1. Association table of Convolvuletum persici (Borza 1931) Sanda et al., 1998	versici	(Boi	za 19	31) Sa	nda e	t al., 1	998											
Relevé number	R1	R2	R3	$\mathbb{R}4$	R5	R6	R7	R8	R9 I	R10 R11		R12 R	R13 R	R14 R1	5 R1	R15 R16 R17	7 R18	×
1	7	ς	4	S	9	7	8	6	10	11	12	13	14 1	15 16	5 17	7 18	19	20
Area (m <sup>2</sup> )	50	50	50	50	50	50	50	50	50	50	50 5	50 5	50 5	50 50	) 50	) 50	50	
Cover (%)	70	80	80	70	50	50	70	70	60	70	809	80 6	60 6	60 60	) 80	02 (	80	
Number of taxa	12	13	12	14	13	15	13	13	12	12	10	11	16 1	13 11	1 10	) 16	11	
Characteristic taxa																		
Convolvulus persicus	Э	4	4	4	ε	Э	ŝ	4	Э	e	Э	4	ŝ	33	4	ŝ	4	>
Festucetalia vaginatae, Scabiosion ucranicae	icae																	
Centaurea arenaria subsp. borys- thenica	+	+	+	I	+	+	+	+	+	+	I	+	+	1	+	+	I	$\geq$
Alyssum hirsutum	+	+	+	I	Ι	Ι	Ι	Ι	Ι	Ι	+			_	+	-	+	Ш
Cynanchum acutum	Ι	Ι	Ι	Ι	+	+	+	Ι	Ι	Ι	+		'	+		+	Ι	Π
Alyssum borzaeanum	I	Ι	I	I	I	I	I	I	I	I	Ì			-	1	0	1	Π
Silene thymifolia	Ι	Ι	+	I	Ι	Ι	Ι	Ι	Ι	I	I				+	1	+	Ι
Carex colchica	Ι	Ι	Ι	Ι	Ι	Ι	Ι	Ι	Ι	Ι	I	1	'	1		I	+	Ι
Festucion vaginatae + Bassio laniflorae -	- Bromion		tectorum	um														
Secale sylvestre	0	ы	0	+	1	+	3	+	1	+	+	1	_	2	0	+	+	>
Bromus tectorum	+	+	+	Ι	1	+	+	+	+	Ι	I	I	_	+	-	1	1	N
Euphorbia segueriana	Ι	+	I	+	I	+	7	I	1	I	· I		+	1	+	+	Ι	Ш
Astragalus varius	I	-	I	+	I	I	I	Ι	Ι	I	· I			+	+	+	+	Π
Silene conica	+	+	+	Ι	Ι	Ι	Ι	Ι	Ι	I			+	+	1	+	Ι	Π
Plantago arenaria	Ι	I	I	I	I	I	I	I	Ι	+	· I	'	+	1	1	Ι	+	Ι
Corispermum nitidum	I	I	I	I	Ι	I	Ι	Ι	Ι	-	+	+		1	1	Ι	Ι	Ι
Apera spica-venti subsp. maritima	Ι	Ι	Ι	Ι	Ι	Ι	Ι	Ι	Ι	I	Ι	Ι	-		1	1	Ι	Ι
																to	be coi	to be continued

rg 1031) Sanda at al 1008 meini (Bo ŝ Table 1 Association table of Convolviuletium

Continuation of Table 1 1	10	m	4	S	9	~	~	6	10	=	12	13	14	15	16	17	18	19	20
Elymetalia arenariae, Elymion gigantei																			
Eryngium maritimum	+	+	Ι	+	+	+	+	Ι	-	2	-	+	+	Ι	I	I	I	Ι	$\sim$
Leymus racemosus subsp. sabulosus	Ι	+	I	-	+	+	+		Ι	-	+	-	+	Ι	Ι	I	I	I	III
Elymus farctus subsp. bessarabicus	-	Ι	I	+	I	I	I	I	I	I	I	I	+	Ι	Ι	I	I	I	Ι
Ammophila arenaria subsp. arundi- nacea	I	Ι	+	+		I	I	I	I	I	I	I	I	I	I	I	I	Ι	Ι
<i>Polygonum oxyspermum</i> subsp. <i>raii</i> Cakiletalia maritimae	+	Ι	+	I	I	I	I	I	I	Ι	Ι	I	I	I	I	I	Ι	Ι	Ι
Salsola kali subso ruthenica	+	I	I	+	+	+	I	I	+	+	I	+	I	I	I	I	I	I	Ш
Cakile maritima subsp. euxina	-	-	+	I	I	I	Ι	Ι	1	1	I	1	I	I	T	I	I	I	I I
Crambe maritima	1	Ι	+	Ι	Ι	Ι	Ι	Ι	Ι	Ι	Ι	Ι	Ι	Ι	Ι	Ι	Ι	Ι	Ι
Lactuca tatarica	I	I	Ι	+	Ι	Ι	Ι	Ι	I	Ι	Ι	Ι	Ι	Ι	Ι	I	Ι	I	Ι
Scolymus hispanicus	I	T	I	I	I	I	I	I	I	I	I	I	+	I	I	I	I	I	Ι
Festucetalia valesiacae + Festucion rupicolae	olae																		
Medicago falcata	Ι	Ι	Ι	+	Ι	+	Ι	Ι	I	Ι	+	Ι	Ι	+	-	+	+	+	Ш
Linaria genistifolia subsp. euxina	Ι	+	+	+	Ι	+	Ι	+	Ι	Ι	Ι	Ι	Ι	+	Ι	Ι	+	Ι	II
Linum austriacum	Ι	Ι	Ι	Ι	Ι	+	Ι	+	+	Ι	Ι	Ι	+	+	Ι	Ι	Ι	Ι	Π
Crepis foetida subsp. rhoeadifolia	Ι	Ι	Ι	+	Ι	+	Ι	+	Ι	Ι	Ι	Ι	+	+	Ι	Ι	Ι	Ι	Π
Seseli tortuosum	Ι	Ι	Ι	Ι	Ι	Ι	I	+	I	Ι	I	Ι	I	I	Ι	I	+	+	I
Cerastium brachypetalum	I	Ι	Ι	Ι	I	I	I	Ι	Ι	I	I	Ι	I	Ι	+	Ι	+	Ι	I
Cynodon dactylon	Ι	I	Ι	Ι	I	I	+	Ι	+	Ι	I	-	Ι	+	Ι	I	Ι	Ι	I
Verbascum banaticum	I	Ι	Ι	Ι	I	I	I	+	+	I	I	Ι	I	Ι	Ι	Ι	Ι	Ι	I
Cichorium intybus	I	Ι	I	Ι	I	I	+	I	I	I	I	I	I	Ι	I	I	I	I	Ι
																	to be	to be continued	nued

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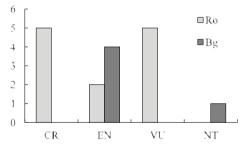
Continuation of Table 1																			
1	7	ω	4	5	9	7	8	6	10	10 11	12	13	14	15	15 16	17	18	19	20
Other species																			
Xanthium italicum	Ι	+	Ι	+	+	0	+	Ι	-	1	+	+	I	+	I	I	I	Ι	Ш
Hippophae rhamnoides	Ι	Ι	Ι	Ι	-	Ι	-	Ι	Ι	Ι	Ι	+	1	Ι	Ι	Ι	Ι	Ι	Π
Elaeagnus angustifolia	Ι	Ι	Ι	Ι	+	Ι	+	Ι	Ι	+	Ι	Ι	+	Ι	Ι	Ι	Ι	Ι	Π
Tamarix ramosissima	Ι	Ι	Ι	Ι	+	Ι	Ι	Ι	Ι	+	Ι	Ι	+	Ι	Ι	Ι	Ι	Ι	Γ
Senecio vernalis	Ι	Ι	Ι	Ι	+	Ι	I	Ι	Ι	Ι	Ι	Ι	I	Ι	+	Ι	+	Ι	I
<i>Cuscuta</i> sp.	I	Ι	Ι	Ι	I	Ι	Ι	Ι	Ι	+	+	1	Ι	Ι	Ι	Ι	Ι	Ι	Ι
Conyza canadensis	Ι	Ι	Ι	Ι	Ι	Ι	Ι	Ι	+	Ι	Ι	Ι	+	Ι	Ι	Ι	Ι	Ι	Ι
Petasites spurius	Ι	Ι	Ι	I	Ι	+	Ι	+	Ι	Ι	I	Ι	I	Ι	Ι	I	I	Ι	I
Amorpha fruticosa	Ι	Ι	I	Ι	Ι	Ι	Ι	Ι	Ι	Ι	Ι	Ι	+	Ι	Ι	Ι	Ι	Ι	Ι
Calamagrostis epigejos	Ι	Ι	Ι	Ι	Ι	Ι	Ι	+	Ι	Ι	Ι	Ι	Ι	Ι	Ι	Ι	Ι	Ι	Ι
Papaver rhoeas	Ι	Ι	I	I	Ι	I	Ι	I	Ι	Ι	Ι	I	I	Ι	I	Ι	+	Ι	Ι
Teucrium chamaedrys	Ι	Ι	Ι	Ι	I	I	I	+	I	I	I	I	I	I	I	I	I	I	Ι
Location of the releves: Durankulak beach, Bu	ulgaria	h (R1-	-4); S	fantu	Gheoi	rghe,	Danube ]		Delta (R5-9); Sul	5-9);	Sulir	ia, Da	nube	Delta	(R10	-13);	Card	on, Di	nube

Location of the releves: Durankulak beach, bugana (n1-4), Delta (R14); Marine Sand Dunes Reserve, Agigea (R15-18).

Twelve threatened plant species according to The Red Book of the vascular plants of Romania<sup>3</sup>, the Red Data Book of the Republic of Bulgaria<sup>4</sup> and the Red List of Bulgarian vascular plants<sup>16</sup>, have been recorded in the phytocoenoses with *Convolvulus persicus: Alyssum borzaeanum* (CR in Romania, EN in Bulgaria), *Convolvulus persicus* (CR in Romania, EN in Bulgaria), *Silene thymifolia* (VU in Romania, EN in Bulgaria), *Eryngium maritimum* (VU in Romania, EN in Bulgaria), *Elymus farctus* subsp. *bessarabicus* (CR in Romania), *Petasites spurius* (CR in Romania), *Cakile maritima* subsp. *euxina* (EN in Romania), *Crambe maritima* (EN in Romania), *Colygonum oxyspermum* subsp. *raii* (syn. *Polygonum mesembricum*) (VU in Romania, NT in Bulgaria), *Astragalus varius* (VU in Romania), *Scolymus hispanicus* (VU in Romania), *Ammophila arenaria* subsp. *arundinacea* (CR according to Ref. 3, but most probably already extinct in Romania).

Species such as Ammophila arenaria subsp. arundinacea, Polygonum oxyspermum subsp. raii, Cakile maritima subsp. euxina and Crambe maritima have been recorded only on Durankulak beach while Alyssum borzaeanum, Petasites spurius and Scolymus hispanicus were noticed only in the releves in Romania.

Of the twelve threatened taxa, five are critically endangered (in Romania), six are endangered (4 in Bulgaria and 2 in Romania), five are vulnerable (in Romania) and one is near threatened (in Bulgaria) (Fig. 4).



**Fig. 4**. Number of species belonging to different Red Data Categories in Romania and Bulgaria (CR – critically endangered, EN – endangered, VU – vulnerable, NT – near threatened)

The high percentage of rare species (26%), compared with the total number of recorded taxa within the plant association (46 taxa), emphasises the conservation importance of the plant community *Convolvuletum persici*. According to Table 1, the most numerous rare plants are on the beach of Durankulak (9 taxa) and fewer of them on the beach of Sulina (4 taxa), in the natural reserve of Agigea (4 taxa) and on the beach of Sfantu Gheorghe (3 taxa).

## CONCLUSIONS

Phytocoenoses with *Convolvulus persicus* are very rare on the western coast of the Black Sea. Such type of plant communities have been noticed in the Danube Delta, in the natural reserve of Agigea and on Durankulak beach in Bulgaria.

The plant association *Convolvuletum persici* can be generally found in the habitat 2110 but on the Durankulak beach, phytocoenoses with *Convolvulus persicus* reach the area of habitat 1210.

In the Danube Delta Biosphere Reserve, the phytocoenoses with *Convolvulus persicus* are vulnerable mainly to anthropogenic pressures such as grazing, tourism or expansion of the recreational areas on the beaches. The parasitic plant *Cuscuta* sp. is another serious threat for the local populations of *Convolvulus persicus*, especially on the beach of Sulina.

On the southern coast of Romania, *Convolvulus persicus* is well preserved in the Natural Reserve of Agigea where its favourable conservation status is a consequence of some effective management measures for the dune habitats.

The Durankulak beach is currently the only location in Bulgaria in which *Convolvulus persicus* was confirmed in the last twenty years; here, it has a favourable conservation status in the frame of the Nature 2000 protected site BG0000154 Ezero Durankulak.

The ecological preferences of *Convolvulus persicus* for the dune habitats and the syntaxonomic affiliation of the accompanying species indicate that the association *Convolvuletum persici* should rather be included in the order *Festucetalia vaginatae* Soó 1957 than in *Cakiletalia maritimae* R. Tx. apud Oberd. 1949.

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Ecology

# EVALUATION OF THE SOIL PROPERTIES VARIABILITY IN RELATION TO DIFFERENT CROP TYPES

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**Abstract**. The aim of the study was to evaluate the variation of the chemical properties of the haplic luvisol under different cultivation conditions (cropland, orchard and meadow) and the influence of this variability on the main ecophysiological groups of the soil microorganisms. The soil samples were collected from an experimental plot located in Cauaceu locality area, Bihor County, Romania, in spring and autumn of the year 2014–2015. Some soil chemical parameters, total number of aerobic heterotrophic bacteria, *Actinomycetes* and fungi were analyzed. Data was subjected to the two-way analysis of variance (ANOVA) (P = 0.05), and in order to determine the samples means statistical differences the Tukey test of pairwise comparisons was used. The results indicate significant differences between soil cultivation practices, for majority of the studied parameters. This fact is also revealed by the multivariate analysis (Principal Component Analysis, Linear Discriminant Analysis, Multivariate Analysis of Variance MANOVA (P = 0.05) and Hierarchical Cluster Analysis).

Keywords: crop type, soil parameters, microorganisms abundance, variability.

# AIMS AND BACKGROUND

In Romania there is a lack of detailed information about soil microorganisms variability and its effect on land use types. Spatial variability is a variation in soil properties which occurs with distance, while temporal variability is a seasonal variation in certain soil properties that display continuous variation depending on the activities on them<sup>1,2</sup>. Spatial variability could be attributed to changes in macro and microflora and fauna<sup>3,4</sup>. The spatial variability is universal to all soils and could be induced by differences in weathering rates, lithology, topographic differences and hydrological characteristics of the soil<sup>5,6</sup>. Other researches have shown that nutrient mining, absence of fallow periods, use of inappropriate farm-

<sup>\*</sup> For correspondence.

ing practices and frequent changes in land uses (over-cultivation), variation in micro-climate, vegetation, parent material, and crops grown escerbate degradation, resulting to constant plummeting of soil fertility levels and productivity<sup>7–9</sup>. Assessment of soil quality and productivity in various agricultural management systems involves essentially the use of bio-physical and chemical soil attributes<sup>10,11</sup>, but the complexity of these properties and their interactions in addition to other environmental variables make the study of crop response to soil very difficult<sup>11</sup>. Many reports showed that soil and crop variability are mostly observed in the fields where fertilities are low and where there is practice of more than one kind of land use within an area<sup>12</sup>. The objective of this study was to assess the variation of the soil properties in relation to land use types. The result of the study could be of great importance for agricultural research.

# EXPERIMENTAL

*Study site*. The Crisurilor Plain is part of the Western geobotanical region of Romania known as the Cris Plain District. The research was carried out in 2014 and 2015 on the haplic luvisol cultivated in three variant such as: cropland (cultivated in 2014 with wheat and in 2015 with maize), orchards and meadow. Soil samples were collected from plots of an experimental field localised at 10 km from Oradea, at village Cauaceu.

*Field data collection*. Sampling took place twice a year, in spring and autumn. For each site were selected six sampling locations. In each location was established a centre from which was drawn a circle with 20 m radius. Soil samplings were taken in accordance with the classical square method that uses five sampling units: one in the square centre and four in the square corners. There were done two replications (by square method) with the same centre, but with shifted squares corners. The sampling depth was 0–20 cm and 20–40 cm. The weight of one sample from each square point was 25 g. The five soil samples from each point of the square (including the centre) were mixed in the laboratory resulting 125 g of soil sample for each replication, resulting n = 12 measured values for each parameter.

Soil analysis. The physical and chemical properties of the soil samples were determined as follows: moisture content (Ur,%) was determined using the gravimetric method by oven-drying fresh soil at 105°C (drying oven SLW 53 POL EKO); soil acidity (acid., me/100 g soil) was determined by means of the Kappen procedure; pH in 1:2.5 soil water suspension using a pH-meter Crison; organic matter content (humus, %) using the Walkley-Black method; nitrate nitrogen (NO<sub>3</sub>–N, ppm) was determined using the colorimetric method and ammonium nitrogen (N–NH<sub>4</sub>, ppm) with Nessler reagent (UV-vis. spectrophotometer UV-1700, CPSA 3320.65-ISO 9001, BRML, Shimadzu, Japan). Available phosphorus was determined colorimetrically by the molybdenum blue method. The potassium content in the extract

1306

was determined by flame-photo meter. The content in organic matter content (humus, %) was determined using the Walkley-Black method. The method used to determine organic carbon (%) was the wet oxidation method and dosage titration (TOC VCSH, CPSA3320.65-ISO 9001, Shimadzu, Germany) and the macro-Kjeldahl method for total nitrogen (%) (Kjeldahl system ISO 9001, BRML, Italy, Velp, CPSA 3320.53) (Ref. 13).

*Bacteriological analysis*. The quantitative variation of the three ecophysiological microbial groups was studied: aerobic heterotrophic bacteria, *Actinomycetes* and fungi. The plate count method was used to estimate the total number of microorganisms on different nutrient medium such as: a sterile solid nutrient medium containing meat extract for aerobic heterotrophic bacteria (pH 7.5, incubation at 37°C, 3 days) (Refs 14 and 15), *Sabouraud* dextrose medium for the total number of fungi (pH 5.4–5.6, incubation at 25°C, 4–5 days). *Actinomycetes* were isolated by spread plate technique on *Actinomycetes* isolation agar (pH 6.5–8.0, incubation at 28°C for 4–5 days). The inoculated Petri dishes were incubated in the incubator Memmert UNB 100 and after incubation the obtained counts (the colonies of microorganisms were counted with the colony counter POL EKO LKB2002) were multiplied by the dilution factor (10<sup>6</sup>) to obtain the number of colony-forming units (CFU) per gram of soil<sup>16</sup>.

Statistical analysis. Data was subjected to two-way analysis of variance (ANOVA) (P = 0.05), and in order to determine the samples means statistical differences the Tukey test of pairwise comparisons was done (Minitab software, Minitab, Inc. Quality Plaz, 1829 Pine Hall Road, State College, PA 16801 USA). Multivariate analysis was done following the sequence: principal component analysis (PCA), and (P = 0.05), in order to determine the possible variables grouping and samples clustering<sup>17,18</sup>.

# **RESULTS AND DISCUSSION**

Significant differences between the crop type and between the two years studied in relation to the counts of microorganisms and the physical and chemical properties of the soil were assessed using two-way ANOVA (Table 1, Figs 1–3), principal component analysis (PCA) (Table 2, Fig. 4), linear discriminant analysis (LDA) (Fig. 5) and multivariate analysis of variance (MANOVA) (Table 3). The highest value of the moisture content (%) was measured in 2014 in wheat crop followed by orchard and meadow in the year 2014 and orchard and maize crop in 2015. The lowest value of the moisture content two years studied is not statistically significant. Under the influence of the maize crop the use of mineral fertilizers produced the soil alkalinisation (Fig. 1). The highest value of pH was measured in maize crop in the year 2015. Soil under the maize crop showed lowest values of pH (the wheat

crop acidifies the soil) but in orchard was registered the lowest value of pH. The soils with high levels of pH are not suitable for orchards<sup>19</sup>. The interaction between the two years studied is statistically significant (Fig. 1). Under the wheat and maize crops was registered a decrease of the hydrolytic acidity (me/100 g soil). The highest values were measured in orchard in the year 2014 and in meadow in 2015. The interaction between the years is statistically significant. The highest values of the humus content (%) were registered in maize crop in the year 2015 and the lowest in orchard and meadow in the year 2014. The highest values of the humus content were found in the cropland in comparison with other variants. The year factor is the dominant factor in the variation of the humus content (%). The use of the mineral fertilisers has led to an increase in nitrate nitrogen (ppm) content which showed high values in 2014 in wheat crop and also in maize crop in the year 2015. The concentration of N-NO<sub>3</sub> was higher in cropland showing that the process of nitrification is more developed in this soil. The year factor is not a dominant factor with effects in the variation of the nitrate nitrogen content. The content of ammonia nitrogen (ppm) presented high values under the maize crop in 2015 and low values in the year 2014. This parameter has not varied significantly depending on the years of the study (the interaction was not statistically significant) (Fig. 1). Concerning the variation of the parameters mobile phosphorus, mobile potassium, organic carbon and total nitrogen, their values varied significantly in both years studied, the interaction for the year factor being statistically significant. The highest values of the concentrations of these parameters were determined in wheat and maize crop and the lowest in uncultivated soil and in orchard (Figs 1 and 2). The dominant factor with effects on the variation of total number of aerobic heterotrophic bacteria (UFC/g soil) was the crop type. The highest values were measured in the year 2014 in meadow. The treatments with pesticides and fertilisers can affect the development of these microorganisms the lowest values being registered in orchard in 2015. In 2014, in the wheat crop the number of aerobic heterotrophic bacteria was more higher in comparison with the number found in 2015 in the soil of maize crop (Fig. 3).

nation of the physical, chemical and microbiological properties of the soil (values are expressed as mean $\pm$ standard deviation)	OIS
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Parameters	Maize crop_2015 Meadow_2014	Meadow_2014	Meadow_2015	Orchard_2014	Orchard_2015	Wheat crop_2014
Ur (%)	11.07bc±4.183	12.53abc±1.795	9.28c±3.742	13.64ab±2.791	11.20bc±3.159	15.59a±2.139
Hd	7.88a±0.151	6.19bc±0.186	6.06bc±0.196	$5.80c \pm 0.289$	6.24b±0.277	7.30a±0.646
Acid. (me/100 g soil)	0.90ab±0.646	2.25b±1.503	3.54a±0.947	3.56a±0.737	3.04ab±0.819	$0.62c\pm0.360$
Humus (%)	3.07ab±0.289	$1.98b\pm 0.552$	2.60ab±0.744	1.89b±0.369	2.82a±0.673	2.51ab±0.345
$N-NO_3$ (ppm)	14.10ab±5.544	2.98c±0.657	1.83c±0.488	5.05c±0.895	6.05bc±3.094	15.48a±6.786
$N-NH_4$ (ppm)	5.85a±7.158	2.18a±1.473	3.83a±1.428	1.35a±0.962	2.30a±1.809	0.56a±0.392
Mobile P (ppm)	338.00ab±131.534	17.75c±9.910	32.25c±29.227	25.00c±12.380	$25.00c\pm 12.380$ 159.25b\pm 142.023	266.25a±67.104
Mobile K (ppm)	710.00b±40.261	117.50d±25.357	172.50cd±67.895	115.00d±20.467 2	$15.00d\pm 20.467$ 297.50bc $\pm 158.045$	877.50a±249.154
C (%)	1.78ab±0.167	$1.15b\pm 0.319$	1.51ab±0.432	$1.09b\pm 0.215$	1.63a±0.389	1.45ab±0.200
N (%)	0.16a±0.008	0.11ab±0.026	0.13a±0.036	0.10b±0.022	0.13a±0.030	0.14a±0.016
AMH (UFC/g soil)	25725000.00bc	32283300.00a	10134300.00bc	16928800.00b	5224610.00c	27250000.00a
	$\pm 5424500.000$	$\pm 6189780.000$	$\pm 11228400.000$	$\pm 5293390.000$	$\pm 4723360.000$	$\pm 1937310.000$
ACT (UFC/g soil)	11440700.00b	16081600.00ab	520975.00b	2295500.00b	532900.00b	24389800.00a
	$\pm 19592400.000$	$\pm 11197800.000$	$\pm 604028.000$	$\pm 167750.000$	$\pm 833045.000$	$\pm 27344400.000$
FNG (UFC/g soil)	185755.00ab	51750.00b	276000.00b	1930920.00a	968550.00b	319500.00b
	±311962.000	±18427.500	$\pm 204746.000$	$\pm 1016760.000$	$\pm 1297440.000$	±425629.000

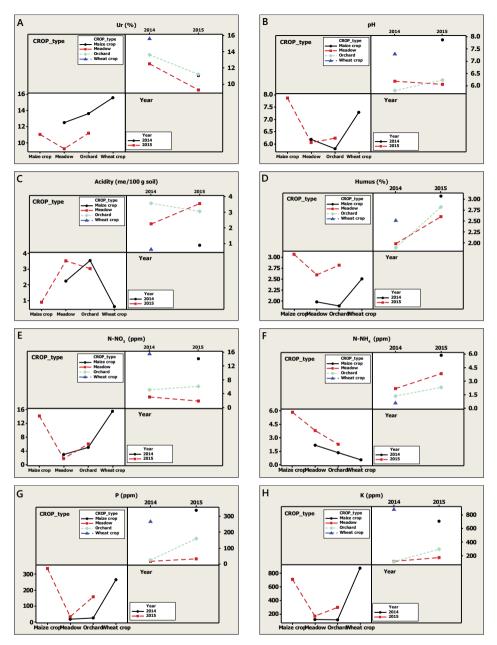
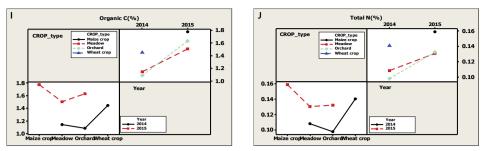


Fig. 1. Interaction plots (two-way ANOVA) for the factors crop type and year for the investigated physical and chemical properties of the soil



**Fig. 2**. Interaction plots (two-way ANOVA) for the factors crop type and year for the investigated physical and chemical properties of the soil (continuation)

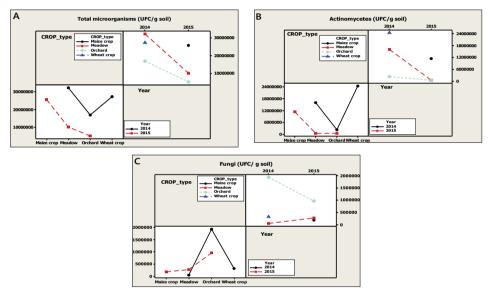


Fig. 3. Interaction plots (two-way ANOVA) for the factors crop type and year for the investigated microbiological properties of the soil

The highest values of the total number of *Actinomycetes* were measured in wheat crop in 2014 and in meadow in 2014. The lowest values were found in orchard in both years due to low pH values (Fig. 3). The orchard soil presented in both years studied a higher number of fungi in comparison with cropland and meadow where registered the lowest values (Fig. 3). PCA analysis was calculated using the correlation matrix of the variables and the between group algorithm. First two principal components explain 86.41% from the total variance of the data. The first three principal components explain 96.35% from the total variance of the data (Table 2). Wheat crop\_2014 and Maize crop\_2015 samples are fully discriminated from the other samples. Orchard\_2014, Meadow\_2014 samples are overlapping; also, Orchard\_2015, Meadow\_2015 samples are overlapping, too. However the Orchard and Meadow yearly sample are not overlapping, thus the Year factor for

these samples is a fully discriminating one. Due to their strong correlation between the variables, the PCA biplot (Fig. 4) also reveals the presence of variables grouping. First variables group is positively strong loaded (i.e. correlated) with the first principal component (i.e. axis) and contains the variables: N-NO<sub>3</sub>, Mobile P, pH and Mobile K. Second group contains the variables: Ur, LgACT and LgAMH. These variables are positive strong correlated with the second principal component and positive week correlated with the first one. The third group gathers the variables: N, C, humus and N–NH<sub>4</sub>. These variables are negatively strong correlated with the second principal component and week positive correlated with the first principal component.

Principal component	Eigenvalue	Variance (%)
1	7.2136	55.489
2	4.02029	30.925
3	1.29263	9.9433
4	0.391195	3.0092
5	0.0822802	0.63292

Table 2. Principal components statistical results

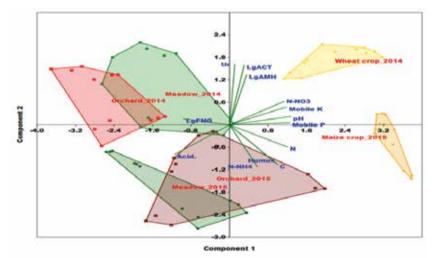


Fig. 4. Principal component analysis (PCA) biplot

Furthermore, the Wheat crop\_2014 sample has highest abundance of the first and second variables group. The Maize crop \_2015 sample displays the highest abundance only for the first variables group. The Orchard\_2014 and Meadow\_2014 have highest abundance of LgFNG and Acid variables, followed by high abundance of second variables group. The Orchard\_2015 and Meadow\_2015 samples have highest abundance of Acid variable (Fig. 4). To alleviate the samples groups overlapping in PCA biplot, there was used the linear discriminant analysis (LDA) which uses canonical projections similar with the PCA method but aims to increase the linear distance between the samples groups (i.e. to get a better discrimination). Figure 5 presents the 3D representation of the samples groups using the first three canonical axes of LDA. The MANOVA statistical results (P = 0.05) are presented in Table 3 where can be noticed that all six sample groups are validate (with 95% accuracy) as sample clusters.

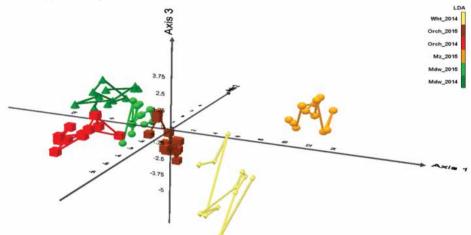


Fig. 5. 3D representation of LDA grouping

<i>p</i> – value	Meadow_ 2014	_ Meadow_ 2015		Maize 4 crop_2015		Orchard_ 2015
Meadow_2014		< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001
Meadow_2015	< 0.0001		< 0.0001	< 0.0001	< 0.0001	< 0.0001
Wheat crop_2014	< 0.0001	< 0.0001		< 0.0001	< 0.0001	< 0.0001
Maize crop_2015	< 0.0001	< 0.0001	< 0.0001		< 0.0001	< 0.0001
Orchard_2014	< 0.0001	< 0.0001	< 0.0001	< 0.0001		< 0.0001
Orchard_2015	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	

# CONCLUSIONS

The results of the study clearly showed the variability of the chemical and microbiological soil properties of the study area. The variables abundance analysis (from the PCA biplot) reveals the fact that the Maize crop\_2015, followed by Orchard\_2015 and Meadow\_2015 samples contain the lower abundance of the microorganisms. If Maize crop\_2015 sample microorganisms have a natural behaviour, the Orchard\_2015 and Meadow\_2015 samples present a negative environmental effect of the chemical treatments that down shifted the higher microorganism abundance displayed from the 2014 year.

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# SOIL POROSITY AND COMPACTION AS INFLUENCED BY TILLAGE METHODS

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Abstract. The research referred here has been carried out in three forest nurseries of Forestry Department in Arad, Romania during 2014–2015, on different soil types. The following tillage methods have been tested in order to point out their influence on soil compaction: classical tillage system (plough + 2X disks) and minimum tillage system (paraplow + rotharrow). The influence of several conditions on soil compaction has been described in our previous studies. In the actual study, a more in-depth analysis of the studied process is presented and discussed, especially through mathematical tools. Multivariate analysis was performed in order to obtain information about the tillage systems statistical differences when considering all variable at the same time. The considered variables are: the total porosity (i.e. TotPoros) and soil compression degree with values at three depth levels: 0-10 cm (i.e. d0), 10–20 cm (i.e. d1), 20–30 cm (i.e. d2). At first point, there was performed the principal component analysis (PCA) with the correlation matrix as input and between groups algorithm. The PCA biplot does not come with consistent information about how considered tillage systems modify the soil properties in quantitative and qualitative ways. However, PCA results shows that soil bulk density and compaction degree are strong correlated and the first variable can be excluded from further analysis. Thus, the multivariate analysis of variance (i.e. MANOVA) followed by canonical variants analysis (i.e. CVA) were performed. The CVA is a generalised version of linear discriminant analysis (LDA) when there are more than two groups involved – as in our case, with 15 samples of combined nursery location and tillage systems. In order to have a consequent samples grouping in CVA, the MANOVA pairwise between groups multi-comparison statistical significance p-results were done considering the sequential Bonferroni correction (data not shown due table huge size). This research attempted to emphasise the fact that the process of compaction plays a negative role on the physical-mechanical properties both in the classical and minimum tillage system. In the case of the minimum tillage system, the state of compaction of the soil is expected to reduce considerably in at least one year, without doing activities of fragmentations. An important role in the soil compaction is also played by the agricultural equipment, through their weight and traffic. Using multivariate analysis we were able to highlight precious information about soil tillage executed in three nurseries for the three sampling depths considering the values of bulk density, total porosity and the degree of compaction. Finally the CVA and HCA analyses emphasised the samples clusters generated by variables total porosity and soil compaction degree, and the pattern of soil modifications after the analysed tillage systems.

Keywords: compaction, soil tillage, bulk density, total porosity, compression degree.

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## AIMS AND BACKGROUND

In the last decades, an enormous quantity of energy was spent at a global level to reduce the negative influences of the soil compaction caused by the influence of the anthropogenic activities. This research is justified by the existence of great agricultural area with compressed soils with weak aeration and weak development of the root of the forestry saplings<sup>1</sup>.

The soil compaction occurs at a certain degree of soil deformation. This fact can be noticed in the mass of the soil through the diminution of the structural pores, and thus the structural aggregates take the form of continuous facies<sup>2</sup>.

The soil degradation is influenced by a series of factors of mechanical, physicochemical and biological nature. The numerous and inappropriate mechanical works contribute to the destruction of the soil structure<sup>3</sup>.

The consequence of soil compaction is the limiting of root growth and inhibition of plant development and thus the reduction of forestry production<sup>4</sup>. Deep compaction can persist for a long period of time and, therefore, it can threaten soil productivity for a long time<sup>3–6</sup>. Efforts to improve deep compaction by deep loosening of soil are often expensive and inefficient, which is why it is best to prevent soil compaction by using appropriate or conservative technologies<sup>7</sup>. Studies conducted by many researchers have highlighted that the risk of undesirable changes in soil structure can be reduced by limiting the mechanical stress applied on soil<sup>5,8,9</sup>, and by limiting the precompression stress<sup>7</sup>. Nowadays, the impact of agricultural machinery on soil properties can be simulated<sup>10</sup> by means of compaction models<sup>4,6,10,11</sup>, which are an important tool for the developing strategy in order to prevent soil compaction<sup>5,10</sup>.

Thus is required the decrease of number of tillage operations and thereby of energy consumed by the machinery, while tillage works must be more efficient and executed in proper conditions. Soil structure obtained by processing is strongly affected by soil moisture, and there is certain moisture for optimum soil processing, called optimum moisture.

In Refs 3 and 8, Dexter showed that the proportion of lumps produced by soil processing at optimum moisture is higher for degraded soils.

Prevention of soil compaction is a significant measure in order to maintain or improve soil quality. Soils with good physical properties, qualitative, are easy to process, which is a prerequisite condition for reducing the energy required for their processing.

The degree of compaction is a static property (characteristic) of soil<sup>4,9,10</sup>. The properties of a particular soil type (as material) generally do not change when its state of compaction undergoes changes. Compaction is defined as the dynamic behaviour of the soil by which its degree of compaction increases<sup>7,9,10</sup>.

The soil structure and, implicitly, the properties which derive from it, represent both a morphological index, characterising different genetic types of soil, and an agronomic index, determining, in an indirect way, its fertility. The agronomic value of the structure is given by its influence on the settlement, of the rule of water and air<sup>10</sup>.

The degradation consists in the elongation and flattening of the aggregates, the apparition of the edges and corners, through a stocky settlement, the increase of the ratio of dusty material which, through the rain action, forms mud and passes through different states of plasticity, to finally harden and crack<sup>9</sup>.

Firstly, the compaction is due to the mechanical forces created by the traffic of the layouts and/or operations of soil processing with a high rate of humidity. The traffic of the layouts on the arable surfaces represents the main factor which contributes to the severe compaction of soils, more often in the last 10–20 years when the weight and dimension of the layouts grew considerably. When the potential of compaction of a layout is evaluated, one must take into consideration the contact pressure created by the wheels of the tractors on the soil and the total upload on the axis<sup>4</sup>.

## EXPERIMENTAL

The research was carried out in three forest nurseries Forestry Department Arad, Romania, during 2014–2015, on different soil types (*Iarac*: alluvial vertic – gleyed; *Agrisul Mare*: luvisol; *Iosasel*: eutricambosol). In the present paper, we display the results obtained after the working of the soil in the classical tillage system and minimum tillage system on the physical and mechanical properties. The work systems applied have been: classical tillage system (plough + 2X disc) and minimum tillage system (paraplow + rotharrow). The usefulness of this paper lays in the research data gathered, processed, analysed and exploited in order to provide a pertinent study material, which could be effectively used by the specialists in the design of obtaining saplings in the forestry nurseries and the choice of the tillage system for the optimum soil.

There were taken samples in the natural settlement with metallic cylinders of 100 cm<sup>3</sup>, in order to determine the physical properties at three levels in depth (0-10; 10-20; 20-30 cm); for each sample, the sampling was repeated six times, after the execution of each technical work.

*Statistical analysis*. All data were subjected to descriptive statistics KyPlot (Kyplot Version 5.0.2, http://www.kyplot.software.informer.com)<sup>12</sup>. Multivariate analysis: multivariate analysis of variance (MANOVA), canonical variants analysis (CVA) and hierarchical cluster analysis (HCA) was performed with P.A.S.T. version 3.04 statistical software, (Palaeontology Statistics, Copyright Oyvind Hammer and D.A.T. Harper (November 2014), http://folk.uio.no/ohammer/past/)<sup>11</sup>.

## **RESULTS AND DISCUSSION**

The bulk density is one of the main indicators of the settlement of the soil and also one of the determining factors of some of the properties of the soil. High values of the bulk density signify the decrease of the capacity to retain water, of the permeability, of aeration and the increase of the mechanical resistance opposed by the soil at works and moreover at the penetration of the roots; low bulk density can reduce sometimes the bearing, making difficult the traffic and the execution of the processing works of the germination bed.

The porosity (the lacunar space) registers higher values while the content of the soil grows in organic matter and offers some important indications in relation with some of the properties of the soil. Thus, high values indicate a high capacity to retain water.

The absolute values of the bulk density or of the total porosity cannot be interpreted accordingly in order to appreciate the state of settlement of the soil, because their practical significance is very different from soil to soil according to their texture.

The determination of the settlement of the soil is well taken by using a synthetic indicator which shows that the compression level and the deficit of total porosity are met. The indicator which includes the bulk density (total porosity) and takes into account the soil texture is the compression degree<sup>1</sup>.

Apart from its significance as general indicator of its state of settlement, the compression degree practically reflects the state of breaking up and compression of the soil. In certain situations, the elimination of the soil compaction is difficult to be carried out, but it is possible to minimise it through the proper soil management. It is easier to avoid the compaction rather than to eliminate it after its installation, because the correction measures can be expensive and cannot totally solve the problem<sup>3</sup>.

To synthesise more efficiently the data taken and to be able to describe completely the intrinsic characteristics of the sample, it was chosen a statistic processing with the aid of the program KyPlot (Ref. 12). The results obtained are given in Tables 1–3, having as a purpose to underline the variance of soil compression degree, comparative with the tillage system (minimum/classical systems). Thus, for each nursery were included in the experiment resulted in fifteen statistical indicators for each technical work performed in the two systems (minimum/classical), but also witness sample. The mechanical processing of the soil through traditional and modern methods is currently put under question due to the high energy consumption and the continuous degradation of the arable horizon through erosion and excessive compaction.

		~	-	-	
Statistical indicator	Witness	2	Soil compre	ssion degree	
	sample	classica	l system	minimun	n system
		classical plough	2X disks	paraplow	rotary harrow
Mean	27.940	0.737	1.397	15.523	32.130
SEM (average standard error)	0.376	1.545	0.627	1.484	1.787
Standard deviation	0.651	2.675	1.086	2.571	3.095
Coefficient of variation	0.023	3.632	0.778	0.166	0.096
Skewness	-0.692	-0.030	0.651	0.702	-0.563
Curtosis	-1.500	-1.500	-1.500	-1.500	-1.500
Mean deviation	0.750	2.697	1.243	2.967	3.490
Median	28.270	0.780	0.920	14.140	33.210

Table 1. Statistical indexes of variation of soil compression degree in Iarac nursery

Table 2. Statistical indexes of variation of soil compression degree in Agrisul Mare nursery

Statistical indicator	Witness		Soil compre	ssion degree	:
	sample	classica	l system	minimun	n system
		classical plough	2X disks	paraplow	rotary harrow
Mean	26.083	4.100	5.177	1.960	23.690
SEM (average standard error)	1.135	5.350	4.127	0.662	5.746
Standard deviation	1.965	9.267	7.149	1.146	9.952
Coefficient of variation	0.075	2.260	1.381	0.585	0.420
Skewness	0.700	0.707	0.338	0.509	0.411
Curtosis	-1.500	-1.500	-1.500	-1.500	-1.500
Mean deviation	2.267	10.700	7.733	1.280	10.920
Median	25.040	-1.140	3.810	1.610	21.330

Table 3. Statistical indexes of variation of soil compression degree in Iosasel nursery

Statistical indicator	Witness	Soil compression degree					
	sample	classica	l system	minimun	n system		
		classical			rotary		
		plough			harrow		
Mean	27.940	-20.463	-4.880	-1.663	1.717		
SEM (average standard error)	0.376	3.430	4.466	0.878	0.728		
Standard deviation	0.651	5.940	7.735	1.520	1.261		
Coefficient of variation	0.023	-0.290	-1.585	-0.914	0.735		
Skewness	-0.692	-0.010	-0.224	0.334	0.097		
Curtosis	-1.500	-1.500	-1.500	-1.500	-1.500		
Mean deviation	0.750	5.957	8.170	1.643	1.293		
Median	28.270	-20.430	-3.920	-1.950	1.650		

The soil processing in the classical tillage system leads to an excessive breakup through repeated interventions, leaving it without vegetal remains through the reversal of the clods in the ploughing process, thus being strongly eroded under the action of the water and wind.

Worldwide, there is the tendency to replace the classical tillage system of the soil, through the extension of the minimum work system, method recommended both from the point of view of the preservation of the soil and for the reduction of energy consumption.

In our country, the extension of these systems of soil processing in the forestry nurseries is slow because of the lack of unitary strategies to sustain the technology, the lack of unitary strategies to correspond to the biological requirements of each culture, the lack of specialised knowledge related to the new system.

*Multivariate analysis*. Multivariate analysis was performed in order to obtain information about the tillage systems statistical differences when considering all variable at the same time. The considered variables are: the total porosity (i.e. TotPoros) and soil compression degree with values at three depth levels: 0–10 cm (i.e. d0), 10–20 cm (i.e. d1), 20–30 cm (i.e. d2).

At first point, there was performed the principal component analysis (PCA) with the correlation matrix as input and between groups algorithm. The PCA biplot does not come with consistent information about how considered tillage systems modify the soil properties in quantitative and qualitative ways. However, PCA results shows that soil bulk density and compaction degree are strong correlated and the first variable can be excluded from further analysis. Thus, the multivariate analysis of variance (i.e. MANOVA) followed by canonical variants analysis (i.e. CVA) were performed. The CVA is a generalised version of linear discriminant analysis (LDA) when there are more than two groups involved – as in our case, with 15 samples of combined nursery location and tillage systems. In order to have a consequent samples grouping in CVA, the MANOVA pairwise between groups multi-comparison statistical significance *p*-results were done considering the sequential Bonferroni correction (data not shown due table huge size).

Figure 1 shows the CVA biplot and prescribes that the total variance explained by the first two axes (out of the 14 axes) is 95.78%. This means that was achieved the main goal of the CVA to reduce the number of variables that explains the tillage systems in several nursery locations. Variables that represent the soil compaction degree (SoilCompDeg\_d0, SoilCompDeg\_d1, SoilCompDeg\_d2) are positively correlated with the first principal axis (Axis 1). The total porosity variables (Tot-Poros\_d0, TotPoros\_d1, TotPoros\_d2) are negatively correlated with the first axis and somehow are opposing the soil compaction degree vectors. Geometrically, these variable vectors prescribe the direction on which the consequent variable is more abundant for the nearby samples. Furthermore, the variables vectors facilitate in soil properties analysis by emphasising the modifications of certain variables after applying the tillage systems.

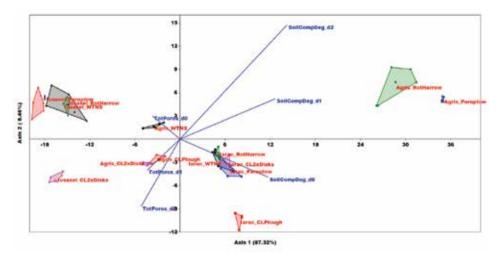


Fig. 1. Canonical variates analysis (CVA) biplot for all nurseries and all tillage systems

The hierarchical cluster analysis (HCA) was performed in order to get the clustering information about the soil properties modifications tillage systems in studied nurseries. Figure 2 presents the dendrogram that describes the 10 clusters that are confirmed by the MANOVA analysis. The input data for HCA consisted of the first two principal axis coordinates from the CVA analysis.

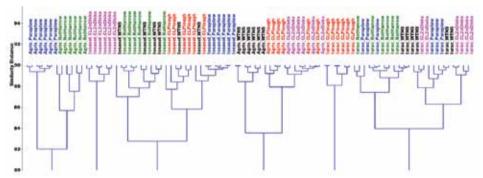


Fig. 2. Hierarchical cluster analysis (HCA) dendrogram with the clustering information for all nurseries and all tillage systems

For Iosasel nursery the CL 2X disks is the only tillage system that changes the soil total porosity at all depth levels. These changes are so high that CL 2X disks is clustered from the other four tillage systems (Fig. 2). The other tillage systems applied in Iosasel nursery are in the same cluster with the witness sample. This

means that the bulk density values are not affected in so large a share of the work carried out, perhaps a question could be sandy granulometric composition of the nursery. In the same way, for Iarac nursery, CLPlough is the only tillage system that changes the soil total porosity at depth levels d1 and d2, and the soil compaction degree at depth level d0. The other tillage systems applied in Iarac nursery are in the same cluster with the witness (WNTS) sample and means that the soil mentioned properties has no significant differences. Accordingly, the ploughing is the only work that changes the total porosity values, because after this work has been a inversion of soil layers, which are not carried to other works.

The Agris nursery displays a different clustering pattern from the other two. It gives four clusters. First cluster with WTNS sample is positively loaded (i.e. the variable has the most abundance) with total porosity at d0 level. Second cluster has samples: CL 2X disks and CLPlough, and is positively loaded with the total porosity at d1 and d2 levels. Third cluster with Paraplow sample and fourth cluster with RotHarrow sample are positively loaded with the soil compaction degree at depth levels d1 and d2. This clustering situation denotes that the Agris nursery has a complex behaviour at tillage systems emphasising huge modifications of the soil properties at d1 and d2 levels. This is not surprising because the first sampling depths (d1 and d2), the soil is much deconstructed, due to multiple interventions applied periodically.

## CONCLUSIONS

The process of soil compaction due to natural factors appears under the form of some genetic consolidated horizons. The situations which lead to the occurrence of the phenomenon of soil compaction are divided between the action of natural and anthropogenic factors.

During the action of the wheeling system of the tractors and the agricultural equipment on the soil, it is subjected to some mechanical efforts, which, through their action, make it shift laterally (refulation), vertically (compression) and horizontally (shear). The effect of the compression is transmitted in the layers of the soil in all directions, under the form of a pressure, and thus their propagation is insignificant at depths greater than 80 cm.

The physical characteristics like bulk density, total porosity and compression degree modify according to the soil works. The modification of these properties is hard to notice (except for the compression degree) during a year because the soil has the tendency, in normal conditions, to get back to the initial state and to estimate the negative effects which appeared after the impact produced by its processing with mechanical means. Several researches show that in a long period of time, the evolution of the physical properties in a certain direction takes place at a slow rhythm, after a short period of time when they start to stabilise. This research attempted to emphasise the fact that the process of compaction plays a negative role on the physic-mechanical properties both in the classical and minimum tillage system. In the case of the minimum tillage system, the state of compaction of the soil is expected to reduce considerably in at least one year, without doing activities of fragmentations. An important role in the soil compaction is also played by the agricultural equipment, through their weight and traffic.

Using multivariate analysis we were able to highlight precious information about soil tillage executed in three nurseries for the three sampling depths considering the values of bulk density, total porosity and the degree of compaction. Finally, the CVA and HCA analyses emphasized the samples clusters generated by variables total porosity and soil compaction degree, and the pattern of soil modifications after the analysed tillage systems.

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# CHARACTERISATION OF SOILS AND PARENT MATERIALS FROM REHABILITATED EMBANKMENTS IN THE AREA OF DPM INC., CHELOPECH

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**Abstract**. 'Dundee Precious Metals Chelopech' Inc. is a company for gold and copper mining and processing within the field of Chelopech. Over the years land disturbances by the mining processes have been analysed and a lot of reclamation projects had been implemented within the field of Chelopech. Purpose of these projects is mitigation the negative mining impact on the environment as well as restoration of an environmentally sound ecosystem within the village of Chelopech. Aim of this article is characterisation of soils and parent materials from rehabilitated embankments in the area of DPM Inc. as part of the ecosystem. Eight projects for reclamation are considered. They have been implemented in nine sites located in the northern part of the DPM during the period of 2004–2012. Results of soil sampling implemented in 2004, 2005 and 2015 are analysed in terms of: morphological description, mechanical and fractional composition, humus and primary nutrient content, acidity and buffering capacity, concentration of heavy metals. Soils and parent materials are characterised by high concentration of heavy metals and high acidity, caused by the mining and processing of gold and copper. The comparative analyses of soil indicators over the years prove that reclamation is relatively successful.

Keywords: mining, processing, land reclamation, soil.

# AIMS AND BACKGROUND

Aim of the study is to assess the state of soils and parent materials from rehabilitated embankments within the north site of DPM Inc., Chelopech as well as state of the adjacent lands influenced by the gold mining and the accompanying ameliorative measures.

The village of Chelopech is located in the low southern part of Stara planina in the region of Etropole. The altitude is approximately 700 m. One of the most famous fields for gold/copper ore is located in the western part of Chelopech. The field of Chelopech is unique in terms of its mineral composition – it contains more than 70 minerals. The ore is characterised with high concentration of copper and gold but also contains high amounts of sulphur, selenium, tellurium and arsenic.

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The field of Chelopech was first recorded in 1840 by Ami Boue who described the geological structure of the area. The first ore was discovered in 1930 and its mining began in 1953. During the 80s of the last century mining of copper and gold ore from the field of Chelopech reaches 500 000 t per year<sup>1</sup>.

In 2003 Dundee Precious Metals Inc. acquires the assets of the company. The main company activity is mining and processing of copper-gold ore from the field of Chelopech as well as production of copper-gold concentrate<sup>2</sup>.

Quality of soils in the region is influenced by the long operation of ore mining and processing companies before introduction of the current environmental standards and norms in the country<sup>3</sup>.

One of the first company projects after acquiring ownership and concession of the mining production is 'Working project for reclamation of contaminated sites by the processing activity of the company before the 70s as well as overall improvement of the northern part of the production site – 2003–2004'. The purpose of the project is to provide all legally required activities for mitigation the negative impacts by the mining process as well as complete restoration of green environment near the production site of the village of Chelopech<sup>3</sup>. Till 2012 the company implemented 8 projects on reclamation of disturbed areas from the north site. A map of location of the rehabilitated areas is presented in Fig. 1 (Ref. 4).



Fig. 1. Rehabilitated areas, located in the northern site of DPM

1 – Heap, mine 'West'; 2 – Regression, bl. 103; 3 – Regression, bl. 150; 4 – mine 'North'; 5 – Regression, bl. 19; 6 – Heap, mine 'Capital – West'; 7 – Heap, mine 'Capital – East'; 8 – Pastovo stopanstvo; 9 – Inclined gallery 'Nadezhda'

In accordance with the climatic zoning of Subev and Stanev (1959), the Zlatishko – Pirdopska valley falls in to the European continental climatic region in particular in the area of hilly and low areas of Western Medium Bulgaria<sup>5</sup>. Climate is temperate and influenced by the geographical location – in a deep valley between two mountains. Sredna gora mountain prevents the entry of Mediterranean climatic impact. The Continental character softens by Stara planina mountain that serves as barrier to cold winds. Mild winters and cooler summers, as well as temperature inversions, particularly in winter are typical compared to another regions in Bulgaria<sup>6</sup>.

In connection with designing the projects on rehabilitation a lot of researches and studies on soils, parent materials and the materials for rehabilitation of the embankments had been conducted.

The main rocks that are established in the area are with various petrological characteristics and age. The basis consists of gneisses, schists and amphibolites whose age is Precambrian and low-metamorphic rocks of Paleozoic age. Cretaceous rocks are developed on them. Silicates and aluminum silicates are the main rock-forming materials that interact with groundwater, and after reaching the surface – interact with the atmosphere. Carbonate minerals have been identified in the Triassic and Cretaceous rocks.

With regard to the acidity of the soil adjacent areas are heterogeneous in nature, but most of them are acidic, with low-buffering capacity.

As in the soil materials, as in bulk substrates, the sulphur is in large quantities – both general and mobile. Exactly the sulphur with its sulphates is generator of the high acidity. Arsenic is an essential concomitant heavy metals toxic element that can be found in highest quantities in the bulk materials and it is a major problem in the rehabilitation of the embankments. Its mobile forms do not necessarily decrease with a decrease in the acidity of the substrates and are the most dependent on the total arsenic.

According to data from 2005, bulk and soil materials are poor in organic matter and the amount of total nitrogen is below the required minimum (0.05%). The content of available phosphorus compounds in the soil material is low. Quantities of mobile phosphorus in the sterile materials are even lower -2-5-6 mg/100 g. The content of available potassium in the tested substrates is too varied, but for the most part soil materials and bulk substrates are rich to very rich in available potassium.

The mobile forms of heavy metals – copper, chromium, zinc and arsenic exceed the control values for protection of soil microorganisms, plant growth and soil filtration water. In some of the samples levels of copper, cadmium and lead exceed the values of contamination of food and feed plants<sup>3</sup>.

## EXPERIMENTAL

Subject to the study is the northern site of the company and the adjacent lands, including all disturbed and rehabilitated lands which are result from the mining activity since the beginning of exploitation the field and apparently undisturbed natural areas on the southern slopes of Stara Planina mountain.

11 soil samples are taken from three soil profiles and 13 groundbreakings. Morphological description of soil and bulk materials is made. Samples are analysed at accredited laboratory for the following parameters: pH, organic matter content and concentration of heavy metals.

## RESULTS AND DISCUSSION

Morphological description of the soil and bulk materials is presented in Table 1.

Table 1. Morphological description of the profiles, samples and places of soil sampling in the area of DPM, Chelopech

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Section/description of the place of	Depth of soil	Morphological description of the
soil sampling	sampling (cm)	profiles
1	2	3
$S_1$ Heel of mine 'West', next to internal road; bulk materials; vari- ous grass	average sample 30–40	red-brown; moist; sandy; rare; peb- bles and boulders – cover landmass from uncontaminated excavations in the area of DPM, Chelopech. Sharp transition to the embankment from mine 'West'
<b>S</b> <sub>2</sub> Heap mine 'West' – slope with inclination $\approx 40^{\circ}$ ; various grass, grass mixture from reclamation grass; oak, pine, acacia, elm; loose rock. Rock; there is not ground water; rehabilitated area; beginning of erosion	average sample 30–40	red-brown; moist; sandy; rare; peb- bles and boulders – cover landmass from uncontaminated excavations ir the area of DPM, Chelopech. Sharp transition to the embankment from mine 'West'
$S_3$ Stripped natural slope above the embankment; rare trees and shrubs –	0-30 (30)	red-brown; moist; sandy-clay; rare pebbles; sharp transition
elm, pine; diorite and gneiss; af- forestration; light erosion; clear differentiation between soil and the parent material	30 – ↓	yellow – grey; moist; stony; rare pebbles
$S_4$ Natural area above the Chugo- vishko dere. Steep eroded slope;	a <sub>0</sub> 14–0 (14)	grey – black; dry; very dry leaves and twigs;
afforested acacia, clematis; deep	a <sub>1</sub> 0–13 (13)	brown-black; dry; clay-sandy; struc-
soil litter; slate and granodiorites;		tured; rare; roots
light erosion	d 13–47 (34)	loose rock; dry; roots

to be continued

Continuation of Table 1

1	2	3
$S_5$ Deep ravine next to mine 'North' and under rehabilitated area; degraded trees and shrubs – elm, howthorn,	a <sub>0</sub> 5–0 (5)	light brown; moist; clay-sandy; rare; turf; sharp transition
cereal and various grass; dolomite, marl, marble and slate; there is not ground water; above the soil sam-	a <sub>1</sub> 0–20 (20)	light brown; moist; clay-sandy; rare; roots; gradual transition
pling – rehabilitated embankment from the mine; light erosion	a <sub>0</sub> 20–70 (50)	grey; moist; clay-sandy; rare; roots
$S_6$ Inclined gallery 'Nadezhda', western part of the rehabilitated area	average sample 30–40	red-brown; moist; sandy; rare; peb- bles and boulders – cover landmass from uncontaminated excavations in the area of DPM, Chelopech

Sections made for soil sampling in the northern part of DPM, Chelopech, are rather diverse both in origin and morphology.

The results from the laboratory analysis are presented in Table 2.

The assessment on state of soils and parent materials from rehabilitated embankments within the north site of DPM Inc., Chelopech as well as state of the adjacent lands influenced by the gold mining and the accompanying ameliorative measures is based on summary of the available information from conducted soil samplings in 2004 and 2005 connected with the reclamation projects and conducted analysis in 2015 connected with the current research. The assessment is presented separately for 7 of the rehabilitated areas which are located in the north area of the field (Fig. 1). Two of the areas – **3** Regression, bl. 150 and **5** Regression, bl. 19 are not assessed because they represent surface subsidence.

Table 2. Concentration of heavy metals, arsenic and sulphur in soil samples from the rehabilitated areas	avy metals, a	ursenic and	l sulphur in	soil sample	es from the	rehabilitat	ed areas				
Place of soil sampling	Depth	Hd	Organic	As	Cd	Cr	Cu	Ņ	Pb	Zn	s
	(m)	$(\dot{H}_2O)$	matter, org. C (%)	(mg/kg)	(mg/kg)	(mg/kg)	(mg/kg)	(mg/kg)	(mg/kg)	(mg/kg)	(%)
s,	average sample	4.07	0.168	175.0	1.7	11.9	424.7	8.5	78.8	176.5	0.7767
$S_2$	average sample	6.67	0.852	81.1	0.7	7.5	434.7	5.1	52.1	101.8	0.1283
S	0-20	4.78	0.553	139.7	1.5	9.3	315.9	6.7	95.5	153.3	0.2519
	20–70	3.31	0.249	502.2	3.6	5.6	579.8	3.5	355.5	135.9	1.6699
S.	5-0	5.59	6.141	104.9	2.5	20.1	564.0	14.9	93.2	119.2	0.2487
	0-20	5.28	3.674	67.0	0.7	22.7	331.8	13.9	61.4	<i>77.9</i>	0.1111
	20–70	5.11	1.175	61.3	<0.5	10.3	206.6	9.2	37.4	64.6	0.0392
S <sub>5</sub>	$a_0 5-0$	8.38	1.012	24.8	<0.5	25.2	107.7	20.5	52.0	82.6	0.0583
	a 0–20	8.34	0.759	24.4	<0.5	26.7	107.4	24.9	42.1	75.0	0.0448
	b20–70	8.49	0.635	25.4	<0.5	25.3	115.6	23.4	29.3	76.0	0.0382
Š	average sample	4.13	0.507	270.5	2.2	12.2	297.2	7.9	187.7	87.6	0.5443
* Maximum allowable concentration (MAC)	Ι	Ι	Ι	40	10	300	500	250	500	600	
*Interventional levels	Ι	Ι	I	120	40	600	1000	700	1000	1500	I
*Background concentra- tion	I	I	I	10	0.4	65	34	46	26	88	I
*Precautionary values	Ι	Ι	Ι	15	0.6	90	50	60	40	110	Ι
MAC – maximum allowable No 3/SG. 71, 2008.	concentration; * interventional level, background concentrations and precautionary values in accordance with Ordinance	on; * inter	ventional le	vel, backgr	ound conce	entrations a	nd precauti	ionary valu	es in accor	dance with	Ordinance

1 - Heap, mine 'West'. The reaction of the soil solution in samples from Heap, mine 'West' is highly acidic. It has not changed over the years. The reason for this is the high concentration of sulphur with its sulphates.

Content of soil organic matter is very low. This assessment is determined by the active erosion, as well as by the typical for the area low forest litter. The area is characterised with high deforestation, degraded and rare tree communities that enrich soil with organic matter (through leaves and roots).

Exceeding values for heavy metals was found in water samples from the area due to leaching of rocks and waste in the embankment.

In terms of heavy metals and metalloids arsenic is with the highest concentration over the years, incl. in the last analysis in 2015. It exceeds the maximum allowable concentration (up to 30 times in 2005 and 4.3 times in 2015) and the interventional level (with 5.66 times in 2005 and 1.5 times in 2015). Concentration of cadmium, copper, lead and zinc is overstated and exceed the precautionary values.

While in 2005 concentration of copper in soil samples exceeded the maximum allowable concentration, there is not such an excess in 2015.

The area of Heap, mine 'West' is covered by shrubby forms of elm (*Ulmus minor*), acacia (*Robinia pseudoacacia*), pine (*Pinus sylvestris*) and various grass – cereals, legumes, mosses.

2 - Regression, bl. 103. Soil materials from the rehabilitated Regression, bl. 103 are characterised with colour, texture and stony inclusions in the soil same as for natural areas. Unlike the natural soils where soil reaction is weakly to moderately acidic on the rehabilitated area it is highly acidic to very acidic in depth of the soil profile. It is necessary to bring additional quantities of lime materials with increased rate in order to reduce soil acidity.

Content of humus is very low. Here, as with Heap, mine 'West' it is explained by the lower organic litter. Proof is the fact that content of humus in the natural area in Chugovishko dere is assessed as very high which is result from high organic litter.

The root layer of the rehabilitated area is with high values of arsenic and copper, that excess up to 12.5 times the maximum allowable concentration for arsenic and for copper – up to 1.16 times, arsenic exceeds 4.1 times the interventional level. Concentration of copper, lead and zinc in soil samples from the rehabilitated area is above the precautionary values. The same excesses are found on close situated natural areas that serve as comparison – stripped natural slope and natural area above the Chugovishko dere. There is a difference – concentration of heavy metals decreases in depth of the soil profile in the natural areas, while in the rehabilitated area it increases. This due to the concentration of heavy metals in the rock materials that builds the embankments. The goal of successful rehabilitation is to reduce their mobility and to bring them into condition that does not allow infiltration into surface and ground water and thir subsequent absorption by plants.

4 - mine 'North'. Natural soils in the area of mine 'North' are dark to black. Results of periodic studies over the years have shown that reaction of soil solution is moderately alkaline throughout the soil profile. There is no need for further introduction of lime materials.

Content of humus is low to very low similar to the other areas in the region. Soils are characterized with very low reserves of nitrogen, phosphorus and potassium compounds.

Concentration of heavy metals is relatively evenly distributed through the soil profile. High pH values determine weaker mobility of the mobile forms of heavy metals. Exceedances of the maximum allowable concentration and the interventional level are not found.

Concentrations of copper, arsenic and lead exceed the precautionary values, with 1.69, 2.31 and 1.3 times, respectively.

The natural area is covered with stones (boulders) from water erosion, and between them is overgrown with wide variety of grasses, shrubs and degraded individuals of elm (*Ulmus minor*), hawthorn (*Crataegus monogyna*), dogwood (*Cornus mas*).

There is light erosion process.

**6** – *Heap, mine 'Capital* – *West'*. Soils from the rehabilitated area of Heap, mine 'Capital – West' are with medium to heavy sandy clay mechanical structure. Soil reaction is slightly to highly acidic. The pH values in water and CaCl<sub>2</sub> are too close, which means that large part of the acid ions are mobile and easy to be taken by the root system of the plants.

Content of humus is very low, resulting in small to medium soil reserves with nitrogen. Soil reserve with potassium and phosphorus compounds is also very weak.

Soil materials analysed for heavy metals show an excess of the maximum allowable concentration for copper with 1.6 times and for arsenic – with 3 to 27 times. Arsenic also exceeds the interventional level with 1.6 to 8.9 times.

Exceeding the precautionary values is established for all heavy metals and metalloids, with the exception of chromium, in which even the background concentration is not exceeded.

Vegetation on the rehabilitated area is represented by birch (*Betula pendula*), acacia (*Robinia pseudoacacia*), beech (*Fagus sylvatica*), pine (*Pinus sylvestris*) and various grass.

7 – *Heap, mine 'Capital – East'*. Soils from the rehabilitated area Heap, mine 'Capital – East' are with medium to heavy sandy clay mechanical structure.

Soil reaction is slightly to very acidic, which due to the high concentration of sulphur.

Content of humus is very low, as well as soil reserves of nitrogen, phosphorus and potassium compounds.

Concentration of heavy metals shows excess of the maximum allowable concentration and the interventional level by arsenic, respectively with 35.3 and 11.8 times. Concentration of copper also exceeds the maximum allowable concentration and the interventional level with 2.5 and 1.26 times, respectively.

There is an excess of the background concentrations and the precautionary values by all heavy metals except nickel and chromium.

There is birch (*Betula pendula*) on the rehabilitated area.

8 - Pastovo stopanstvo. Soils in the area of Pastovo stopanstvo are characterised with medium sandy-clay to clay-sandy mechanical structure.

This rehabilitated area is with low organic matter similar to the other rehabilitated areas. Soil reserves with nitrogen and phosphorus are very low. Potassium compounds provide soil with average availability.

Concentration of arsenic exceeds the maximum allowable concentration with up to 4.13 times and the interventional level with 1.38 times.

Concentration of all of the heavy metals exceed the background concentrations and precautionary values, with the exception of chromium and nickel.

9 - Inclined gallery 'Nadezhda'. Soils from the Inclined gallery 'Nadezhda' are with medium to heavy sandy-clay mechanical structure. Soil reaction is strongly acidic. Soils are characterised with low sorption capacity and low rate of exchange base saturation.

Content of humus is very low to medium. Soil reserves with nitrogen are low to medium. Phosphorus compounds are with low concentration. Soil reserves with potassium compounds are medium.

While in previous analysis was established exceeding of the maximum allowable concentration by arsenic with up to 50 times and the interventional level with 16.9 times, the results from 2015 show an excess over the maximum allowable concentration with 6.76 times and over the interventional level with 2.25 times.

Also in previous analysis was established exceeding of the maximum allowable concentration by copper with 2.56 times and exceeding the interventional level -1.28 times, but in 2015 the concentration of copper is under the maximum allowable concentration.

The precautionary values are exceeded by copper, cadmium and lead.

There is pine (*Pinus sylvestris*) and black pine (*Pinus nigra*), oak and grass cover of grasses and mosses on the rehabilitated area of Inclined gallery 'Nadezhda'.

Light erosion is established.

# CONCLUSIONS

As a result of this study it can be concluded that reclamation has a relatively good success, for example the reclamation of mine 'West', mine 'North' and the Inclined gallery 'Nadezhda'.

State of the rehabilitated embankments is relatively good. Major problem for the rehabilitated areas as well as for part of the undisturbed soils in the area remains the high concentration of heavy metals and high acidity caused by mining and processing. Concentration of copper, arsenic, and in some areas of lead, zinc and cadmium is above the precautionary values. In some of the areas it is even above the maximum allowable concentration.

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# **RESTORATION PROCESSES IN ECOSYSTEMS WITHIN THE REHABILITATED MINING SITES OF DPM INC., CHELOPECH**

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**Abstract**. The soil-forming proceeds as a complex of biological, physical and chemical processes. In its initial stage the soil-forming process is connected with development of bacterial and actinomycete forms, which are capable to do decomposition of primary and secondary minerals and forming chelates. Successional processes run as a result of the initial soil-forming. These processes are initial phase of biological restoration. Low profile soils form. Studying the initial soil-forming processes on disturbed lands for purposes of the biological reclamation provides to the investigators valuable information on the speed and stages of soil-forming. These studies provide information and justify the necessary ameliorative measures for speeding the succession and soil-forming processes. The purpose is successful biological reclamation and development of sustainable ecosystem. The aim of this study is to carry out a comparative analysis of the initial soil-forming process within the rehabilitated mining sites of DPM Inc., Chelopech. The comparison is between sites without ameliorative measures, with afforestation, and sites with conducted certain ameliorative measures for speeding the successional processes on disturbed areas. The most appropriate conditions for speeding up soil formation and restoration of the disturbed ecosystems are established after analysing the data.

Keywords: restoration, ecosystems, soil-forming, reclamation.

# AIMS AND BACKGROUND

The aim of this study is to carry out a comparative analysis of the initial soilforming process between sites without ameliorative measures, with afforestation, and sites with conducted certain ameliorative measures for speeding the successional processes on disturbed areas.

Seven large facilities for management of mining waste and landfills for metallurgical waste are located within the Pirodpsko-Zlatishka valley. They don not only disturb the landscape because of the high reactivity of the materials accumulated there, but they also represent source of contamination the air, water and environment with toxic elements. In Bulgaria there is not another industrial district with such a high amount of mining waste – embankments and tailing ponds located in

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such a small area and representing diffuse sources of particulate emissions. The companies operating the tailing ponds within the valley contribute to the studies on mitigation the soil contamination. After the companies had been privatised the tailing ponds comply with the existing regulations and their operating is in accordance with the best world practices.

Soils in the Zlatishko-Pirdopska valley have been first recorded by N. Poushkarov. In his papers in 1919–1920 he describes the orography, hydrography within the valley, geology, petrography, subsoil moisture, vegetation. He identifies the soil types and links them with the sediments as well as makes fairly complete characterisation of their physicochemical properties<sup>1</sup>.

During the 70s of the XX century experts from the Institute of soil science 'Nikola Poushkarov' began research on the problematic areas affected by industrial contamination within the companies from the Zlatishko-Pirdopska valley. At the same time Chulzdhian and Hinov make a research on the valley in terms of concentration copper in soils<sup>2</sup>.

Soil contamination by copper is connected with high acidity caused by sulphur as well as partly by selenium, arsenic, lead, etc. The low values of pH show there are free acid due to the sulphur oxides. The advanced acidification is specific for the soils within the valley. As a consequence circular forms of heavy metals appear (copper, aluminium, manganese). This is toxic for the plants. Removal of the toxic circular forms of heavy metals from the plants is ameliorative possible and economically but only after cessation the contamination.

Till the 80s of the last century the conducted studies within the valley are mainly on the soils from the agriculture, woods and water that are affected by emissions from the companies. These studies lead to development of effective technologies for mitigation the soil acidity and the toxic effect of the heavy metals in the soil as well as opportunities for rehabilitation of disturbed areas<sup>3</sup>.

#### EXPERIMENTAL

Subject to the research were disturbed and rehabilitated areas, originating from the mining and processing work of Dundee Precious Metals, Chelopech (DPM, one of the mining companies located within the Zlatishko-Pirdopska valley) before and after its privatisation.

The main method used was the systematic-ecological. Results from studies conducted by team from University of Forestry, Sofia, during the period of 1995–2015 (Ref. 4) in connection with projects on rehabilitation of disturbed areas were used for the analysis of the restoration processes within the DPM.

Average soil samples were taken from all of the described embankments. The soil samples were analysed in accordance with methods of Bulgarian State Standard (BSS) or international standards for mechanical composition, general physical and water properties (BSS, p. 2, 1989).

The chemical properties and chemical composition which are of high importance for the biological reclamation, were analysed in accredited laboratories – University of Chemical Technology and Metallurgy (salt ingredients and anions) and National centre of hygiene, medical ecology and nutrition (NCHMEN) (elemental composition), laboratory Aquateratest – Sofia. The main nutrients (N, P, K) were analysed in the laboratory of ecology and environmental conservation in University of Forestry, Sofia. The methods used were in accordance with the BSS and international practice (ISO-11047, BBM 414/CMA:2000, etc.).

Results obtained during the years represented the changes in the rehabilitated areas under the conducted ameliorative measures. The data from 2004 were used as base.

# **RESULTS AND DISCUSSION**

*Concentration of primary nutrients.* Fertility of the reclamation substrates is an important prerequisite for conducting successful biological reclamation. The main preconditions for fertility are the availability of water, humus and primary nutrients in the reclamation substrates – both in soil materials that will be used to cover the rock, as well as in the bulk substrates on which the biological reclamation will be conducted. In order to be fertile soil materials they must have not in excess soluble toxic ions and microelements.

Data from the analysis on humus and nutrients in the embankments, soil materials and clays that are used for reclamation and in soils near the site are presented in Table 1. They are analysed sequentially and in relation to other elements of fertility.

Results for the content of humus show that soils are with very low content of humus. This is due to its natural low concentration in the soils within the valley which is connected with the erosion and the low organic litter as in wood as in grass communities. The slope dump is characterised with a little bit better concentration of humus -2.22-1.87%.

Results show the concentration of total nitrogen is under the required minimum (0.05%). Only the natural soils near the incline gallery 'Nadezhda' (mine 'Nadezhda') are characterised with average concentration of nitrogen -0.312-0.349%. The section in the eastern part of the gallery (SS<sub>6</sub>) is with better results in terms of humus and nitrogen.

The concentration of digestible phosphorus compounds in the substrates is too various. In most of the cases it is low ( $P_2O_5$  is under 10 mg/100 g), in separate cases  $P_2O_5$  is above 13–14 mg/100 g. Sterile bulk materials contain even lower quantities of mobile phosphorus – 2 – 5–6 mg/100 g.

No	Place of soil	Depth (cm)	р	Η	Hu-	N (%)		K <sub>2</sub> O
sec- tion	1 0	-	H <sub>2</sub> O	CaCl <sub>2</sub>	mus (%)		(mg/100g)	(mg/100g)
$\overline{\mathbf{S}}_{1}$	SS <sub>1</sub> soil disposal	average sample 0–20	7.55	6.64	0.40	0.043	10.17	37.59
$S_4$	SS <sub>2</sub> slope dump	average sample 0–20	5.77	5.46	2.22	0.102	7.74	52.78
<b>S</b> <sub>5</sub>	SS <sub>2</sub> slope dump	average sample 0–20	5.92	5.40	1.87	0.312	13.00	34.22
$S_6$	SS <sub>3</sub> steep slope	average sample $0-20$	3.31	3.00	_	0.032	9.66	17.35
<b>S</b> <sub>7</sub>	$SS_4$ natural slope	A 0–17	3.70	3.26	1.10	0.100	3.66	27.96
$S_8$		BC <sub>1</sub> 17–33	3.78	3.21	0.74	0.349	2.47	32.78
S <sub>9</sub>		$BC_{2}^{'}33-50$	3.61	3.18	0.73	0.049	4.67	27.96
S <sub>10</sub>		CD 50–84	3.71	3.24	0.40	0.076	1.05	20.73
<b>S</b> <sup>10</sup> <sub>15</sub>	$SS_6$ entrance of the gallery	0–15	6.81	6.11	0.69	0.124	3.07	75.19
$\mathbf{S}_{16}$		15-38	7.14	6.50	0.33	0.047	2.66	29.88
S <sub>17</sub>		38-50	3.46	3.30	0.27	0.039	4.76	14.94
<b>S</b> <sub>18</sub>	SS <sub>7</sub> E from old flotation plant (FP)	average sample 0–20	3.01	2.85	0.45	0.044	5.91	17.11
S <sub>19</sub>		average sample 0–20	3.48	3.32	-	0.038	3.71	32.29
S <sub>22</sub>	SS <sub>9</sub> S from old FP	average sample 0–20	3.76	3.64	0.78	0.052	3.07	8.44
S <sub>23</sub>	SS <sub>10</sub> over retain- ing wall, mine 'Capital'	average sample a 0–20	4.04	3.90	_	0.037	4.72	26.75
S <sub>24</sub>	SS <sub>11</sub> N from mine 'Capital'	average sample 0–20	3.74	3.67	0.55	0.045	3.02	20.97

Table 1. Concentration of humus and primary nutrients in soil samples within the area of DPM

Concentration of digestible potassium in the substrates is also too various. In most of the cases soil materials and bulk substrates are rich to very rich with digestible potassium. There are a lot of samples with medium concentration. The data show also an average availability of potassium compounds (between 15 and 20 mg/100 g). However, part of the bulk materials are with very low concentration of potassium, especially those of the embankment around the old flotation plant. Because of this the potassium should be included in the combination of fertilisers for ameliorative activities on the reclaimed embankments.

Summarising information on the concentration of primary nutrient in the bulk materials, soil materials and soils within the area of DPM, it can be concluded that

the future rehabilitation should be carried out by bringing mineral fertilisers or other organic matter in accordance with the purpose of the rehabilitation, requirements for reclamation vegetation and the availability of nutrients in these materials.

Analyses on the acidity of soils and the parent materials from the embankments as well as their buffering capacity are particularly important in the reclamation of damaged areas. The level of acidity (pH) depends as on the mobility and absorption of nutrients, as on the mobility of heavy metals and other toxic ions and their saturation in the soil solution. These properties are particularly important for water retention in soil bulk materials which are subject to reclamation.

In terms of the acidity soils from the adjacent area have a heterogeneous character. Most of them are alkaline to slightly acidic, with not high exchange and hydrolytic acidity and therefore there is no need for further introduction of lime materials. However, another part of the natural soils and the majority of the bulk materials are very acidic to acidic, with low sorption capacity and low saturation conversion bases. Such are soils from  $SS_4$ ,  $SS_7$ ,  $SS_9$ ,  $SS_{10}$  and  $SS_{11}$ .

For these soils liming should be with different, but usually pretty high amounts. Taking into consideration the oxidative processes in these substrates are started, most appropriate is lime to be entered on the bulk materials once and after that they can be buried with rock materials with low acidity and covered with clay and soil materials. The biological reclamation of these embankments will be very difficult and lengthy process.

*Concentration of heavy metals.* The nature of the activity of DPM Inc. is connected with high concentration of some heavy metals in soils and excavation materials which are disposed on its territory. The high concentration of heavy metals that is naturally above the maximum allowable concentration (MAC) requires such a reclamation that will minimise their mobility and bring them into a condition that prevents their absorption by plants, animals and humans.

Because of these reasons subject to the analysis on concentration of heavy metals are the total and mobile forms of heavy metals from the soils of adjacent areas, from the soil materials, which will be used for reclamation of damaged areas in the eastern part of the field as well as from the materials from the embankments subject to the first stage of rehabilitation.

Results for the concentration of heavy metals and arsenic are presented in Table 2 (total forms) and Table 3 (mobile forms).

Table	Table 2. Concentration of heavy metals, sulphur and arsenic in soils within the field of Chelopech	/y metals, sulphu	ır and ars	enic in so	oils with	in the fie	ld of Che	lopech						
No	No Place of soil samplin	ing Depth	hд	Cu	Fe	Cr	Mn	ïZ	Co	Zn	Cd	Pb	$\mathbf{As}$	s
sec-	4		$(\tilde{H}_2O)$	(mg/	(%)	(mg/	(mg/	(mg/	(mg/	(mg)	(mg/	(mg/	(mg/	(%)
				Ng)		Ng)	Ng)	Ng)	Ng)	Ng)	Ng)	Ng)	Ng)	
Mini-	. 1.			<0.2	$\heartsuit$	<0.5	$<\!0.1$	<0.2	<0.2	<0.2	<0.2	<0.2	<0.5	$\heartsuit$
mum														
Ś	SS <sub>1</sub> soil dispoal	av. sample	7.55	61.0	2.68	5.8	621.2	10.7	10.2	71.9	0.5	38.6	131.1	0.381
$\mathbf{S}_{4}$	$SS_2$ slope dump	av. sample	5.77	288.8	2.59	27.7	867.9	16.3	8.9	172.8	1.2	91.9	47.1	0.123
Š	$SS_2$ slope dump	av. sample	5.92	229.7	3.06	7.4	973.6	18.2	9.8	176.8	0.9	146.6	220.8	0.382
š	SS <sub>3</sub> steep slope	av. sample	3.31	980.5	3.84	5.4	145.1	7.4	5.1	116.5	0.8	71.9	245.8	2.477
Š		BC <sub>1</sub> 17–33	3.78	191.3	2.73	2.6	40.9	2.7	0.7	35.7	0.4	158.4	373.4	1.484
s		$BC_{2}$ 33–50	3.61	162.2	4.30	1.9	28.1	7.8	1.9	29.3	0.9	181.9	2026.1	2.693
$\mathbf{S}_{10}$		CD 50-84	3.71	725	2.03	3.4	44.5	2.2	0.4	43.5	0.5	167.4	290.3	1.171
$\mathbf{S}_{15}$		0-15		404.6	2.25	3.4	315.6	11.8	6.1	230.4	2.9	52.9	201.6	0.670
$\mathbf{S}_{16}$		15-38	6.81	455.8	3.09	3.9	277.9	7.7	8.0	164.2	1.2	178.9	225.6	1.127
$\mathbf{S}_{17}$		38–50	7.14	246.3	3.94	5.6	83.8	3.3	2.6	122.8	1.1	131.2	329.9	4.155
$\mathbf{S}_{19}$		av. sample	3.01	72.4	2.06	V	58.1	1.8	1.2	52.2	0.2	160.1	114.5	1.249
$\mathbf{S}_{21}$	SS <sub>8</sub> behind the old flor tion plant (FP)	ta- av. sample	3.38	178.8	2.62	17.9	428.5	22.2	7.6	87.5	6.0	57.8	88.3	0.393
$\mathbf{S}_{22}$	SS <sub>9</sub> S of the old flotation av. sample plant (FP)	on av. sample	6.02	788.8	3.32	6.1	242.0	77.0	4.8	183.6	1.5	140.7	448.9	3.362
$\mathbf{S}_{23}$	SS <sub>10</sub> mine 'Capital'	av. sample	3.76	332.9	3.07	10.3	196.6	10.8	5.8	121.8	0.9	39.5	194.3	1.260
$\mathbf{S}_{24}$	SS <sub>11</sub> N from mine 'Capital'	av. sample	4.04	388.1	3.45	5.3	234.8	10.9	6.1	117.6	6.0	76.3	294.6	1.734
The c	The darker colour values show which elements exceed the maximum allowable concentration in accordance with Ordinance No 3/SG. 71, 2008; the other	which elements e	exceed th	e maxim	um allow	vable cor	ncentratio	n in accc	ordance v	vith Ordir	nance Nc	3/SG. 7	1, 2008;	the other

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coloured values show exceeding the background values; not coloured are not contaminated samples.

Tabl	Table 3. Concentration of mobil	obile forms of heavy metals, sulphur and arsenic within the field of Chelopech	vy metals	s, sulphur	and arse	enic with	uin the fie	ld of Ch	elopech					
No	No Place of soil sampling		Hq	Cu	Fe	C	Mn	ïZ	S	Zn	Cd	$^{\mathrm{Pb}}$	As	s
sec-		(cm)	$(H_2O)$	(mg/	(mg/	(mg/	(mg/	(mg/	(mg/	(mg/	(mg/	(mg/	(mg/	(%)
tion			1	kg)	kg)	kg)	kg)	kg)	kg)	kg)	kg)	kg)	kg)	
Ś	SS <sub>1</sub> soil dispoal	av. sample	7.55	7.09	71.3	V	121.16	1.04	0.77	2.84	0.04	3.2	1.4	0.053
$\mathbf{N}_{4}$	SS <sub>2</sub> slope dump	av. sample	5.77	115.40	120.6	V	116.83	1.29	0.57	21.14	0.34	12.2	0.4	0.010
Š	$SS_2$ slope dump	av. sample	5.92	71.84	118.0	V	163.57	1.13	0.78	17.68	0.27	12.8	0.4	0.012
s,	SS <sub>3</sub> steep slope	av. sample	3.31	172.25	696.5	0.3	29.45	0.96	0.98	21.34	0.12	1.9	5.5	0.439
$\mathbf{S}_{1}$	SS <sub>4</sub> natural slope	A 0–17	3.70	73.83	586.3	V	5.00	0.15	0.09	3.23	0.05	1.2	5.9	0.023
Š		BC <sub>1</sub> 17–33	3.78	49.11	603.7	V	3.29	0.41	0.11	2.14	V	2.9	6.3	0.021
s		BC, 33–50	3.61	42.28	929.8	V	3.66	0.24	0.13	3.27	0.05	1.6	26.1	0.033
$\mathbf{S}_{10}$		CD_50–84	3.71	46.46	587.4	V	3.61	0.07	0.11	3.64	0.06	0.3	13.4	0.014
$\mathbf{S}_{15}$		0-15	6.81	102.03	51.8	V	97.22	1.07	0.97	9.64	0.13	12.5	2.7	0.019
	gallery													
$\mathbf{S}_{16}$		15-38	7.14	100.87	66.3	V	48.46	0.74	0.84	29.46	0.27	6.9	3.2	0.128
$\mathbf{S}_{17}$		38-50	3.46	129.07	462.8	0.2	24.78	0.68	1.12	32.16	0.32	0.9	12.4	0.749
$\mathbf{S}_{19}$		av. sample	3.48	17.22	208.3	V	2.02	0.19	0.10	3.24	V	1.9	4.2	0.281
$\mathbf{S}_{21}$	SS <sub>8</sub> behind the old flotation plant (FP)	av. sample	6.02	37.02	57.8	$\vee$	174.77	1.96	1.53	2.91	0.18	3.8	0.5	0.140
$\mathbf{S}_{22}$	SS <sub>9</sub> S of the old flota- tion plant (FP)	av. sample	3.76	91.42	215.5	$\vee$	44.44	0.52	0.80	24.01	0.13	3.5	2.8	0.366
	SS <sub>10</sub> mine 'Capital'	av. sample	4.04	95.71	185.0	$\vee$	41.75	0.86	0.88	18.05	0.12	0.6	1.1	0.318
$\mathbf{S}_{^{24}}$	SS <sub>11</sub> N from mine 'Capital'	av. sample	3.74	109.57	207.5	V	77.72	1.28	1.11	19.86	0.14	0.7	2.0	0.255

Copper and arsenic are with the highest concentration, followed by lead, cadmium, zinc, chromium. This is due to the nature of the mining and metallurgical production which is mainly gold and copper as well as the processed ores are mainly sulphide, polymetalic. In accordance with the data obtained the concentration of mobile forms of heavy metals depends on the acidity of the substrates as well as on the concentration of the total forms of heavy metals. Their mobility is also connected with the concentration of sulphur in the minerals. Concentration of sulphur is above the average concentration. It is 5-10 times more in the soils and in the rock materials from the embankments -5-30-50 times more. The sulphur mobility depends on the acidity of the environment which is connected with the activity of sulphate-reducing bacteria.

The higher concentrations of active sulphur (Table 3) connected with the high acidity of the rock materials in the embankments within the old flotation plant (FP) and near mine 'Capital' determine higher concentration of heavy metals – especially copper.

The mobile copper and rarely zinc, cadmium and lead excess the maximum allowable concentration for total forms in a lot of the soil samples. Their concentration in the substrates subjected to rehabilitation is due to their concentration in the ore materials and the technique of their extraction. Despite the higher pH of the bulk materials from SS<sub>6</sub> 52–54% from the copper is in mobile forms.

Concentration of total copper is lower in the bulk materials from  $SS_7$ ,  $SS_8$  and  $SS_9$  as well as in the substrates from  $SS_{10}$ . They are with high acidity, i.e. oxidation processes occurring there a long time and average 23–28% of the total copper is in mobile forms.

High concentration of copper – total and mobile as well as concentration of sulphur in the rock materials leads to the conclusion that during excavation of the rock materials from the gallery should not be allowed excitation of rapid oxidation processes, i.e. during the filling embankments with parent materials they should be limed and compacted.

Results from the analysis from 2015 on the 'West' show variation compared to the analysis from 2004. The soil solution is highly acidic, it had not changed over the years.

In terms of heavy metals and metalloids arsenic is with the highest concentration. It exceeds the maximum allowable concentration (up to 30 times in 2005 and 4.3 times in 2015) and the interventional level (with 5.66 times in 2005 and 1.5 times in 2015). Concentration of cadmium, copper, lead and zinc is overstated and exceeds the precautionary values. While in 2005 concentration of copper in soil samples exceeded the maximum allowable concentration, there is not such an excess in 2015.

These concentrations and the chemical status of the rehabilitated area are due not only to the low content of organic matter because of the low organic litter, not even to bad rehabilitation. This due to the high quantities of chemicals – waste from underground mining, that enter into reaction with ground water in the embankment, leach at its base and acidify and contaminate water and soil with heavy metals. That is way grasses on the embankment develop progressively, but trees are with good growing during the first years, but with deepening of their root systems they wither.

Soils from the rehabilitated area from the western part of mine 'Capital' are slightly to highly acidic. The pH values in water and  $CaCl_2$  are too close, which means that large part of the acid ions are mobile. Content of humus is very low, resulting in small to medium soil reserves with nitrogen, very weak with potassium and phosphorus compounds.

Soil materials analysed for heavy metals show an excess of the maximum allowable concentration for copper with 1.6 times and for arsenic – with 3 to 27 times. Arsenic also exceeds the interventional level with 1.6 to 8.9 times. Exceeding the precautionary values is established for all heavy metals and metalloids, with the exception of chromium, in which even the background concentration is not exceeded. We should note that north and in proximity to the embankment earthworks are held which largely affects the soil formation processes – through toxic dust, causing high acidity. Vegetation on the rehabilitated area is represented by birch (*Betula pendula*), acacia (*Robinia pseudoacacia*), beech (*Fagus sylvatica*), pine (*Pinus sylvestris*) and various grass.

Soils from the rehabilitated area in the eastern part of mine 'Capital' are with slightly to very acidic soil reaction, which due to the high concentration of sulphur. Content of humus is very low, as well as the soil reserves of nitrogen, phosphorus and potassium compounds.

Concentration of heavy metals shows excess of the maximum allowable concentration and the interventional level by arsenic, respectively with 35.3 and 11.8 times. Concentration of copper also exceeds the maximum allowable concentration and the interventional level , respectively with 2.5 and 1.26 times. There is an excess of the background concentrations and the precautionary values by of the all heavy metals except nickel and chromium.

Soils from the inclined gallery 'Nadezhda' (SS<sub>6</sub> with S<sub>15</sub>, S<sub>16</sub> and S<sub>17</sub> from 2004) are with medium to heavy sandy-clay mechanical structure, strongly acidic soil reaction, low sorption capacity and low rate of exchange base saturation. Concentration of humus is very low to medium. Soil reserves with nitrogen are low to medium, phosphorus compounds are with low concentration, potassium compounds are with medium concentration.

While in previous analysis was established exceeding of the maximum allowable concentration by arsenic with up to 50 times and the interventional level with 16.9 times, the results from 2015 show an excess over the maximum allowable concentration with 6.76 times and over the interventional level with 2.25 times. Also in previous analysis was established exceeding of the maximum allowable concentration by copper with 2.56 times and exceeding the interventional level – 1.28 times, but in 2015 the concentration of copper is under the maximum allowable concentration.

This shows that for the period of ten years part of the heavy metals are leached in depth with the high reaction of soil solution. Meanwhile pH in the soil coating reduces from approximately 6 to 4.13 and the parent material keeps its high acidity, i.e. the sulphate sulphur is in high concentration and continues to nourish soil solution with sulphte ions.

That is way in terms of vegetation only tree species resistant to high acidity have kept their species composition. There is pine (*Pinus sylvestris*) and black pine (*Pinus nigra*), oak and grass cover of grasses and mosses on the rehabilitated area of inclined gallery 'Nadezhda'.

## CONCLUSIONS

Based on the presented data and discussions on results from researches on the embankments formed by the mining activities within the field of 'Chelopech' before their rehabilitation in 2004–2005 and the analyses of soil samples taken in 2015 from the reclaimed areas the following can be concluded:

-Acidity of the rock materials decreases slightly, but increases in the surface soil materials at the expense of capillary rise of sulphate ions through water from the rock substrates. Acidity of the embankment, mine 'West' is characterised with highly acidic materials buried in the embankment.

- Parent materials remain poor in organic matter, total nitrogen, digestible forms of phosphorus and potassium, despite the conducted fertilisation during the first three years of rehabilitation (requirement for cultivating cares).

- Despite the low fertility of the rehabilitated areas, grass coverage imported during the biological reclamation is developing relatively well, thereby preventing the embankments from erosion and enrich their surface layers with organic matter.

– The specific granule composition, impaired water regime, availability and mobility of the above mentioned chemical elements in the waste from mining activities represent the greatest difficulty in reclamation of the embankments within the DPM Inc. For these reasons, specific ameliorative measures should be conducted during the rehabilitation of lands damaged by the activity of DPM and its previous owners. The most important of the ameliorative measures is the introduction of substances that reduce the acidity of substrates – limestone materials, increased rates of phosphorus fertilisers, etc. as well as substances enhancing soil fertility through nitrogen and potassium fertilisers.

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Ecology

# STUDY CONCERNING THE EXCESSIVE HUMIDITY ON THE WALLS OF A RESIDENTIAL BUILDING AND THE HEALTHY IMPACT

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Abstract. Nowadays many buildings and historical monuments are affected by the phenomenon of capillary ascension of the water through foundation and walls, leading to the appearance of dampness. The presence of excessive humidity in a brick wall leads to the plaster and wall degradation, to an increased heat loss in the affected area and also causes the appearance of mould (bacteria and fungus) on the interior plaster, which leads to a toxic microclimate inside for those who live in the household. This paper represents a study regarding the effects of moisture on the walls of a household and on the human health. It also presents methods which can be used to remove humidity from the walls. Excessive moisture has a negative effect on the indoor climate, leading to the appearance of microorganisms, such as fungi, bacteria, which affect the health of the inhabitants of the building. In areas affected by humidity and where heat losses occur there is a degradation of the materials that the brickwork is made of. House rehabilitation interventions made in the past will be corrected by appropriate measures. Intervention methods will be based mainly on ventilation walls. The materials used will respect the datasheet. The conclusions indicated that the higher levels of humidity were due to poor maintenance of the building, but also to inappropriate interventions, which instead of reducing moisture problems led to their amplification.

Keywords: capillary ascension, excessive humidity, brick wall.

## AIMS AND BACKGROUND

Cultural goods, historical monuments, modern buildings are made from materials that are more or less porous, more or less chemically active, which constitute the support for the water migration in the form of solutions of salts. The water migration takes place from a higher energetic level to a lower energetic level<sup>1</sup>.

The excessive humidity has a negative effect on the interior microclimate, leading to the appearance of microorganisms, such as: moulds, bacteria etc.<sup>1,2</sup>

Moulds that spread through the air have a series of adverse effects on health, entering into the human body by inhalation, but also through skin contact. When

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mould spores are present in a large number in the breathable air, they may cause allergic reactions, asthma, infections and other diseases<sup>3–7</sup>.

Mycotoxins released by moulds and inhaled by people affect the liver, inhibit the protein synthesis and cause haemorrhages. The cellar mould (*Penicilium galucum*) appears in any enclosed space that is poorly ventilated and moist. Sticking to the walls, this mould gives a specific, heavy/foul smell. The fungus is toxic and allergenic<sup>1,2</sup>. One of the mould species that can be mentioned is *Aspergillus fumigatus*.

Bacteria may have negative effects on the state of health, causing inflammation of the respiratory tract, but also skin infections.

#### EXPERIMENTAL

The object of the case study is a building for human habitation situated in the Cut village, Alba county, which was built at the beginning of the 18th century. The building was abandoned some decades ago and at the beginning of the year 2000 began its renovation. Due to the lack of maintenance, the construction elements deteriorated considerably. The building consists of a basement, a ground floor and an attic.

Within the project, we performed measurements in different points, up to the height of 100 cm, in the basement and the ground floor, given that the humidity signs in the masonry were clearly visible. It was considered that no measurements were necessary in the attic, since it was rebuilt with new materials.

The measurements for the walls were performed with a Testo 600-1 device, and the interior atmospheric humidity was determined with a hygrometer<sup>8</sup>.

#### **RESULTS AND DISCUSSION**

*Basement*. The atmospheric humidity of the basement is 55%, being thus optimal for its use as a cellar.

The walls have undergone operations consisting in the application of a waterproofing membrane on the exterior side and the execution of ventilation openings with PVC tubes and the plaster was removed on the exterior walls. The high humidity in the walls was caused by meteoric water, which was flowing directly on the wall, infiltrating into the masonry.

The effects of humidity were transferred especially on the bricks, causing their deterioration (Fig 1). The humidity was measured in three points, as can be noticed in Table 1.

H (cm)	W1 (%)	W2 (%)	W3 (%)	W atm. (%)	Temperature (°C)
20	10.3	12.8	9.3	55	10
40	12.5	10.2	7.2		
60	14.2	9.4	14.1		
80	14.4	2.3	14.9		
100	13.1	5.2	6.4		

Table 1. Humidity level in the basement

H-height; W1, W2, W3 - the humidity measured in the 3 points; W atm. - atmospheric humidity.

In the case of the ground floor, the ascension of the humidity reaches in some places a height of 2 m. This humidity is present due to the fact that the roof of the building is affected by considerable decay and thus the waters from precipitation came directly on the masonry, infiltrating into the structure. Another cause is the flow of water on the outside of the building, caused by the difference of level between it and the ground.



Fig. 1. Presence of the humidity: the basement, corridor, smoke room, living room

One of the causes of excessive humidity is the floor, which is made of concrete and floor tiles. These materials, being impermeable, did not allow the water to evaporate, determining a capillary ascension of the water from the foundation ground towards the higher zones in the masonry.

The ventilation openings made by the owner. Due to the fact that the walls are quite thick (80 cm), they got clogged in time, thus becoming ineffective.

Another cause of the humidity in the walls is the fact that the level of the ground behind the house reached the height of the windows.

The humidity leads to the appearance of spots and the fall of the plaster. The humidity levels were measured in two points, presented in Table 2.

Sampling point – ground floor – corridor				
Н	W1	W2	W atm.	temperature
(cm)	(%)	(%)	(%)	(°C)
20	5.5	3.4	49	12.5
40	6.2	2.5		
60	6.2	5.9		
80	5.9	6.4		
100	5.8	6.3		
cma	3–5	3–5		

Table 2. Humidity in the corridor

W1, W2 - the humidity measured in the two points; W atm. - atmospheric humidity.

In the case of smoke room, the causes which lead to a high humidity level are the same as in the case of the corridor. The lack of ventilation also determined the persistence of humidity in the walls. Dampness can cause failures of the electrical installation.

Although the humidity in the wall is high, the atmospheric humidity in this room is normal (Table 3).

Table 3. Humidity in the smoke room

Ground floor – smoke room				
$H(\mathrm{cm})$	W1 (%)	W atm. (%)	temperature (°C)	
20	6.3	52	12.6	
40	6.8			
60	6.1			
80	3.1			
100	6.2			

*H* – height; W1 – the moisture measured in 1 point; W atm. – atmospheric humidity.

*Cultural centre*. This phenomenon of excessive humidity was caused both by the capillary ascension and the horizontal migration of the water from the foundation ground towards the wall of the room.

The meteoric waters also lead to an increase of the humidity in the walls.

In this room were performed wall repair works. Ventilation openings were made here also, but as in the case of the corridor, they proved to be ineffective. The humidity levels were measured in two points (Table 4).

Ground floor- cultural centre				
H (cm)	W1 (%)	W2 (%)	W atm. (%)	temperature (°C)
20	6.2	5.9	51	11.9
40	5.9	6.4		
60	2.7	5.9		
80	6.4	6.2		
100	2.9	5.8		

 Table 4. Humidity in the cultural centre

H-height; W1, W2 - the humidity in two point; W atm. - atmospheric humidity.

*Living room.* Dampness appeared due to meteoric rains, which infiltrated into the masonry following the deterioration of the roof. The relative humidity in this room is above the normal limits, reaching as much as 65% (Table 5). The effects of humidity on the masonry can be noticed in Image 5, but they tend to diminish, as the wall is beginning to dry.

Table 5. Humidity in the living room

Ground floor-living room					
$H(\mathrm{cm})$	W1 (%)	W atm. (%)	temperature °C		
20	5.5	65	18.3		
40	2.4				
60	2.5				
80	3.1				
100	5.8				

H-height; W1-the moisture measured in 1 point; W atm. - atmospheric humidity.

*Kitchen.* The highest level of atmospheric humidity (68%) was measured in this room. Here, the effects of humidity in the wall are visible up to a height of over 2 m. This is caused by the floor, which is made of concrete and floor tiles, but also by the facing with wall tiles.

## CONCLUSIONS

As a result of the evaluation, the following measures have been proposed:

- The floors made from floor tiles and concrete in the corridor will be removed and they will be replaced with breathable floors;

- In the kitchen, where the atmospheric humidity is very high, the floor and the wall tiles will be removed. The floor will be remade, the wall tiles will be placed only where it is necessary and the rest of the wall will be plastered with breathable mortar;

- On the interior/exterior walls ventilation ducts will be made, and airing openings will be performed at 1.0 m, ventilation grilles being placed on the exterior side of the wall;

- Given the fact that the ground on which this building for human habitation is placed is sloping, a drainage canal will be made on the rear frontage for removing rainwater;

- Rainwater will be carried from the gutters to the sewerage system through PVC tubes;

- In all the rooms in which there are obvious signs of humidity, the plaster will be removed with 50 cm above these visible signs of humidity. The plaster will be reapplied with breathable mortar;

At the lateral side at the exit from the house on the exterior waterproof pavement will be removed and replaced with a permeable pavement. The foundation will be protected vertically with a Fondaline membrane.

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# TRADITIONAL FOOD AS A TOURIST ATTRACTION IN ECOLOGICAL FARMS FROM ROMANIA

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**Abstract**. This article presents the relationship between rural tourism and culinary heritage. To investigate the tourists perception on traditional foods consumed in ecological farms, a number of 143 tourists were interviewed in 2016 regarding the importance of traditional food in the assessment of a successful holiday, the percentage of the average budget per holiday spent on traditional dishes and the type of meal services preferred. Statistical correlations were made between the various answers to questions and the results were presented in graphs and tables. The importance of the local traditional food in the assessment of a successful holiday is appreciated by 58.8% of the respondents ranking between 20 and 50%. The preferences related to the choice of the number of meals served show that half board and breakfast are preferred by 69.2% of the total tourists surveyed. The data obtained highlight that 40.6% of tourists spend more than 50% of holiday budget on food, 39.9% spend between 20 and 50% and 19.5% allocate less than 20% of expenditures on traditional food. These case study results are based on the interviews, being followed by a discussion and conclusions offering further research directions.

Keywords: traditional food, ecological farms, tourism attraction, Romania.

## AIMS AND BACKGROUND

Tourism constitutes one of the most important economic sectors on a global scale<sup>1</sup>. Many countries suffering of recessions and economic turmoil can benefit from tourism to bring foreign capital in the country to stimulate growth<sup>2</sup>.

Rural tourism is a type of tourism that becomes integrated with rural culture, natural environment and agriculture<sup>3</sup>.

For human beings, food is a critical contributor to physical well being, a major source of pleasure, worry and stress, a major occupant of waking time and, across the world, the single greatest category of expenditures<sup>4</sup>. Traditional foods as part of the cultural identity of a region are considered as an important component of ecological tourism.

<sup>\*</sup> For correspondence.

As identity marker of a region and as a means of promoting farm products, gastronomy meets the specific needs of consumers, local producers and other actors in rural tourism<sup>5</sup>.

Fischler<sup>6</sup> has developed the concept of a 'back-to-nature myth' as a countertendency to urbanisation: city dweller escapes from the daily routine to find solace in so-called 'traditional' food. Combined environmental and social concerns about the adverse impacts of large scale factory farms and industrialised agriculture have facilitated movement from intensive, producing agriculture to alternative, more sustainable agricultural forms, including organic farming<sup>7</sup>. Agritourism has been given increased attention with respect to its conservation potential and is clearly growing and many studies forecast further growth in supply and demand<sup>8</sup>.

Ecotourism criteria used in the literature may be summarised as follows:

- (1) Minimum physical and social impacts on the visited area;
- (2) Ecological education of the tourist at the natural site;
- (3) Notable economic participation of local residents<sup>9</sup>.

An agritourism system model identifies three primary stakeholders (agritourism providers, destination marketing organisations, and agritourists) and asserts that the conservation of traditional family farms can contribute to the conservation of communities, landscapes, and ecosystems<sup>10</sup>.

Organic farming emerged in the period 1920 - 1950 as a reaction to the increased use of chemical fertilisers, pesticides, and herbicides, and to their perceived impacts on the environment and food<sup>11</sup>. Farm tourism in particular has received much attention and nowadays it is considered an established means of agricultural diversification<sup>12</sup>.

Organic farms are also seen as an opportunity not only for the protection of natural systems but also for assisting small farmers<sup>13</sup>. Farm tourism offers economic opportunities, and hopes have been expressed since the late 1970's that farm tourism could support local economies and contribute to the preservation of landscapes and cultural heritage in the countryside<sup>12</sup>. Tourism in ecological farms is seen to meet the needs of visitors who seek traditional rural hospitality, nature, and cultural experiences, as well as thematic holidays such as health-oriented and outdoor activities, while helping farmers to maintain agricultural viability and economic diversity<sup>14</sup>. Farm tourism has been conceptualised as an 'alternative farm enterprise' comprising one of several potential forms of farm business diversification<sup>15</sup>. It is often perceived to be intrinsically sustainable since it generally hosts a small number of tourists and does not require extensive infrastructure<sup>16</sup>. Farm tourism is part of rural tourism, and the location of the accommodation in a part-time or full-time farm is its distinguishing criterion<sup>17</sup>.

Despite the importance of food as an input in the tourism sector, food continues to be given very little attention in the literature<sup>18</sup>. Gastronomy is a fundamental

component of tourist traveling: eating is an integral part of holidays and rural tourism can be a part of the re-appropriation of history in terms of eating habits<sup>5</sup>.

The present paper brings into discussion issues on tourists' interest in traditional foods from ecological farms in Romania.

#### EXPERIMENTAL

A total of 143 people were surveyed regarding the importance of traditional food in the assessment of a successful holiday in ecological farms, the percentage of the average budget per holiday spent on traditional dishes and the type of meal services preferred. Statistical correlations were made between various answers to questions and the results were expressed in graphs and tables.

To make statistical analysis, specialised software called IBM SPSS Statistics, version 23 was used. The significance coefficient was  $\alpha = 0.05$ .

## **RESULTS AND DISCUSSION**

The importance of traditional food in assessment of a successful holiday was firstly determined. The summarisation of results can be seen in Fig. 1.

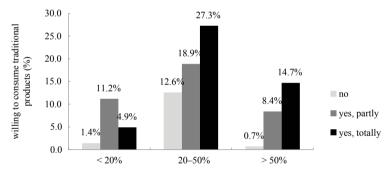


Fig. 1. Importance of the local traditional food in the assessment of a successful holiday

The data from Fig. 1 indicate that for 58.8% of the respondents traditional culinary importance rank between 20 and 50%, whereas 23.8% of those surveyed give greater importance than 50% to traditional food preparations, and the rest of them, 17.5% give less importance than 20%, respectively. The intention of consuming only traditional gourmet cuisine is expressed by 46.7% of respondents, a percentage of 38.5% want to eat partially this kind of food and only 14.8% are not interested to consume such products.

We have determined by statistical analysis if there is a correlation between the importance of the local traditional food in the assessment of a successful holiday and willing to consume traditional products. The Pearson coefficient of the chi square

test of the association is  $\chi^2 = 15.269$ . Since this value is associated probability  $p < 0.004 < \alpha = 0.05$ , we accept that there is an association between the importance of traditional food and demand to consume traditional foods.

Secondly we tried to determine the percentage of holiday budget spent on eating traditional food. The data can be seen in Fig. 2.

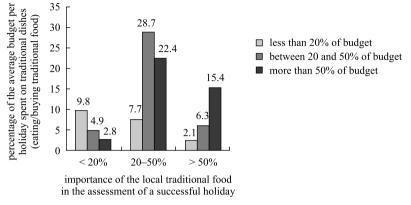


Fig. 2. Percentage of the average budget per holiday spent on traditional dishes

The data presented in Fig. 2 show that 40.6% of tourists spend more than 50% of traditional holiday budget on food, 39.9% spend between 20 and 50% and 19.5% allocate less than 20% of expenditure to traditional food.

Statistical analysis aimed at establishing correlations between the percentage of budget spent on eating traditional foods and the importance given by tourists to traditional food when assessing a holiday. Important variables standing for traditional holiday foods and percentage of budget allocated are considered as ordinal (below 20% < between 20–50% < 50%). The values of the Kendall correlation coefficients  $\tau b$ ,  $\gamma$  (Gamma) and the Somer correlation coefficient *d* for ordinal variables and probabilities associated with these values were calculated. If these coefficients are associated with probabilities of p < 0.05, one should admit that there is a correlation between the importance of traditional food and the percentage of budget spent on eating traditional foods.

The coefficients-associated probabilities are  $p < 0.001 < \alpha = 0.05$ , thus suggesting that the variables are correlated. As for tourists food preferences, we tried to find out what type of meal services on vacation rather small, breakfast, half board or full board is preferred. Responses are summarised in Fig. 3.

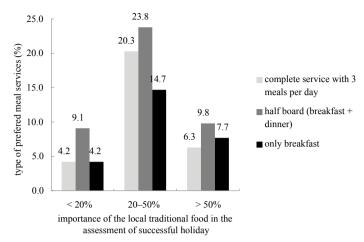


Fig. 3. Type of preferred meal services

The data presented in Fig. 3 show that 42.7% of tourists prefer as meal type half board (breakfast + dinner), whereas 30.8% prefer full service with three meals per day, and 26.5% prefer to serve only breakfast in accommodation.

Statistical analysis results show that there is no correlation between food against traditional mass and type of services chosen.

#### CONCLUSIONS

Traditional foods in organic farms are given special attention by tourists. The importance of local traditional food in the assessment of a successful holiday is appreciated by 58.8% of the respondents ranking between 20 and 50%. Preferences related to the choice of the number of meals served in accommodation shows that half board and breakfast are preferred by 69.2% of the total tourists surveyed. Half-board and breakfast meals only feature amateur tourists hiking in the area who are interested in visiting the area, attractions provided by and serving lunch in the places where they travel to.

The data obtained show that 40.6% of tourists spend more than 50% of holiday budget on food, 39.9% spend between 20 and 50% and 19.5% allocate less than 20% of expenditure on traditional food. In a typical holiday food represents approximately one-third of all tourist expenditures<sup>18</sup>. The difference is due to organic foods which are more expensive than those obtained from classical intensive farming.

This article presents the relationship between rural tourism and culinary heritage. Further research is needed to answer the question: *Why do local cuisine and so-called 'traditional' products arouse such interest?* Also, taking into consideration tourists' interest in traditional foods, we intend to design an on line site to display a complete inventory of the culinary heritage of the Romanian provinces.

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# UTILISATION POTENTIALS OF MAQUIS SHRUBLAND IN SUSTAINABLE LANDSCAPE DESIGN

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**Abstract**. In Turkey, about 7.5 million ha of maquis shrubland are basically found in the Mediterranean region. The characteristics of the ecological conditions in the region help the maquis components to continue their existence even under the least maintenance circumstances and hard conditions. Maquis areas are, nevertheless, being steadily destroyed in Turkey due to construction and urban design as in the other Mediterranean countries. This study was planned in three stages. Firstly, the protected maquis areas located in the parks of Antalya City are to be assessed. Secondly, a 3D Project presentation on transformation of present natural maquis areas into cultural landscaping with simple interferences is to be prepared. Finally, the reasons of maquis protection in the coastal regions and the utilisation ways within the light of sustainability term are to be explained by using examples. The aim of the study is to determine the advantages of maquis areas which can be used in the suitable landscaping in their nature. It is also an idea that this study may help to increase the utilisation of these maquis areas in cultural landscaping during planning studies in the future.

Keywords: maquis areas, sustainability, planning, natural landscaping, cultural landscapes.

## AIMS AND BACKGROUND

The French word 'Maquis' is used as 'Macchia' in Italian<sup>1</sup> and accepted as 'Maki' in Turkish. Called Maquis or Macchia in Mediterranean Basin, this vegetation type is called under different names in different countries<sup>2,3</sup>.

So what does maquis actually mean? The bushes with leathery leaves, dense form and up to 5 meter high mostly woody and some herbaceous bushes in the Mediterranean basin<sup>4,5</sup>. Maquis is secondary vegetation formed with destruction of permanent evergreen forest cover<sup>6</sup>. Due to cut, overgrasing and continuous use

<sup>\*</sup> For correspondence.

of maquis, first garriques cover and later step vegetation appears<sup>7</sup>. It is seen in the Fig. 1 issued by Polunin and Huxley<sup>8</sup> what essentials of Mediterranean vegetation are with the phases experienced with different factors.

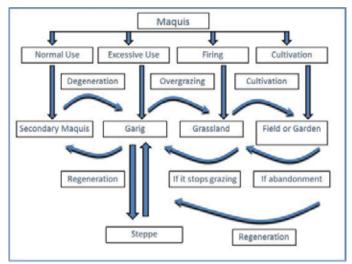


Fig. 1. Phases experienced by maquis with different factors8

Maquis elements are not very choosy in terms of soil requirement. They can grow on any soil from acidic to alkaline<sup>9</sup>. Maquis spread over up 1000 m above sea level in respect to topography and climate and over arid regions to semi-arid regions. Covering approximately 7.5 million ha area in Turkey, maquis is preliminary common in Mediterranean, Aegean and Marmara Regions<sup>10,11</sup>. Distribution of maquis in Turkey is given Fig. 2.



Fig. 2. Natural distribution of maquis of regional base in Turkey<sup>11</sup>

The regions where maquis cover intense areas are the Tourism centres of Turkey, which has been receiving the highest migration. Therefore maquis, in the places where highest amount of constructions areas take places, has been very quickly destroyed in Turkey in the last two decades. With each passing day is expanding in the city limits an uncontrolled manner in the regions of the maquis and these regions are being destroyed by staying in the city.

Forest Code numbered 6831 (Official Newspaper numbered 8.9.1956/9402) left maquis areas outside of forest lands. Maquis areas do not have the protection of forest lands and therefore all type of pressure is seen on maquis areas within the cities. While the studies are carried in the last two decades to increase use of natural vegetation in landscape areas of developed countries, in Turkey maquis elements that are natural vegetation of Mediterranean is defined 'degraded forest'. Thereby, they have been subjected to a treatment they do not deserve and they have been removed and replaced by monoculture foresting or touristic development<sup>12,13</sup>.

It is known that maquis is generally densely consumed by grass eating animals and is a valuable food for animals<sup>10</sup>. Moreover, maquis has great significance in soil and water preservation, treatment of degraded areas, fuel and construction material production and forming habitat for wild animals<sup>13,14</sup>. This use of maquis has in the attracted many scholars and many researchers have been conducted last 10–15 years<sup>3,15–18</sup>.

There are some researchers<sup>19–22</sup> present for production of some plants that are maquis elements and use of them in cultural landscape. However, there is no scientific study on how to use or protect maquis as a whole within a city; but there is existing landscape projects applied in this area. Most beautiful two examples of these are in Ataturk Culture Park and Akdeniz City Park within the borders of Antalya City Centre. While the black goats eat the young leaves and fresh shoots of maquis, thereafter, they unconsciously give various shapes to the plants. The shapes are nearly as successful as artificially shaped ones (Fig. 3). Tavsanoglu and Cosgun have conducted a research on effect of black goats grazing on developmental forms of maquis species<sup>23</sup>. However, the use of trimmed maquis in landscape applications has not been done yet in literature.

In the study, it would be liked to determine the advantages of maquis areas which could be used in the suitable landscaping in their nature especially in the coastal areas. It is also an idea that the study may assist to increase the utilisation of the maquis areas in cultural landscaping during planning studies in the future.



Fig. 3. Juniperus oxycedrus subsp. oxycedrus plant shaped by black goats<sup>24</sup>

#### EXPERIMENTAL

The materials of this study are Antalya City Centre maquis areas. Maquis include mostly woody and some herbaceous plants such as: *Quercus coccifera* L., *Sytrax officinalis* L., *Myrtus communis* L., *Arbutus unedo* L., *Arbutus andrachne* L., *Phillyrea latifolia* L., *Olea europaea* L. var. *sylvestris*, *Pistacia terebinthus* L., *Pistacia lentiscus* L., *Rhus coriaria* L., *Cotinus coggygria* L., *Laurus nobilis* L., *Ceratonia siliqua* L., *Anagyris foetida* L., *Erica arborea* L., *Erica verticillata* L., *Cersis siliquastrum* L., *Jasminum fruticans* L., *Nerium oleander* L., *Cistus creticus* L., *Cistus salviifolius* L, *Cistus parviflorus* L., *Calicotome spinosa* L., *Sarcopolerium spinosum* (1.) spach, *Genista acanthoclada* L., *Lavandula cariensis* (L.) Mill., *Rosmarinus officinalis* L.<sup>1,25</sup>

The city of Antalya, one of the cities where maquis is most densely seen in Turkey, has been chosen. The province of Antalya is located in the Southwest of Turkey on 29°20′–32°35′ East longitudes and 36°06′–37°02′ North latitudes. The province is the 5th biggest province of Turkey both in terms of its population (2 222 562 people) and surface area (20 723 km<sup>2</sup>) with 620 km sea cost. Another reason of choosing the province of Antalya is that in the province the maquis has been heavily damaged due to tourism and constructions. It was identified in the study of Mansuroglu et al. that total maquis area within the zoning area of the city is 42 962 ha and covers space around 5% (Refs 26 and 27). As maquis is intertwined with other biotopes in some regions, some researchers have separately mentioned

these areas. Accordingly; it was identified that the zoning borders of city is: agriculture areas + maquis + forest 5.2% (2821 ha), maquis + forest 6.8% (3726 ha), rocks + maquis 0.1% (25 ha), maquis + forest + garriques 1.3% (697 ha), valley + maquis + forest + garriques 0.3% (146 ha), maquis + settlement areas 1.1% (61 ha).

The study was conducted at Ataturk Culture Park, Akdeniz City Park and Akdeniz University Campus within zoning area borders of city of Antalya (Fig. 4). Gokturk identified that maquis vegetation type is particularly seen at Akdeniz University Campus and locally at the West, North and East sections of the area<sup>24</sup>. Mansuroglu et al. stated in their study that Akdeniz University is one of the 3 example areas where maquis biotope is seen most beautifully within zoning area borders of city of Antalya<sup>27</sup>.



**Fig. 4**. Ataturk Culture Park, Akdeniz University Campus and Akdeniz City Park from the Antalya satellite view<sup>28</sup>

The study was successively conducted in three phases. In the first phase, following general literature related to the researches on maquis areas in general in the city of Antalya have been investigated and summarised in the introduction section of the study. In the second phase, two important city parks, Atatürk Culture Park and Akdeniz City Park where maquis areas have been protected included within cultural landscape have been documented with photographs and its harmony with the park in general has been associated in terms of landscape design. In the third and the last phase of the study, the situation of the maquis areas in Akdeniz University Campus, where maquis biotope is most densely seen has been identified. It has been seen that the maquis areas seriously decreased especially in the last 10 years due to indicated reason above Therefore, prospective precautions have been brought to protect maquis areas within the campus area. Finally, the trimming of the maquis by making small interventions through remarkable custom concept design with 3D Max program was carried out.

## **RESULTS AND DISCUSSION**

In the first phase of the study, it was determined that the most beautiful examples of maquis biotopes in the city centre of Antalya were seen in Foundation Farm which is the 1st level Natural Archeological Site, north of Yamansaz Wetland and the Campus Area of Akdeniz University. It was seen in the construction plan that maquis and garriques biotopes were divided among houses, workplaces and other urban uses. This clearly showed us that maquis in Antalya are vitally important to protect and use them in sustainable ways.

In the second phase of the study, protected maquis areas in the two important city parks were analysed.

*Ataturk Culture Park*. Ataturk Culture Park is the biggest one in the city. It is within the area of Antalya Culture Centre and Glass Pyramid Sabanci Congress and Fair Center. The construction of the center in 1992, 1st phase, 50 ha of the park which is 80 ha was finished and opened to public use in 1997. Although a bigger part of the park was established on maquis area, the current maquis area in the park is today around 2 ha. The maquis area is mainly protected in the South of Atatürk Culture Centre and used as a natural landscape. The maquis at the right and left side of the King Road were naturally protected within the park (Fig. 5).



Fig. 5. Satellite picture of the maquis area in Ataturk Culture Park<sup>28</sup>

As the maquis area is very dense, free grass area was left at the entrance in order to decrease its effect (Fig. 6). This arrangement was increased the effect of the maquis. 50 - 150 cm gap was left in the front side of the maquis area and arranged as grass area and framed with curb (Fig. 7). Walk paths were left between

the maquis areas and included into the landscape in harmony with the whole park. Cliff stones in the natural structure have also increased the visual quality of the maquis. Ataturk Culture Park is a good example of using maquis area protected within the landscape.



Fig. 6. Maquis area arranged in Ataturk Culture Park



Fig. 7. Maquis area arranged in Ataturk Culture Park

*Akdeniz City Park*. Completed in 2013, having 9 ha area and being the third biggest park of the city, Akdeniz City Park is one of the most beautiful examples where the maquis area was protected and displayed within the landscape (Fig. 8).



Fig. 8. Situation of the maquis in construction of Akdeniz City Park<sup>29</sup>

In some sections of the park, the front side of the maquis area is arranged as grass area. *Bougainvillea glabra* L. was planted right next to the cliff stones within the maquis and created a contrast with the green texture of the maquis. *Prunus cerasifera* L. and *Lagerstroemia indica* (L.) Pers. was planted in the grass area in front of the maquis and contributed colour to intense green appearance of the maquis (Fig. 9). In some sections of the park, the front side of the maquis area is arranged as grass area and right in front of the curb, creeping shrubs were planted (Fig. 10). The arrangement with planting *Berberis thunbergii* L. and *Juniperus horizontalis* L. right in front of the maquis made this arrangement very impressive.



Fig. 9. Maquis area arranged within Akdeniz City Park



Fig. 10. Maquis area arranged within Akdeniz City Park

Approximately one fourth of the Akdeniz City Park includes maquis area. Thereby, this park is a great example for other parks. Moreover, the maquis area was not left random within the park but flowers or dark red color plants having a contrast with the maquis were used. These plants also increased the visuality of the maquis.

*Akdeniz University Campus*. Campus located in the west of the city. Akdeniz University Campus has in total 339.77 ha (Ref. 30). Akdeniz University Campus is identified to be the place where the maquis is most densely seen according to the researchers and our estimations<sup>26,31,32</sup>. Unal and Gokceoglu stated in their study that the dominant vegetation of the campus was *Quercus coccifera* L. due to fast growing of Akdeniz University in both number and constructions the last 10 years also had a negative effect on maquis area in total<sup>33</sup>. According to 2015 data, structural area of the campus is 67.72 ha (Ref. 30). This makes one out of twenty percent of the area. The maquis area in the Akdeniz University campus has been steadily destroyed not only because of the structural areas but also due to landscape arrangements. Following completion of the buildings in the campus, the maquis in the gardens shall be removed and total cultural landscape will replace it (Fig. 11).



Fig. 11. South Garden of Faculty of Fine Arts at the Akdeniz University, Antalya - Turkey

A very urgent solution is needed to protect the maquis area at Akdeniz University Campus. Undergraduate students, A. Mirzatürkmen and A. Nugay, from Interior Architecture and Environmental Design in the Department of Faculty of Fine Arts at the Akdeniz University prepared a project to create awareness for this issue under the supervision of the Dr. Z. K. Elinc (Figs 12 and 13).



Fig. 12. Shaping the maquis area by the trimming project (designed by A. Mirzaturkmen under the supervision of Dr. Z. K. Elinc in 2015)



**Fig. 13**. Shaping the maquis area by the trimming project (designed by A. Nugay under the supervision of Dr. Z. K. Elinc in 2015)

The starting point of both students is the shapes of the maquis after grasing of the black goats in nature (Fig. 3). The students thought after application of such project, the maquis area would be in focus and later on protection studies would be started.

#### CONCLUSIONS

Enforced in 1956, 'Forest Law' numbered 6831 abandoned maquis which are outside of the forest definition to its fate. It is known that maquis areas have a lot of beneficial functions<sup>34</sup>. For example, Liacos listed in his study the functions of maquis groups (firewood, energy production, landscape and recreation, protection of soil, water production and grazing) where *Quercus coccifera* L. is dominant<sup>35</sup>.

Ozel et al. added in their study on Aegean groups the function 'of using maquis for food and medical reasons'<sup>2</sup>. Also one of the functions of maquis is a good source of food for wildlife. Important functions of maquis, landscape and recreation have always been ignored.

Especially places like Akdeniz University where maquis is densely and variously seen are very important to create awareness in maquis plants use. Students, academic and administrative personals at the University campuses are an important element in protection of bio-diversity and their activities are effective on change. The structures to be obtained through trimming maquis, given as an example in the study on maquis shall be used within the university campus. Moreover, these studies should be performed under the name maquis festival and public awareness should be created. In addition to these studies, the book study on 'Maquis Areas in Mediterranean Region' should be immediately carried out. The findings obtained from this study shall contribute to the plan, design and practice, harmony of urban areas with natural habitats and sustainability of natural areas. Moreover, as these plants are resistant against any and all negative conditions (drought, illness, pests, etc.); their cost is highly low compared to cultural landscape planning and sustainability as well.

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Ecology

# UNMODIFIED EXPRESSION OF rbcS AND rbcL GENES MAY BE RESPONSIBLE FOR THE RESISTANCE OF ALBANIAN LOCAL WHEAT CULTIVARS DAJTI, LVS AND PROGRES, TOWARD MEDITERRANEAN HIGH TEMPERATURES

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**Abstract**. In this study were evaluated the expression of rubisco genes (rbcS and rbcL) from three local wheat cultivars of Albania, named Dajti, LVS and Progres, in order to elucidate molecular factors responsible for their resistance toward Mediterranean high temperatures. Seeds donated from the Centre for Genetic Resources of the Agricultural University of Tirana were germinated in culture following a temperature stress regime, and gene expression was controlled at vegetative and anthesis phase. For this, total ARN was extracted from leaves and RT-PCR was developed using primers specific for rubisco genes following the instructions of HS RT-PCR-SIGMA Aldrich Kit. The electrophoresis of the RT-PCR products was done in 1.4% agarose gel and evaluation of gene expression rate was done based on the concentration of the product in gel. Results show that rbcS and rbcL coding genes were expressed from the three wheat cultivars under stress conditions; Modification of stress regime from 25 to 30°C to 35°C did not affect the expression rate at both phases. In conclusion, at three local wheat cultivars there is not suppression of the expression of rubisco genes, however research should be continued in order to verify the synthesis of Rubisco activase, which can modify the catalytic efficiency of rubisco even though it is produced normally under specific stress conditions.

Keywords: Rubisco genes, temperature stress, RT-PCR.

## AIMS AND BACKGROUND

Temperature stress modifies the photosynthesis rate, water exchange and cell membranes, which may bring extreme reduction of the yield of wheat cultivars. In Albania, a typical Mediterranean country, high temperatures of above 30°C are present from June to September having a strong impact at agriculture, wheat cultivars included. Among many anonymous wheat cultivars, which are extensively used from the food industry in the country because of the favourable prices, there are still in use a number of local cultivars created through long breeding programs from public institutions, named Dajti, LVS and Progres. The last are considered as

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the most adapted to environmental conditions in the country considering their agronomical performance, which was evaluated extensively for more than thirty years.

In general, the genetic basis of wheat heat adaptation is poorly understood and no heat-tolerance genes have been cloned so far. That is why the physiological traits associated with adaptation constitute the best available handle for genetic improvement of crops<sup>1</sup>.

It is known that temperature stress modifies the photosynthesis rate, water exchange and cell membranes, and that understanding at molecular level the mechanisms of impact at physiological cycles is crucial. Temperature stress accompanied by draught under non-irrigated conditions can bring a wide variety of total nitrogen content at wheat varieties<sup>2</sup>. Other factors such as the accumulation of heavy metals in soils are related to the impairment of many biochemical processes important for plant growth<sup>3</sup>. The most affected by high temperatures is Calvin cycle, at which the Rubisco activation by Rubisco activase is reported to be inhibited<sup>4,5</sup>. Rubisco is the most abundant protein on Earth<sup>6,7</sup>, and it is characterised by a number of severe limitations, however is still the only enzyme capable of supporting the net assimilation of carbon that leads to biomass gain. Given its central role in carbon assimilation, the improvement of Rubisco function and regulation is tightly linked with irradiance, nitrogen and water use efficiencies. It is known that Rubisco affinity for CO<sub>2</sub> decreases with temperatures<sup>8</sup>, and research should be conducted to elucidate the genetic variability of Rubisco kinetic properties in order to understand a possible mechanism to increase this affinity, which means to improve the adaption toward higher temperatures. As temperature increases the Rubisco activase becomes less effective in maintaining Rubisco in a catalytically active state9 with its active sites progressively becoming inactive<sup>10</sup>, however, there are reports on the natural diversity in the optimum temperature of Rubisco activases at different plant species<sup>1</sup>. The expression of Rubisco genes is also believed to be reduced by high temperature stress, however, a number of reports describe differences in the heat tolerance of Rubisco synthesis even within a species<sup>5</sup>. Considering the above, in this study we evaluated the expression of so-called Rubisco genes (rbcS and rbcL) from three local wheat cultivars of Albania under HS conditions aiming to understand if the resistance is related to the possibly modified expression of these genes.

#### EXPERIMENTAL

Seeds of wheat cultivars Dajti, LVS and Progresi (100 for each cultivar) were donated from the Centre for Genetic Resources of the Agricultural University of Tirana.

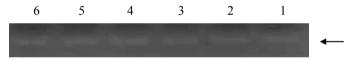
After sterilisation seeds were germinated in culture Hoagland, half of them serving as control  $(25^{\circ}C/20^{\circ}C - day/night regime)$ , and the other half following the temperature stress regime according to Ref. 5  $(30/25^{\circ}C - day/night)$  at the

growth chamber. The effect of temperature stress on rubisco genes expression were controlled after one week (vegetative phase), and after 30 days (anthesis phase). For this, total ARN was extracted from leaves and RT-PCR was developed using primers specific for Rubisco genes following the instructions of HS RT-PCR-SIGMA Aldrich Kit. The electrophoresis of the RT-PCR products was done in 1.4% agarose gel and evaluation of gene expression rate was done based on the concentration of the product in gel.

#### **RESULTS AND DISCUSSION**

The genes encoding Rubiscos (Large and Small) subunits named rbcL and rbcS, chloroplastic and nuclear genes respectively, are reported to be among the genes, which express differentially under heat stress in different wheat cultivars<sup>5</sup>.

In this study we evaluated the expression of rbcS and rbcL genes from three local wheat cultivars of Albania, named Dajti, LVS and Progres, which are well adapted to the Mediterranean high temperatures. The temperature regimes were 20–25°C and 25–30°C, for control samples and heat stressed, respectively. Even though the optimum temperature for the growth of wheat is about 18–24°C, the above temperatures were chosen because they are closer to the natural temperature regime in Albania. The specific RT-PCR for (Rubisco large subunit coding genes) rbcL expression during control and treatment conditions showed that there was unmodified expression from the three cultivars (Fig. 1).



**Fig. 1**. Transcription products of rbcL genes from three wheat cultivars Dajti, LVS and Progres Fragments dimensions are 700 bp (from right to left: 1 – Dajti control; 2 – Dajti treated; 3 – LVS-control; 4 – LVS treated; 5 – Progres-control; 6 – Progres treated

The specific RT-PCR for (Rubisco small subunit coding genes) rbcS expression during control and treatment conditions showed that there was expression from the three cultivars also (Fig. 2).



**Fig. 2**. Transcription products of rbcS genes from three wheat cultivars Dajti, LVS and Progres Fragments dimensions are 560 bp (from right to left: M – molecular marker 1kbp; 1 – Dajti control; 2 – Dajti treated; 3 – LVS-control; 4 – LVS treated; 5 – Progres-control; 6 – Progres treated

A number of works on understanding of the regulation of Rubisco activity report that chloroplast metabolites, inorganic ions<sup>11,12</sup> and tight binding inhibitors<sup>11</sup> interact with Rubisco and regulate its activity and/or prevent its degradation under stress conditions.

Also, the Rubisco activase, a nuclear encoded protein, is reported to be among the molecules that constraint the photosynthetic potential of plants at high temperatures because it is responsible for the regulation of the activity of Rubisco. The elucidation of this mechanism of regulation under heat stress is complicated because of findings on the presence of two or more isoforms of Rubisco activase<sup>13</sup>, the number of genes which code for them<sup>14</sup>, as well as on the heat induced differential expression of Rubisco activase at different plant species<sup>15</sup>. Previous studies report that the decline in Rubisco activity *in vitro* does not increase with temperature<sup>16</sup>, and that a negative interaction with heat inhibited Rubisco activase could contribute or explain the negative impact of elevated temperatures on Rubisco activation state in planta<sup>17,18</sup>.

In conclusion, at three local wheat cultivars of Albania, Dajti, LVS and Progres there is not suppression of the expression of Rubisco genes during vegetation and anthesis phase, however research should be continued in order to verify the synthesis of Rubisco activase, which can modify the catalytic efficiency of Rubisco even though it is produced normally under specific stress conditions.

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Biology

# STATE OF SOME COMMERCIALLY IMPORTANT FISH POPULATIONS IN NATURA 2000 ZONES OF BULGARIAN BLACK SEA AREA

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**Abstract**. Methodology for monitoring of fish fauna in Black Sea was developed. This methodology applies to all fish species monitored to the national system for monitoring biodiversity. Status assessment is carried out in three groups of parameters relevant population data, data distribution and data threats. Analysis of the resulting data is based on tracking the trends of changing the values of various parameters in different years. Species, for which no reference values for such adopt monitoring data exist in the first year, are subject to revision every ten years. Additional monitoring sites were proposed in this project in order to reach the sufficiency of data to assess at geographic (only for species that are reported under Art. 17 of the Habitats Directive) and national level. Such species in Black Sea is Pontic Shad (*Alosa immaculata* E. T. Bennett, 1835). Additional research on growth patterns, migration routes, spawning and feeding areas of this anadromous species with special interest under Habitat Directive is of high importance.

Keywords: Black Sea, monitoring, fish, Alosa, status assessment.

## AIMS AND BACKGROUND

The aim of this study is to collect biological and environmental data in order to make assessment on state of the fish populations of interest in Natura 2000 zones and outside. The state<sup>1–5</sup> of the population assessment was performed by group of parameters<sup>6</sup>, concerning population data and distribution of the species. Bulgaria is one of the EU countries with the richest biodiversity. This also determines the size of the protected areas network in Bulgaria. Protected areas under Directive 92/43/EEC in Bulgaria are 234, protected are 90 habitat types and 121 species of plants and animals, including – 24 species of fish (Multiannual National strategic plan for Aquaculture in Bulgaria, 2014–2020). The particular geological and geographical location of Bulgaria has determined the formation of a wide variety of species of aquatic organisms<sup>7–9</sup>. It has a significant number of endemic species, unique to our or the Balkan Peninsula waters. Therefore, nowadays the protection of the genetic pool and natural fish populations in the country are becoming

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increasingly important in national, regional and even global scale. Fisheries in the Natura 2000 zones - Aladzha Bank (BG0001500), Aheloi - Ravda - Nessebar (BG0000574), Galata (BG0000103), Lake Durankulak (BG0000154), Lake Shabla-Ezerets (BG0000621), Emine-Irakli (BG0001004), Emona (BG0001501), Kamchia (BG0000116) Complex Kaliakra (BG0000573), Beach Shkorpilovci (BG0000100), Beach Garden, Goldfish (BG0000146), Ropotamo (BG0001001), Otmanli (BG0001502) and Strandzha (BG0001007) (Fig. 1) of Bulgarian Black Sea area are characterised by the considerable variety of species caught, as most frequently a fishery object are the small fish species: European sprat (Sprattus sprattus sulinus), Anchovy (Engraulis encrasicholus ponticus), Mediterranean horse mackarel (Trachurus mediterraneus ponticus) and Whiting (Merlangius merlangus euxinus). Other fishes are represented with smaller quantities in the fish yield: Grey mullet (Mugil cephalus), Picked dogfish (Squalus acanthias) and Turbot (Psetta maxima). This study focuses on state of fish populations in the Natura 2000 areas, which offers critical habitat<sup>2-5</sup> to species included in Art. 17 of the Habitats Directive, namely Pontic shad (Alosa immaculata). The species is vulnerable according to IUCN and Bulgarian Red Data Book (http://e-ecodb.bas.bg/rdb/bg/vol2/Alpontic. html; http://www.iucnredlist.org/apps/redlist/details/907/0). It is also included in Annex 2 and 4 of the Bulgarian Biodiversity Act (http://www.biodiversity.bg/files/ File/zak bg biodiv.pdf). During the summer, it is also an important spawning area for anchovy (Engraulis encrasicholus ponticus) and horse mackerel (Trachurus mediterraneus ponticus). Most of the species caught in this Natura 2000 areas are commercially important as Psetta maxima and Sprattus sprattus.

The aim of the study is to assess the ecological status of five species (*Psetta maxima*, *Sprattus sprattus*, *Alosa immaculata*, *Engraulis encrasicolus* and *Trachurus mediterraneus ponticus*) in Natura 2000 zones of Bulgarian Black Sea area.

#### EXPERIMENTAL

*Area of interest in front of Bulgarian coast.* Figure 1 shows the areas specified under Habitat Directive (Council Directive 92/43/EEC) with one mile buffer (under WFD Directive 2000/60/EC) and 3 mile zone (change of Order № RD09-152/09.03.2015).

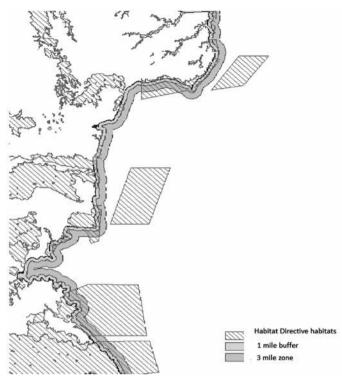


Fig. 1. Map of areas specified under Council Directive 92/43/EEC; Directive 2000/60/EC; change of Order № RD09-152/09.03.2015 in Bulgarian marine area

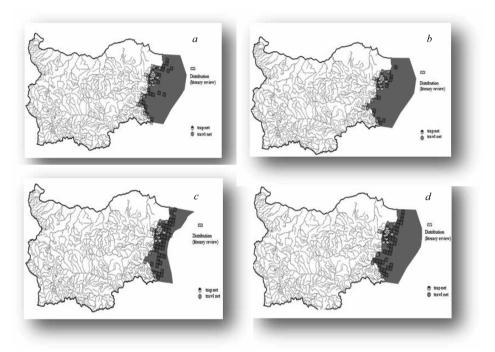
*Gears used for sampling: Uncovered Trap nets.* They are set close to the shore line in shallow waters direct in the migration tracks of the fishes. The fish enters voluntarily, but is hampered from coming out. Fishermen visit traps every day collecting only the captures and leaving the gears set in the same place for the whole season. Mid-water otter trawls (OTM) were used towing gears in order to collect the ichthyologic samples at different depths. Literature survey on distribution of four commercially important species was conducted, as the results were mapped and presented in Fig. 2. According to the survey under 'Operational Program Environment 2007–2014' Field studies of the distribution of species/assessment of the status of species and habitats throughout the country – Phase I – 'Fishes', 2 otter trawl surveys were conducted in July and September 2014. Additionally, in almost all stations replicates were done. Historical data from surveys were used for mapping the spatial and temporal distribution of turbot. The coverage of surveys was in the 12 mile zone of Bulgarian Black Sea waters.

*Size structure*. All caught individuals are distributed in L classes, as interval is 0.5 cm. Vizualisation is done by using hystograms. 'Unvarouble – unsatisfactory'

state is when only one maximum, placed in right or left side of histogram was observed. All the rest of cases are 'favourable'.

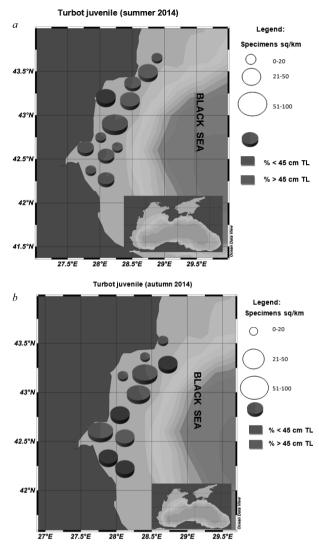
## **RESULTS AN DISCUSSION**

The results from literature survey<sup>2–4,6</sup> on distribution of four commercially important marine fish species are presented in Fig. 2. The main used fishing gears were uncovered pound nets – trap nets (FPOs) and midwater trawls (OTMs).



**Fig. 2**. Map of literature survey on distribution of: anchovy -a; sprat -b; turbot -c, and horse mackerel -d (trap nets and trawls)

The coverage of surveys was in the 12 mile zone of Bulgarian Black Sea waters. The results of juvenile distribution are presented in Figs 3a, b. The field studies were conducted in July and September 2014.



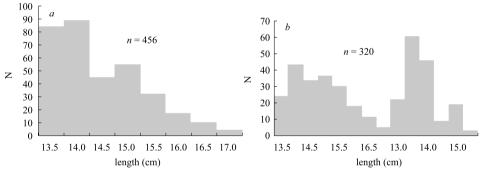
**Fig. 3**. Map of turbot juvenile distribution in Bulgarian Black Sea in July 2014 (*a*) and in September 2014 (*b*)

In July turbot immature individuals (< 45 cm TL) were predominant in the Bulgarian marine area, especially in shallow waters (Fig. 3*a*). The maximum length observed was 56.2 cm. Turbot individuals ranged from 22 to 40 cm in summer in the near shore area between 22 and 65.3 m depth. In September 2014 (Fig. 3*b*) the bulk was composed by length groups 33–40 cm, distributed in depths of 26.6 to 68.1 m, as the specimen with total length of 15–28 cm were less in terms of

abundance in the near shore zone with depths of 21 to 58.2 m. This corresponds with the species distribution revealed in previous surveys conducted<sup>7,8,10</sup>.

Juveniles and immature specimen displayed restricted habitat requirements, mainly occupation of sandy locations.

Size structure. The histogram of Pontic Shad (*Alosa immaculata* E. T. Bennett size structure, caught in uncovered trap nets (in September, 2014) reveals bi-modal distribution on the left side of the histogram with a preponderance of size groups 13.5–14 cm. 14.5–15.0 length groups have lower numbers, as the smaller numbers have 15.5–17 cm groups (Fig. 4). There are no individuals larger than 17 cm, which indicates the presence of young, not sexually mature individuals aged 1+ years. Similar size distributions were observed in previous research<sup>2,3,10</sup>. The state according to this criteria is estimated to be 'unfavourable unsatisfactory'. The number of caught specimen in autumn (n = 320) were lower, as clear bi-modal distribution and lack of individuals with higher than 15 cm size, reveals the state according to this criteria as 'unfavourable unsatisfactory'.



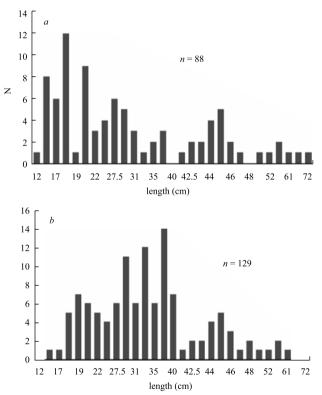
**Fig. 4**. (*a*) *A. immaculata*, spring-summer, length groups (cm); (*b*) *A. immaculata*, autumn, length groups (cm); sampling gear: uncovered pound net (FPO)

The turbot (Fig. 5) size structure (July) varies within 14–53 cm. Predominant size class of 18 cm, followed by size classes of 15, 17, 20, and 22 cm (corresponding to age 1+ year) – An indicator of mass presence of undersized non-sexually mature turbot studied in investigated area. The greatest abundance was of the size class of 18 cm, which is much below the minimum allowable size<sup>6</sup> for turbot (TL = 45 cm, FAA, 2001). The presence of similar size structure<sup>7–9</sup> spoke of 'unfavourable-bad' state in terms of this parameter level monitoring Black Sea area.

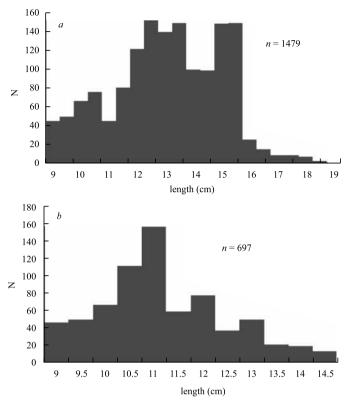
The data on the dynamics of the size structure of scads show a sharp dominance of separate classes ( $\sim 10, 10.5, 11$  and 11.5 cm) and relative length variation order (Fig. 6). The increased participation of these age groups contributes to lower average data dimensional structure.

It is notable absence of individuals with size 15-18 cm. This is probably due to the fact that in the samples presented juveniles and young-of-the-year -0+, 1-1 + 2-2 + 3-3 +. The increased participation of these age groups contributes to lower average data dimensional structure. This corresponds to a 'favourable condition' of the species in the monitoring area.

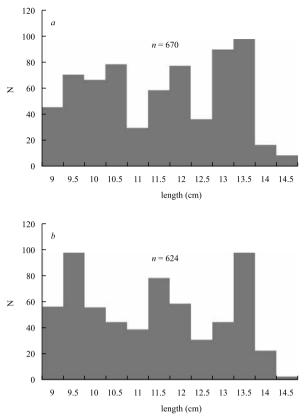
Anchovy length groups varied from 9 to 13.5 cm (age: 1-3 years +). Minimum size for catch of the species identified in FAA is 8 cm (Fig. 7). The state in 2001 is estimated as 'favourable' as regards parameter sized structure. Such distribution corresponds with historically observed for this species<sup>10</sup>.



**Fig. 5**. (*a*) Turbot, *Psetta maxima*, spring-summer, length groups (cm); (*b*) Autumn, length groups (cm), sampling gear: midwater trawl OTM



**Fig. 6**. (*a*) Scad, *Trachurus mediterraneus*, spring-summer, length groups (cm), sampling gear: uncovered pound net (FPO); (*b*) Scad, *Trachurus mediterraneus*, autumn, length groups (cm), sampling gear: midwater trawl (OTM)



**Fig. 7**. Anchovy, *Engraulis encrasicolus*, spring-summer, length groups (cm), sampling gear: uncovered pound net (FPO) – a; anchovy, *Engraulis encrasicolus*, autumn, length groups (cm), sampling gear: uncovered pound net (FPO) – b

#### CONCLUSIONS

The state of the species of commercial interest in Bulgarian marine zone and most particularly in Natura 2000 marine zones, showed highly seasonal character, especially as regards size structure. The seasonal character of different type of gears in use was also very important in analysing the state of species and populations.

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# CLASSIFICATION OF ICT IN EU ENVIRONMENTAL STRATEGIES

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Abstract. ICTs (Information and Communication Technologies) constitute an important tool for the environmental protection and the sustainable use of natural resources. ICTs include any communication device or system encompassing, inter alia, radio, television, mobile phones, computer and networking hardware and software, satellite systems, as well as the various services and applications associated with them. As a global actor, the European Union plays a key role in international efforts to promote environmental protection and sustainable development globally. ICTs are now a major part of European sustainable development strategy. The environmental strategies of the European Union help green the economy, protect nature, and safeguard the health and quality of life of people living in the European Union. This improves both the exchange of information between competent authorities across borders and the direct delivery of online public services to businesses and citizens, which can also be across borders. This paper looks at the current use and trends in the use of ICTs for dealing with environmental issues in European Union in an attempt to enhance sustainable development. The European Union environmental strategies are assessed and classified according to the ICT application categories, which are the following: observation, analysis, planning, management and protection. Findings reveal that the ICTs support most new European Union strategies.

Keywords: ICTs, environmental strategies, the European Union, ICT application categories.

## AIMS AND BACKGROUND

The definition of Information and Communications Technologies (ICTs) is an umbrella term that include any communication device or system encompassing, inter alia, radio, television, mobile phones, computer and networking hardware and software, satellite systems, as well as the various services and applications associated with them<sup>1</sup>. ICTs are the actual platforms where different socioeconomic forces of Humanity have the opportunity to interact, without geographical barriers<sup>2</sup>. The potential for economic growth through new ICTs has barely been tapped. These new possibilities exist largely as a result of two converging forces, the first one being the quantity of information available around the world, exponentially greater than that available only a few years ago and growing at an accelerating

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pace and the second one being the advances in global communications and technological infrastructure<sup>3</sup>.

ICTs such as Internet provide various advantages and benefits by offering a rich, dynamic environment for the exchange of information and resources<sup>4,5</sup>. These modern technologies present very substantial opportunities for us to advance in all areas<sup>6</sup>. Information refers to the transformation of an economy and society through the effective deployment of information and communication technologies in business, social, and public functions<sup>7–9</sup>. Some have posited that ICT has become a general purpose technology (GPT) and have compared ICT to electricity as a fundamental enabling technology, based on its pervasiveness (spread to most sectors), improvement (propensity to get better while lowering the costs of its users) and innovation spawning (make it easier to invent and produce new products or processes)<sup>10,11</sup>.

Positive impacts can come from dematerialisation and online delivery, transport and travel substitution, a host of monitoring and management applications, greater energy efficiency in production and use, and product stewardship and recycling<sup>12</sup>. ICT-based solutions have greatly impacted the environment in a positive manner by reducing greenhouse gas (GHG) emissions<sup>13,14</sup>.

Bringing about transitions to sustainability has emerged as one of the key organising global challenges over the past four decades<sup>15</sup>. Sustainable development nowadays represents a role model with nearly universal appeal<sup>16</sup>. In terms of sustainable development, by definition it is 'development that meets the needs of the present without compromising the ability of future generations to meet their own needs<sup>'17,18</sup>. In March 2010, the European Commission launched the strategy called 'Europe 2020' for smart, sustainable and inclusive growth<sup>19</sup>. According to OECD Report Greener and Smarter<sup>20,21</sup>, Information and communication technologies (ICTs) are a key enabler of 'green growth' in all sectors of the economy. ICTs are a key part of government strategies for a sustainable economic recovery<sup>20</sup>. Other document of the European Union (EU) 'A Green Knowledge Society' also strongly emphasises sustainability, ecological aspects of information system innovation and their impact on sustainability<sup>22,23</sup>. ICT should be seen as a key positive element, empowering EU citizens, growing businesses, and helping us build an open, innovative, secure and sustainable knowledge economy<sup>22</sup>. ICT are now a major part of European economic growth strategy<sup>24</sup>.

In recent years, the role of ICT in the protection of the environment has received significant attention<sup>25,26</sup> and has become the focus of public policies as climate change has been scientifically verified while energy and environmental resources are continually eliminated, thus governments need to enable management strategies to tackle the phenomena and control their use<sup>27,28</sup>. The implementation of most new EU legislation is supported by ICT systems<sup>29</sup>. ICT Implications are defined as the consequences a legislative act can have in relation to the use of ICT

for the implementation thereof<sup>29</sup>. The use of ICT in the implementation might entail the development of new ICT solutions or the adaptation of existing ones and is likely to impact on existing processes and IT systems of the Commission and the Member States<sup>29</sup>. Implementation of legislation will impact in almost every case the processes, the data which needs to be stored, the data exchange between businesses, citizens and governmental organisations or the applications which are used to execute the processes<sup>29</sup>.

This paper looks at the current use and trends in the use of ICTs for dealing with environmental issues in European Union in an attempt to enhance sustainable development. The European Union environmental strategies are assessed and classified according to the ICT application categories, which are the following: Observation, Analysis, Planning, Management and Protection.

#### EXPERIMENTAL

The data for this study were collected from the official European Union website (www.europa.eu). The topics of the EU policies that concern the environment are the following: the environment, the regional policy, the maritime affairs and fisheries, the development and the energy. The first step was to record the regulations, the directives, the decisions, the communications and other acts regarding the environmental issues.

Each policy was studied in order to define whether ICT tools are adopted. In case there was ICT exploitation, the ICT implications were recorded. As ICT implications, we consider: Satellite monitoring systems, Web portals, Traceability systems, Early Warning Systems, Lifelong learning systems, Computer simulation software, E-freight, Electronic tax payment systems, Web-based databases, Telematic systems, Electronic dissemination of information, Geographic Information System, Electronic media, Automatic measurement systems, Systems of electronic reporting, Internet/Broadband networks, Automatic Identification Systems, Systems of Integrated Pollution Prevention and Control, Online shopping, Electronic submission systems, E-navigation, E-government, E-commerce, e-business, Smart grids.

The classification of ICTs in European Union environmental strategies is based on the four application categories as they have been defined by the International Telecommunication Union in 2008 (Ref. 1). The four categories are briefly reviewed below<sup>1</sup>:

– Environmental observation: terrestrial (earth, land, soil, water), ocean, climate and atmospheric monitoring and data recording technologies and systems (remote sensing, data collection and storage tools, telemetric systems, meteorological and climate related recording and monitoring system), as well as geographic information systems (GIS) as it applies to data recording and georeferenced data formats;

– Environmental analysis: land, soil, water and atmospheric quality assessment tools, including technologies for analysis of atmospheric conditions including Greenhouse Gas (GHG) emissions and pollutants, and the tracking of both water quality and availability;

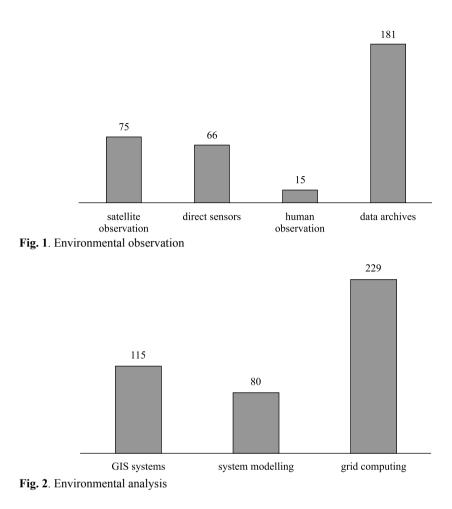
- Environmental planning: planning proceeds from environmental analysis in order to forecast short-term and long-term environmental conditions and objectives. The planning activity may include classification of various environmental conditions for use in agriculture and forestry and other applied environmental sectors. Planning is often focused on specific issues such as protected areas, biodiversity, industrial pollution or GHG emissions. In addition to improving environmental conditions, planning may also include the anticipation of environmental conditions and emergency scenarios, such as climate change, man-made and natural disasters;

- Environmental management and protection: environmental policy and strategic direction set during planning must reach the implementation phase in order to have a direct impact on the environment. In the area of climate change, management and protection deals with issues related to mitigating the impacts of climate change as well as adaptation to climate change.

## **RESULTS AND DISCUSSION**

The research in the EU website resulted in the retrieval of 449 policies (regulations, directives, communications and other acts) concerning environmental issues in a broad range such as environment, regional policy, maritime affairs and fisheries, development and energy. There are 213 policies about the environment, 118 policies about the development, 90 policies regarding the energy sector, and 47 policies regarding regional policy and maritime affairs.

Regarding the category of Environmental observation (Fig. 1), most environmental strategies (181) use archival data, while 75 of them use satellite observation, 66 strategies use direct sensing tools and only 15 strategies use human observers to collect data. As for the Environmental analysis (Fig. 2), most environmental strategies (229) use computational grid, 115 strategies use GIS and 80 strategies use models to conceptualise and construct systems in business and IT development.



Regarding the category of Environmental planning (Fig. 3), most environmental strategies (191) apply data correlation methods, while 102 of them forecast environmental conditions and objectives, and 73 strategies contribute directly to policy formation. As for the Environmental management and protection (Fig. 4), more environmental strategies (166) refer to the planning of mitigating the impacts of the climate change and the adaptation to climate change, while 160 environmental strategies are enforcement strategies to climate change.

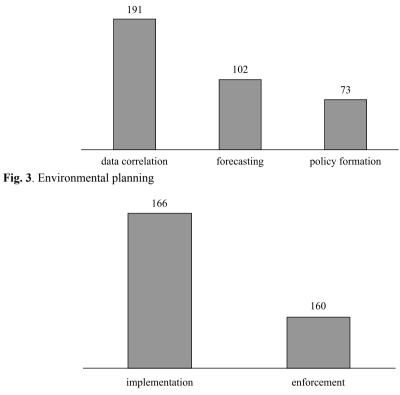


Fig. 4. Environmental management and protection

#### CONCLUSIONS

ICTs have changed many traditional types of community, but also enabled the appearance of new ones<sup>30,31</sup>. On the European Union level a set of Directives, Regulations, Decisions and Communications affects the impact of ICTs on environmental protection and sustainability.

Findings reveal that most EU environmental strategies use archival data and distribute them through grid computing. In most cases, researchers apply correlation methods to the data aiming to find a way to mitigate the impacts of the climate change and to adapt to climate change, while 160 environmental strategies are enforcement strategies to climate change.

The introduction of new ICT tools, concerning network connection and services is necessary within the context of e-innovation society. To prepare for and to take account of the ICT aspects of the EU policies, stakeholders involved in the drafting and the implementation of legislative proposals must be more aware of the ICT exploitation of such proposals. Within that aspect, e-readiness of member

states of the EU should be improved in order to participate in the information age (networked world).

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# TOOL TO MEASURE AND EVALUATE THE ENVIRONMENTAL IMPACT OF PRODUCTION ACTIVITY IN THE ROMANIAN MANUFACTURING SMEs

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Abstract. In the context of sustainability, today and in the near future, all organisations working in the field of production are facing growing pressure to develop production systems that: consume less resources and energy, using renewable resources and materials that are not toxic, minimise the occurrence of undesirable results which can lead to adverse effects on the environment, take into account the degree of recycling, etc. Starting from the studies done concerning the identification and the analysis of the patterns to integrate the 'environmental impact' indicator within an organisation and about the current situation of the Romanian industry, the main goal of our research was to help the Romanian SMEs from the manufacturing industry to transform the current production systems in a sustainable one. This transformation can be done by using a tool, which is presented in this article, to integrate sustainability at the organisational level based on the practices and principles identified at the operational level in the context of eco-innovation. This tool can be considered as a starting point in assessing the efficiency of resource used to measure the environmental impact of production activity.

Keywords: manufacturing, organisations, environmental impact, sustainability, production.

## AIMS AND BACKGROUND

'The environment' and 'the production systems' are two very important points which are debated and studied in our days. Nowadays and also in the near future, all the organisations which activate in the production industry are facing a growing pressure for becoming sustainable<sup>1</sup>.

Regarding the environmental protection, Romanian Government and the public authorities have elaborated a series of regulations which must be respected by all the organisations which activate in the industry. In order to align to the requirements, all organisations, no matter their dimension, have to make modifications to the

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products, to the production process or to services, changes which will reduce the environmental impact. Unfortunately, this change is very hard to be achieved by the SMEs, which are, in generally, less equipped in terms of resources, knowledge and necessary skills for the environmental compliance. In many cases, SMEs are not fully aware of the laws governing their activities, or they do not know what to do. The large number of laws applicable in this area can be confusing and makes all the regulations therefore not fully known and understood<sup>2</sup>.

All studies on the relationship between sustainability and organisations size showed that, for reasons such as the costs involved, the company culture, the support of top management effort required and others, a larger enterprise is more open to become sustainable than a SME (Ref. 3).

The problem is that Romanian industry is dominated nowadays by SMEs and more than that, many of them activate in the production industry.

If we analyse the data on the involvement of SMEs in Romania to protect the environment and in particular those working in manufacturing, we can conclude that the situation is alarming: only 42% of SMEs are operating in accordance with the law and only 25% of them are trying to be more resource efficient<sup>4</sup>. They are not yet aware that, due to the environmental policies implemented at national and EU level, performing the activity in a responsible manner towards the environment is no longer just a 'trendy' or 'nice to have' but will become very soon a must in order to be accepted and to withstand on the market. Moreover, factors such as: the cost of increasingly high raw material, pressure from the local communities, the requirements of investors and customers, require decisions in the shortest possible time on active involvement in environmental protection from organisations working in the manufacturing industry<sup>5</sup>.

Based on the research studies carried out to identify and analyse patterns of the 'environmental impact' indicator integration in an organisation, as well as those focused on involvement in environmental issues of the Romanian SMEs working in manufacturing filed, the general objective is to develop a model of the concept 'Green manufacturing' integration at the organisational level, in order to assist SMEs in the transformation of current production systems in sustainable ones.

#### DISCUSSION

The research aims to develop a tool to support SMEs operating in the manufacturing industry in Romania in their transition towards sustainability through the development of eco-innovative production systems.

The two main reasons for which we choose to support SMEs from production industry are:

• Manufacturing is considered as one of the most important Romanian industry; • Manufacturing industry is considered the main consumer of natural resources and energy that releases the largest amount of greenhouse gas emissions.

The instrument that we propose to SMEs is called the 'Model Integration of Green Manufacturing' (MIGM) and can be considered as a starting point to assess the resources efficiency used to measure the environmental impact of the production activity.

MIGM involves three steps: Assessment, Diagnosis and Improvement Measures (Fig. 1).

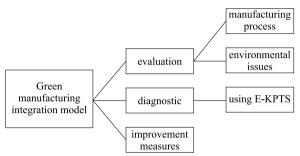


Fig. 1. Green manufacturing integration model

## EVALUATION

In this first phase it is done a data base and an analysis of the information regarding the manufacturing process within the organisation. This 'mapping' of the current situation requires some actions like those given in Table 1:

• description of each stage of the production process;

• inventory of issues regarding environmental protection and the practices used to protect the environment (analysis and environmental impact assessment for each stage );

• preventing emergencies situations: training, response, attribution of responsibilities (identifying pollution prevention measures used, risk and hazard analysis). 
 Table 1. Example of assessment of the production process

Assessment of the production process

Stage description of the production process Phase 1 (description) Phase 2 (description) Phase *n* (description) Analysing and assessing of the environmental impact at each stage of the production process Phase 1 ..... Analysis of how the raw material and materials used affect environmental impacts (water, soil and air) ..... Analysis of how this stage of the production process affects the environmental impacts (type and amount of hazardous and non-hazardous waste, noise, water and energy consumption, etc.) ..... Phase 2 Analysis of how the raw material and materials used affect environmental impacts (water, soil and air) ..... Analysis of how this stage of the production process affects the environmental impacts (type and amount of hazardous and non-hazardous waste, noise, water and energy consumption, etc.) ..... Phase *n* ..... Analysis of how the raw material and materials used affect environmental impacts (water, soil and air) ..... Analysis of how this stage of the production process affects the environmental impacts (type and amount of hazardous and non-hazardous waste, noise, water and energy consumption, etc.) ..... Analysis of how to prevent emergencies: training, response, responsible people (identifying pollution prevention measures used, risk and hazard analysis Phase 1 Identify pollution prevention measures used Risk and hazard analysis ..... Description of how to act in emergency situations ..... The person designated to take measures in case of emergency Name and surname..... Function..... Phase 2 ..... Identify pollution prevention measures used ..... Risk and hazard analysis ..... Description of how to act in emergency situations ..... The person designated to take measures in case of emergency Name and surname..... Function..... Phase *n* ..... Identify pollution prevention measures used ..... Risk and hazard analysis ..... Description of how to act in emergency situations ..... The person designated to take measures in case of emergency Name and surname..... Function.....

## DIAGNOSIS

At this stage, in order to obtain a clearer picture of the current situation within the organisation, we decided that the best solution is to use both qualitative and quantitative indicators.

Therefore, in order to evaluate the quality of the production process, after having obtained a first 'X-ray' of the manufacturing process, there will be made a classification of the environmental problems identified by their level of impact (high, medium low) on the environment (energy, air quality, water quality, green house emissions, use of non-regenerable resources, use of toxic materials, etc.) and the impact within the organisation (costs, stopping the production process, closure of the factory, sales, fines, company image, safety of employs, etc.) (Table 2).

Evaluation of identified en	vironmental problems
Stating the problem	
Description of environmental impact	
Description of impact within the organisation	
Level of in	mpact
Environmental impact	Impact within the organisation
High	
Medium	
Low	

In order to assess the quantitative production process we proposed utilisation of Environmental Key Performance Indicators (E-KPIs). Key Performance Indicators (KPIs) are defined as a set of quantifiable measures which help organisations in defining and measuring the achieved results in comparison with the objectives<sup>6</sup>. Based on this definition F. John Reh deemed Environmental Key Performance Indicators provide organisations with a tool to assess performance in terms of environmental protection, reported to the objectives related to this<sup>7</sup>.

In order to identify the most suitable E KPI's, which will be used to obtain a clear and realistic situation, we used the selection REM criteria that is:

- relevant for activity, operations, objectives;
- easy to use;

• measurable in terms of quantity and quality<sup>8</sup>.

We also took into account the objectives of environmental protection, the necessary time for a fair assessment, the evaluation indicators periodicity (day, week, month, year).

Based on the indicators proposed in the self-assessment tools presented in literature<sup>9–17</sup> and taking into account the selection criteria listed above to assess the quantitative production process, we propose the following indicators:

(1) Ratio of non-renewable resources in the manufacturing process;

(2) Use of hazardous substances in the manufacturing process;

(3) Utilisation of recycled materials in the manufacturing process;

(4) Water consumption in manufacturing process;

(5) Energy consumption (if it is the case, can be calculated also the amount of renewable energy consumed) in the manufacturing process;

(6) Amount of greenhouse gas released during the manufacturing process;

(7) Amount of waste produced in the manufacturing process.

(8) Quantity of waste water resulting from the manufacturing process released into surface waters;

(9) Amount of emissions from the manufacturing process waste released into the air (e.g. SO<sub>x</sub>, NO<sub>x</sub>, NMVOC, PM10, etc.).

In order to reduce the time required going through this stage each of these indicators can be calculated using an Excel file (in which are placed the indicators with related formulas and in the time period recommended for assessment) that we put at the SMEs disposal (Fig. 2).

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3	FORMULA FO	R CALCULATING			Amount of	water consum	ed (cubic met	ers) / NU	MBER OF PRODUCTS (or qua	ntity) produ	ced in the	organizatio	'n
4													
5													
6													
7			Amount of v	water consumed	NUMBER	OF PRODUCT	s						
8	YEAR	MONTH		u.m.		u.m.	Water co	nsumptio	on in manufacturing process				
9	2015	lanuarie											
10		February											
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Fig. 2. Indicators calculation

# IMPROVEMENT MEASURES

In the last stage, called 'Improvement measures' after all required data were collected and processed for a certain period of time (determined by indicator: days, weeks, months or years), the values obtained for each indicator can be analysed in order to evaluate the impact on the environment (positive, negative or constant). On the basis of the results of this assessment, SMEs can analyse various options and scenarios (taking into account the impact on production process, productivity, adaptability of the organisation to certain changes, costs involved, etc.) on reducing environmental impact the production process like:

• replacement of non-renewable raw material derived from non-regenerable or toxic sources with one from regenerable or non-toxic sources;

• changes in the production process in order to be more efficient and to reduce waste and emissions;

• modification of the product in order to minimise environmental impact during the production process;

- efficient use of energy;
- if possible, use of energy primarily from renewable sources;
- use of recycled materials or reuse of materials from the same organisation;
- establish a plan for maintenance of equipment, etc.

#### CONCLUSIONS

In the context of sustainability, although the word 'green' is used to reflect awareness of the threat of environmental pollution, when is associated with the production system, it is used to describe an approach to production that takes into account the impact of production/products on the environment and resources which includes such impact in order to improve planning and control.

Studies have shown that when large and very large organisations make the transition towards sustainable production like 'Green manufacturing', it is much easier and faster than that of SMEs. But in Romania, nowadays, the industry is dominated by SMEs. Moreover, considering the added value and number of employees, one of the most important sectors both at EU level and in Romania is represented by the industry.

According to the report by the EU 'SMEs and the environment in the European Union', estimations show that in each country (including Romania) SMEs in manufacturing are responsible for about 64% of the pollution, being considered the bigger consumers of energy and natural resources and emitters of the largest amount of greenhouse gas emissions.

Taking into consideration the results above, the general objective pursued in the research was to prepare an evaluation model that can be considered a starting point in improving resource efficiency by measuring the environmental impact of production activity.

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# SUSTAINABLE USE OF AGRICULTURAL LANDS IN THE SCOPE OF URBAN AGRICULTURE: THE SAMPLE OF MALATYA, TURKEY

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**Abstract**. Agriculture is a strategically very important sector for every country since it supplies all vital food elements people need, meets the raw material demands of national industries, offers job opportunities to people from every levels of society and inputs foreign currency to national economy by exporting crops. However, it is one of the most affected sectors by the increasing urbanisation impacts due to rapid human population increase, improving socio-economic conditions and advancing technology. The province of Malatya located in Eastern Anatolia region of Turkey meets nearly 80% of world dried apricot need. Urban agriculture lands taking place in urban tissue of Malatya city and composed mainly of apricot orchards (AO) have been lost at an important rate for the last 15 years. From this point of view, in the scope of the present study, land cover (LC) and land use (LU) types were determined first to determine the rate of lands lost to the use types other than agriculture. The rate of the areas gained and lost was determined by investigating the changes in these LC/LU types between 1984 and 2014. Protective and improving strategies were developed by determining socioeconomic and legal and administrative reasons for the lost agricultural lands (AL) and AO.

Keywords: urban agriculture, apricot, urbanisation, Malatya, Turkey.

## AIMS AND BACKGROUND

Rapidly growing world human population in the last years, people wish to live in suburbs away from city centres, changing socio-economic structure, land use policies and advanced technology cause the transformation of natural lands into urbanised areas. Urban sprawl can cause dense stress on natural and cultural landscapes taking place in close proximity of cities<sup>1–10</sup> by affecting AL the most<sup>7</sup>.

Urban agriculture can be defined as growing plants and raising animals for food and other production types and related activities such as transportation of outputs or inputs, processing agricultural raw materials (e.g. cutting, packaging, etc.) and various marketing activities<sup>11</sup> based on products within and around cities

<sup>\*</sup> For correspondence.

and towns. Urban agriculture may mean higher quality, fresher and cheaper food, a production activity to evaluate and clean domestic wastes and larger green area. Because of such characteristics, urban agriculture is advocated to be a possible solution for various problems in cities<sup>12,13</sup>. From this point of view, in recent years, several studies related mainly to socio-economic development have had a closed look at agricultural activities within and around urban areas<sup>14–16</sup>. Applications such as urban farms, community and allotment gardens can produce values and make contributions to local, regional and national economies. It is the typical of ordinary urban systems to end natural ecology in and around their borders. Instead of a healthy ecosystem for every living thing, where nutrients are largely recycled; most cities prefer to dump, haul, or pipe away tons of organic garbage and sew-age<sup>17</sup> by wasting a huge and valuable source and polluting nature in many ways.

The province of Malatya, in eastern part of Turkey, inhabits an important rate of urban agriculture activities due to apricot growth. The city alone accounts for 60 and 95% of fresh and dried apricot production in Turkey and 80% of world dried apricot need is met by the city<sup>18</sup>. Objective of the present study is to develop protecting and improving strategies for sustainability aims about fertile AL and AO in and around Malatya city centre by determining land losses due to urbanisation in urban agriculture and apricot growth.

It was aimed also to follow 30-year (1984–2014) changes of urban sprawl and its narrowing effect on urban AL, in other words, to analyse the change in AL lost in time in compact urban structure. Based on this aim, the method developed consists of 4 main components (Fig. 1).

At Stage 1, pre-operations were applied to satellite images of LANDSAT 5 TM and LANDSAT 8 OLI in WGS 84 37N projection before their classification. This stage involves two sub-stages: (a) atmospheric removal which means that atmospheric removal was applied through FLAASH algorithm, and (b) study area was derived from main image.

At Stage 2, object based classification of satellite image was performed and this stage includes two sub-stages, i.e. segmentation and rule based classification.

At Stage 3, the method of analysing the changes in landuse types obtained from the classification was conceptualised by the following relationship:

 $t_{-1}$ : Baseline (Time 1)

 $t_0$ : Present time  $G_a$ : Gained area

 $t_{-1} \cap t_0 = P_a - t_{-1}/P_a = L_a - t_0/P_a = G_a$ 

L<sub>a</sub>: Lost area

P<sub>a</sub>: Persistent area

At Stage 4, change analyses of LU types were evaluated together with socioeconomic data.

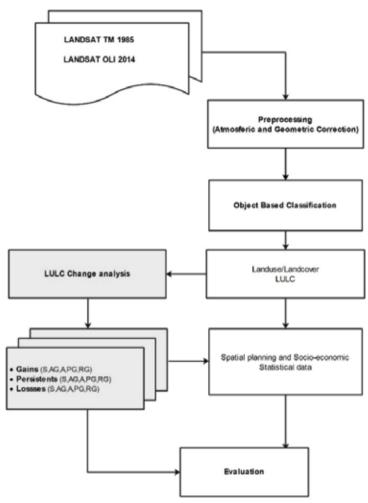


Fig. 1. Flow chart

# EXPERIMENTAL

Study area is composed of the Central, Battalgazi and Yeşilyurt districts of Malatya province (Fig. 2). The province of Malatya gained the legal administrative status of grand municipality required by the Code enacted on 12th November 2012. City centre has recently expanded and involved the districts of Battalgazi and Yesilyurt. Total human population of Malatya city centre and other combined settlements is 447 240 and mean population density is 66 ppl/ha. By considering the present situation and trend of the city, population projection and density of the city are estimated to reach 1 000 000–1 100 000 and 73 ppl/ha by 2040.

Malatya city centre has developed around D-300 highway in east-west direction. Industrial areas accumulated in west side of the city. According to Territorial Plan and Regional Development Plan (2014–2023) (Ref. 19), leading sectors were determined by considering present trends, potentials and strategies produced in Malatya city centre to be service, industry, agriculture and tourism, respectively<sup>20</sup>.

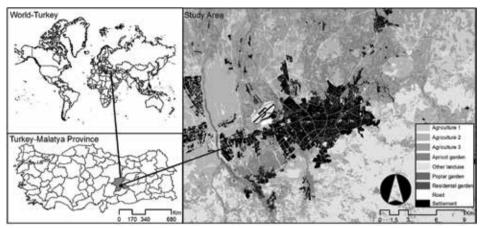


Fig. 2. Location of Malatya province and study area

Data obtained as the result of environmental observations in the qualitative and quantitative changes of LC/LU are accepted to be significant tools to be used to achieve sustainable LU targets<sup>21</sup>. As the result of the classification carried out for Malatya, 12 LC/LU classes were determined (Table 1, Fig. 2).

Land cover/use classes	1984	2014 Surface (km <sup>2</sup> )		
	Surface (km <sup>2</sup> )			
AL 1	132.19	55.35		
AL 2	168.19	103.22		
AL 3	83.94	161.07		
AO	36.77	43.89		
Arid land	146.06	166.82		
Industrial areas	2.32	8.60		
Mining and construction areas	0.50	1.30		
Rocky areas	1.01	1.07		
Roads	7.84	10.10		
Housing areas	15.10	38.38		
Urban green spaces and house gardens	8.34	12.54		
Water surfaces	0.75	0.66		
Total	603.02	603.02		

Table 1. Land cover/use change in Malatya

It can be observed when Table 1 is evaluated that total AL decreased from 383 to 319 km<sup>2</sup> while the total area of roads, industrial zones and housing areas increased from 25.2 up to 57 km<sup>2</sup> (an increase of 126%, Fig. 3*a*). An increase by 19 and 50% is observed in AO and urban green spaces, respectively. Analysis of the changes in land use is given in Table 2 and Fig. 3*b*.

Land use	Permanent (S <sub>a</sub> )	Lost (K <sub>v</sub> )	Gained (K <sub>a</sub> )
	(km <sup>2</sup> ) "	(km <sup>2</sup> )	(km <sup>2</sup> ) "
Urban land use change	14.8	10.30	41.8
AO	14.7	22.00	29.1
Rocky areas	0.64	0.37	0.43
Urban green spaces and house gardens	2.5	5.80	8.00
AL	261.1	123.10	58.5

Table 2. Land use change

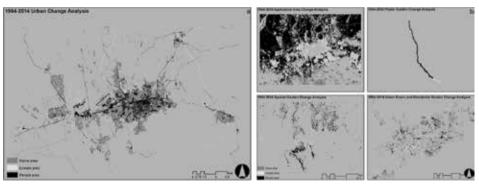


Fig. 3. Urban change analysis (a) and land use change analysis (b)

#### **RESULTS AND DISCUSSION**

It seems as the result of the analysis that total area of the AO increases, however; in especially the close proximity of the city, AO and AL shrink clearly depending partially on human population in city centre. According to data of TurkStat 2015, net migration rate has been decreasing since 1984 in the city centre showing that the number of urban people increases by 8% while rural population decreases by 11%. Industrialisation, including the increase in the number of industrial facilities and zones can be counted among the reasons for migration from rural to urban. Increasing apricot crop yield through the financial supports from governmental side caused an increase in prosperity level and so in the demand for new housing estate areas and the number of motor-vehicle. New housing areas have been opened in

especially west part of the city by also increasing total length and traffic density of the roads and thus driving AO and AL to threats.

One of the reasons for apricot orchards and arable lands to face urbanisationbased threats may be the absence of the protective legal and administrative framework including these areas. Malatya city was granted the legal status of grand municipality through the enforcement of a Turkish Legal Code in 2012. However, no planning effort has so far been performed to take under control the consistently increasing urbanisation pressure on agricultural lands in the periphery of the city. In development plans and related documents, the city development direction and new road systems are defined to be towards agriculture lands and any alternatives have not been offered for such situation. From this point of view, the first action to perform may be the preparation of the agricultural master plan which can sustain agricultural activities based on fruit production by determining suitable areas for such activities.

#### CONCLUSIONS

Threatening pressure and negative impacts of urbanisation on agricultural/arable lands have extensively increased in the world and Turkey to a level at which agriculture design, product diversity and food security have been taken under several threats. Agricultural activities conducted in and close proximity of urban are (i.e. urban agriculture) is accepted to be an important part of urban development strategies and food security policies<sup>22,23</sup>. In this respect, management and protection strategies to be implement in both Malatya and Turkey may include: (1) urban and rural spatial setup should be thought in comprehensive planning approach while preparing spatial plans; (2) spaces where urban agricultural activities can be performed should be chosen in city development plans; (3) existing national agricultural product design should be integrated with the mentioned development plans, and (4) local products should be supported by governmental incentives. It can also be suggested specifically to Malatya city that protective planning attempts should be performed for agricultural lands in close proximity of the urban area of the city to form a protected agricultural corridor which can direct and manage compact city development, prevent transformation of arable lands into urban area in periphery and remove fragmented structure of agricultural lands.

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# IMPORTANCE OF POLLINATING INSECTS FOR MAINTAINING SUSTAINABLE AGRICULTURE IN EASTERN CROATIA

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**Abstract**. Significant efforts are currently undertaken in Europe for estimating the size and stability of insect populations, especially the useful pollinators. In Croatia, there is a lack of researches on size and stability of the population of pollinators not managed by the man. Insect pollination benefits to plant reproduction within natural and agro-ecosystems, and contributes to conservation of wild flora, as well as crop production. Furthermore, wild pollinators are known as indicators of the environmental stability. Importance of honey bee is high in the intensive agricultural areas, where wild pollinators are suppressed due to shortage of food sources and nesting spaces. In the research, we used pan-traps to estimate pollinator diversity in three different habitats in eastern Croatia: Nature Park 'Kopacki Rit' as natural and undisturbed habitat, countryside near the Park as semi-natural habitat, and agricultural habitat during one-day period for capturing pollinating insects. Highest richness, diversity and evenness values are recorded at semi-natural habitat, while lowest values are recorded in agricultural habitat. For maintaining sustainable agriculture it is recommended to conserve some natural habitats for wild pollinators in the intensive agricultural areas.

Keywords: pollinators, insects, habitat, agriculture, Croatia.

## AIMS AND BACKGROUND

The main aim of this study is to estimate actual diversity of wild pollinating insects in eastern Croatia, to determine population trends, and analyse impact of intensive agriculture on pollinating insects.

Presence and abundance of pollinating insects highly effects the global food production and today 35% of global crop production highly depends on the animal pollinators<sup>1</sup>. Pollinating insects increase fruit or seed quantity and quality of 39 of 57 major crops worldwide<sup>1</sup>. The value of insect pollination on global scale is estimated on \$153 billion (Ref. 2), and in Europe on €14.6 billion per year (Ref. 3).

While feral honey bee (*Apis mellifera* L.) colonies have mainly disappeared, colonies managed by man suffer evident loss in Europe<sup>4</sup> and USA<sup>5</sup> every year. Due

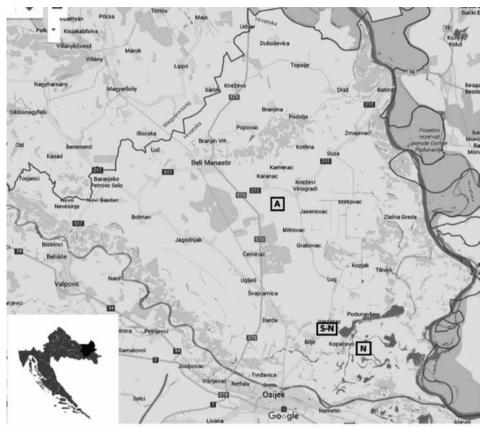
<sup>\*</sup> For correspondence.

to efforts of beekeepers and because of their economic importance, most of the honey bee colonies are repopulated before next winter period. This is not a case in populations of non-bee pollinators. Trends of evident wild pollinator decline<sup>6</sup> and consequently disappearance of some plant species that depend on them is confirmed in the recent years. The most significant reasons for pollinator decline are change in land use, application of pesticides, environmental pollution<sup>7</sup>, decreased resource diversity, invasive species, the spread of pathogens and climate change. Since the natural habitats for wild pollinators are decreasing and cultivated area for food production is growing, pollination depending crops today mostly depends on honey bee as pollinator<sup>8</sup>. Intensive agricultural operations have negative effects on species richness, abundance and diversity of wild pollinator in Europe<sup>9</sup>, while species richness and benefit for biodiversity<sup>10</sup> increases with more areas under semi-natural habitats in the landscape.

For monitoring the diversity of pollinating insects in some area it is necessary to provide a representative population sample. Pan trap method is very efficient method for monitoring populations of pollinating insects in both agricultural and semi-natural habitats<sup>11</sup>. Pan traps presents standard and verified method for trapping pollinating insects and evaluation of their diversity<sup>12,13</sup>. Important features of the systematic monitoring of changes in population of pollinators are simplicity, repeatability, low-cost implementation and ability to detect small changes in their status.

#### EXPERIMENTAL

*Study area*. Three different sampling locations for trapping the pollinating insects were selected in Baranya region of eastern Croatia (Fig. 1), considering the habitat characteristics and anthropogenic impact. First location was in natural habitat (N) in the area of Nature Park 'Kopacki rit' where no agricultural interventions or any chemical usage is allowed. Second location was in semi-natural habitat (SN) in the Vardarac village, where some agricultural practice is taking place, but still there are unpolluted and neglected areas nearby. Third locality was in agricultural habitat (A) with intensive crop production for decades. The most common sown crops are rape seed, wheat, barley, corn, sugar beet, sunflower and soybean.



**Fig 1**. Geographical position of the study area with distribution of sampling location (N – natural habitat, S-N – semi-natural habitat, A – agricultural habitat)

*Sampling and analyses.* Three pan traps in different colour: white, yellow and blue, were used for attracting pollinating insects. Pan traps were white plastic plates 20.5 cm wide and 4 cm deep. Plates were sprayed with fluorescent yellow and fluorescent blue colour or left white. Before exposing traps were filled with water and few droplets of dishwashing liquid were added for reducing the surface tension of water. So, if insect was attracted with plate colour and landed it would simple sink in water. At each location, 10 of each colour pan traps were placed at distance approximately 10 m one from each other. The pan traps were exposed for one day, on 11 July 2015, from 9:00 till 17:00. After collection, all specimens were preserved in 70% ethanol until identification. All trapped insects were identified to the lowest possible taxonomic level with entomological identification key<sup>14</sup>.

We estimated richness and calculated the Shannon-Weaver<sup>15</sup> and Simpson

indices as a measure of diversity. The Shannon-Weaver Diversity index (H) was calculated according to the following equation:

$$\mathbf{H} = -\Sigma \mathbf{P}_i (\ln \mathbf{P}_i)$$

where  $P_i$  is the proportion (n/N) of individuals of one species (n) divided by the total number of individuals found (N). The value of Shannon-Weaver diversity index varies between 0 and 4.6 and higher index value presents higher diversity.

The Simpson Diversity index (D) was calculated according to the following equation:

$$D = 1 - \frac{\sum n_i (n_i - 1)}{N(N - 1)}$$

where n is the total number insects of one species and N – the total number of individual insects found at habitat. The Simpson index ranges between 0 and 1, higher value indicates higher diversity.

The Pielou J' evenness index<sup>16</sup> was calculated to estimate species evenness distribution in a habitat. For calculation following equation was used:

$$J' = H/(\ln S)$$

where H is the Shannon-Weaver index and S – the total number of species found in habitat. Values of evenness index are between 0 and 1 and the less variation in habitat is, the value of J' is closer to 1.

The Sorensen similarity index (CC) was calculated to provide information on similarities between two habitats according to:

$$CC = \frac{2C}{S1 + S2}$$

where C is the number of species two habitats have in common and S1 and S2 – the total number of species found at first and second habitat. The value of the Sorenson coefficient can vary between 0 and 1 and more habitats are similar, value of coefficient is higher.

#### **RESULTS AND DISCUSSION**

Total of 196 insect individuals were identified (Table 1). In total, 87% of all pollinating insects captured in traps were identified up to family level and 47% to species level. According to entomological classification system, identified insects belong into 6 orders, 11 families and 7 species. The most numerous families are Cicadelidae (41 individuals), followed by Apidae (35 individuals) and Muscidae (32 individuals).

Taxonomic category	Habitat type					
order/family/species	natural	semi-natural	agricultural			
Odonata	5	6	_			
Diptera						
Muscidae						
Musca domestica	16	15	1			
Hymenoptera						
Apidae						
Apis mellifera	5	4	20			
Osmia spp.	_	6	_			
Formicidae						
Formica rufa	9	_	10			
Ichneumonidae	6	15	3			
Vespidae						
Polistes gallicus	5	_	1			
Coleoptera						
Chrysomelidae	_	3	_			
Coccinellidae						
Psyllobora 22-punctata	_	4	_			
Mordellidae						
Mordella aculeata	1	_	4			
Unidentified	5	9	_			
Hemiptera						
Cicadelidae	3	8	30			
Neuroptera						
Chrysopidae						
Chrisoperla carnea	1	1	_			
Total	56	71	69			

Table 1. Taxonomic arrangement and distribution of pollinating insects according to habitat

Out of 13 entomological taxa identified at order, family, genus and species level, 10 were recorded in natural and semi-natural habitat, each and 7 in agricultural habitat.

Regarding each particular habitat, most frequent insects in natural habitat belong into Muscidae family, in semi-natural habitat into Muscidae and Ichneumonidae families, while in agricultural habitat most frequent insects belong into Cicadelidae family. In all habitats, insects from families: Muscidae, Apidae, Ichneumonidae and Cicadelidae are recorded, while insects from Chrysomelidae, *Psyllobora 22-punctata* and *Osmia* spp. species were recorded only in semi-natural habitat.

In agricultural habitat, 72% of all identified insects belong into two families: Cicadelidae and Apidae. The percentage of honey bee (*Apis mellifera*) was about 30% in agricultural habitat, 9% in natural and 5% in semi-natural habitats. This could be explained with the fact that sunflower bloom ended just few days before sampling time, and probably some beekeepers still had their hives present nearby.

The number of captured insects was lowest in natural habitat (56) and did not differ between semi-natural (71 insects) and agricultural habitat (68 insects recorded). Natural habitat<sup>17</sup> offers good conditions for nesting space to pollinating insects, but semi-natural and agricultural habitats provide more diverse food sources than natural habitat.

Higher values of Shannon-Weaver and Simpson index (Table 2) indicate higher diversity of pollinating insects in natural and semi-natural habitat than in agricultural habitat. Similar trends are recorded in other countries<sup>18</sup>. Values of Shannon-Weaver and Simpson diversity index were highest in semi-natural habitat, like in some previous studies<sup>19</sup>.

Table 2. Values of calculated arversity indexes for three anterent hashat types					
Habitat	R	S	Н	D	J'
Natural	10	56	2.05	0.86	0.89
Semi-natural	10	71	2.10	0.87	0.91
Agricultural	7	69	1.43	0.73	0.62

Table 2. Values of calculated diversity indexes for three different habitat types

R – richness, number of different species identified; S – number of individual insects identified; H – Shannon-Weaver index; D – Simpson index; J' – Pielou Evenness index.

Lowest values of Shannon-Weaver diversity index in agricultural habitat mean that captured insects are not evenly distributed, and most of insects belong into Apidae or Cicadelidae families. In natural and semi-natural habitat, value of H index is higher, but still around mean index value.

It seems that semi-natural habitat offers the best living conditions for wild pollinators. They can easily find nesting site and shelter, while various flowering crops and fruits from surrounding area provide quality food source<sup>18</sup>.

Similar values of Shannon-Weaver index were recorded in semi-natural habitats in Vojvodina (Serbia)<sup>20</sup>. However, there are some studies explaining greater abundance and diversity in agricultural habitat<sup>21</sup>, probably due to the pollen and nectar sources from flowering crops.

Intensive agriculture in some way can enhance abundance of pollinators<sup>18</sup>, but concurrently flowering of the cultivating and wild plants potentially jeopardises wild plant species throughout a competition. Lowest value of Pielou evenness index was recorded at agricultural habitat, showing that number of insect found in habitat are not evenly distributed among species. Natural and semi-natural habitat had highest evenness values (0.89 and 0.91) indicating equal distribution of individuals among the species found. Sorensen's index of similarity (CC) between natural and semi-natural habitat was 0.7 and between semi-natural and agricultural habitat

was 0.47, which means that there is no much of overlap between semi-natural and agricultural habitat, as it is expected.

## CONCLUSIONS

Our results suggest that highest richness, diversity and evenness of pollinating insects are recorded in semi-natural habitat. Such environment provides nesting sites for pollinators and lot of diverse food sources available from crops and fruits grown in the area.

For securing the sustainable agriculture and saving many of the native wild plants, it is important to keep the stable and healthy populations of pollinating insects.

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## ASSOCIATIVE STRUCTURES, FACTOR OF PROGRESS IN THE DEVELOPMENT OF ANIMAL HUSBANDRY IN ROMANIA – A CASE STUDY, THE NATIONAL ASSOCIATION OF GOAT BREEDERS CAPRIROM

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**Abstract**. Because the goat breeding sector has steadily increased in the past decade in Romania, reaching a livestock of 1 605 860 heads, and due to the interest to consume goat products – meat, milk and derivatives, the importance of managing these issues by a husbandry associative entity, an important branch of agriculture, describes an active and positive impact socially and economically. By managing 20% of the Romanian goat population (stock from 410 goat farms) and having 25% of the national livestock in Official Control of Milk Production, the National Association of Goat Breeders CAPRIROM (NAGB CAPRIROM) leaves its mark on the economy of the entire goat breeding sector, solving specific and real economic and social problems, in an environment often difficult and competitive. With a relevant activity in the goat farming field, NAGB CAPRIROM influences positively and resolves important issues of a developing economic sector that is founded on an occupation that has always defined the Romanian space – shepherding.

Keywords: animal, husbandry, associative, structures, survey.

## AIMS AND BACKGROUND

The aim of this study is to highlight the importance of the animal husbandry associative structures, the objective being the support of Romanian livestock farmers and the representation of their interests, taking as a case study the National Association of Goat Breeders CAPRIROM, founded in 1993. This goal was achieved by analysing the impact of NAGB CAPRIROM services on goat farms development and consequently the importance of this animal husbandry association for the entire sector of exploitation of this species in Romania. The association impact on livestock farming seen as a pole of competitiveness cluster type that manages association of companies, farms, research and training organisation, acting in partnership and implement a common development strategy<sup>1</sup>.

<sup>\*</sup> For correspondence.

## EXPERIMENTAL

The impact assessment process for the evaluation used the investigation based on a survey and used as a research tool a specific questionnaire containing 20 questions relevant to the association work in the territory<sup>2</sup>. The impact analysis of NAGB CAPRIROM services and activities on the development of the goat husbandry sector was achieved through the following indicators: the image of NAGB CAPRIROM in the field, the farmer interest in a professional association, the quality of NAGB CAPRIROM activities and services, and the association impact on the goat farms development. The following stages were covered in the design of the questionnaire-based survey: establishing and delineating the research topic, statement of the objectives, documentation, determination of the research sample, drafting the questionnaire project, applying the questionnaire, processing and interpretation of data, and conclusions as a result of the research. The preliminary documentation took into account the database regarding the NAGB CAPRIROM members, which includes the farmers identification data, and also data regarding the goat farms. The target population was represented by 400 farmers, members of NAGB CAPRIROM, the sample on which the study was done counting 100 responders. They were given both closed and open questions, allowing the responders to express their opinions freely. The period of questionnaire fill out was 01.02.2016–29.02.2016. The sampling unit corresponding to the selection of the survey sample was individual – the farmer.

## **RESULTS AND DISCUSSION**

In what follows, we are presenting the questionnaire given to the target population and its answers interpretation.

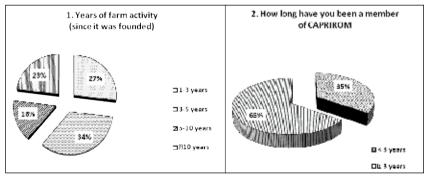


Fig. 1. Responders answers to questions 1 and 2

Regarding answers 1 and 2 (Fig. 1), it is noted that most CAPRIROM members are farmers for 1–5 years (61%), opting for the registration in a professional association right from the beginning or very early of their activity (65%). If from the viewpoint of the total number of animals from a farm, it can compete with known countries in the field, the ratio of households with 1–2 heads (over 32%) is to the breeders disadvantage. The livestock of most NAGB CAPRIROM farms (52%) counts 100–500 heads, considerably above the livestock average numbers per farm, with 88% farms with less than 10 heads (Ministry of Agriculture and Rural Development – Romania, 2015, http://www.madr.ro/) (Fig. 2).

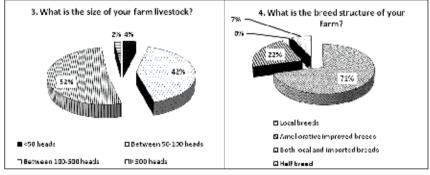


Fig. 2. Responders answers to questions 3 and 4

This aspect is positive in terms of the farms economic efficiency, knowing that it becomes lucrative at over 100 goat heads. In Romania, approximately 80% of the livestock is Carpatina, a rustic breed, with low production. The rest of 15% is represented by White of Banat, an improved breed, with superior production and 5% are imported specialised breeds. In what regards the breed structure in the NAGB CAPRIROM farms, the goat livestock is made up of non-specialised local breeds -71%, the production direction being predominantly towards milk. but also meat, however, with small amounts and poor quality (Fig. 2). With the purpose of eliminating these shortcomings, a directional selection was made towards milk production by accomplishing lines of hybrids with superior productive potential -7%, adapted to the market requirements and keeping in line with the Western-European countries tendency. 22% of the breeders own animal farms with imported specialized breeds. Goat milk has not had and does not have an imposed quota and the request is still higher than the supply in all EU members (Fig. 2). As long as livestock are decreasing in the European Union, the development of associative organisations in this field can only be in favor of Romanian farmers<sup>3</sup>.

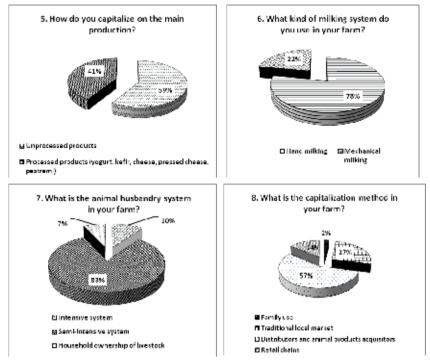


Fig. 3. Responders answers to questions 5, 6, 7 and 8

The vision of the NAGB CAPRIROM members commands the continuous improvement of the rearing and exploitation technologies (semi-intensive and intensive systems) by expanding the mechanisation of farm activities, mechanical milking and the in-farm processing of the main productions – milk and meat (Fig. 3). In fact, 90% of the farmers rear goats for milk. Thus, 10% of the farms use intensive systems, this aspect being correlated with 41% of the farms that process their own production and with 22% of the farms in which mechanical milking is employed (Fig. 3). These have the possibility to capitalise the farm production to retailers – 14% (Fig. 3). Instead, much of the production obtained inside NAGB CAPRIROM permits, thanks to the used livestock, the obtaining of a sustainable production from goats<sup>4</sup>. The improvement programs take into account the coordination of all the works done with the purpose of ameliorating the breed: registration in the Herd book, official control of the milk production, reproduction organisation (the testing of reproducers, certification of the value and quality of the goats to be used for reproduction, AI – artificial insemination), nutritional modulation (Fig. 3).

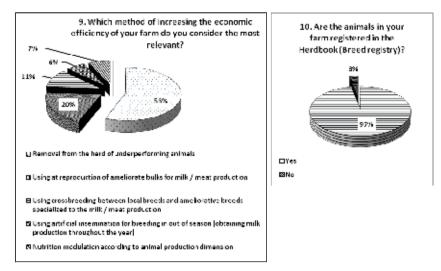


Fig. 4. Responders answers to questions 9 and 10

The members have animals registered in the Herdbook in a significant proportion. NAGB CAPRIROM is accredited by the National Agency for Animal Husbandry for owning the Herd book of Carpatina, White of Banat, Saanen, Alpine, Murciana Granadina (breeds specialised in milk production), as well as Boer and Nubian.

In 56% of the interviewed people, the economic efficiency increased by the elimination of the underperforming animals, based on the data provided by OCMP. 31% use AI as efficiency method, with seminal material from ameliorative bucks. 6% of the farms resorted to AI out of season in order to have milk production in October-February when this product traditionally misses from the market (Fig. 4).

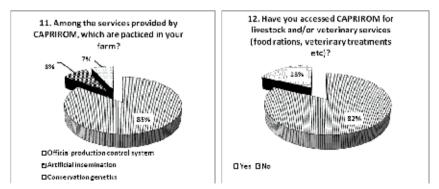


Fig. 5. Responders answers to questions 11 and 12

Thus, the association specialists:

 Check and approve the breeders requests regarding the taking over of the animals in the official control and provide the necessary personnel required for the official control;

- Execute the official control of the milk production (OCMP) at national level (Fig. 5);

- Transmit to the zonal laboratory milk samples for the quantitative analysis, accompanied by the control bulletin (optional);

- Transmit to the breeders the results of the official control, after each control accomplished in the farm, for a more efficient farm management<sup>5</sup>.

This way, milk is capitalised for a good price and this aspect facilitates closing contracts with buyers. Another method to increase productivity is nutrition modulation, in the sense of an efficient exploitation of the nutritive principles, correlated with the type of production and its level (Fig. 5).

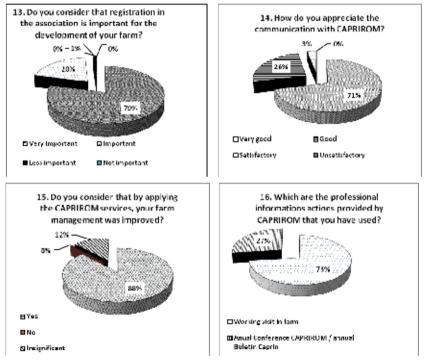


Fig. 6. Responders answers to questions 13, 14, 15 and 16

The NAGB CAPRIROM objectives are:

 to co-interest goat breeders for the numerical increase of the livestock as well as production and reproduction performances, with the purpose of increasing the income; - to apply technical measures regarding the genetic improvement using reproduction biotechnologies, especially artificial insemination – AI;

- to offer technical assistance for projects (National Rural Development Programme Romania, the European Agricultural Fund for Rural Development-EAFRD);

- to offer information and technical support for a diversified capitalization of goat productions<sup>6</sup> (Fig. 6);

- to support breeders in activities of supply with feed, medicine and disposable elements specific to goats;

 to organise round table, conferences, communications and publications regarding the promotion of new scientific and practical information in the field of goat breeding;

 to implement development policies in the goat breeding sector by zonal branches and of appropriate marketing strategies<sup>7</sup>;

- to defend the rights and interests of goat breeders in relation to various governmental or non-governmental organisms.

Consequently, 79% of the breeders declare as very important the collaboration with the association, 71% of them having a very good communication with NAGB CAPRIROM (Fig. 6). 88% of the farmers believe that their farm management was improved by using NAGB CAPRIROM services, the main communication method being the work visit in the farm – 73%. Every year there is a National Conference of Goat Breeders of Romania – CAPRIROM, a conference to which the following are invited: branch presidents, participants to the genetic conservation program, breeders, farmers and specialists in goat breeding, but also specialists from governmental organisations, Ministry of Agriculture and Rural Development – Romania, the National Agency for Animal Husbandry- Romania, universities and agricultural institutes, as well as members of the IGA board (The International Goat Association) (Fig. 6).

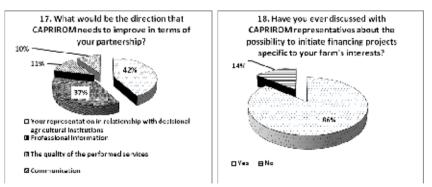


Fig. 7. Responders answers to questions 17 and 18

With the purpose of improving communication with NAGB CAPRIROM, 42% of the farmers interviewed want a better representation in relation with decisional agricultural institutions and 86% are interested in available information regarding access to national and European funds in the zootechnical domain, and not only (Fig. 7).

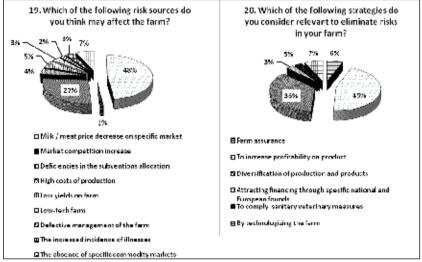


Fig. 8. Responders answers to questions 19 and 20

There have been identified other development adjacent targets that can attend to the welfare of farm: agro-tourism, organic production, handicraft, etc.<sup>8</sup>

Most responders (45%) believe that risks decrease by increasing profitability per product, by eliminating underperforming animals, by rearing a smaller number of bucks, by using AI and by reducing the feed losses though nutritional modulation. The Romanian consumer is not educated to consume goat dairy and milk, which is why there is no stable market for this sector at the moment. That is why the price per product may fluctuate considerably. Added to this, subsidy delays or repeated legislative amendments disrupt the annual activities planned in the farm<sup>9</sup>. Accessing national and European projects is a solution for remove these shortcomings and for farm development (Fig. 8).

## CONCLUSIONS

The end-user of animal amelioration is the consumer, namely the society, as animal improvement leads to an increased production and this translates to a larger offer of products at the level of national economy. The main activity of NAGB CAPRI-ROM is the official control of milk production, an activity without which there would be no selection and improvement of productions, thus no genetic progress.

Between 1995 and 2015 NAGB CAPRIROM managed to create the premises for an infusion of valuable genetic material and, implicitly, for a progressive increase of goat production performances.

In this regard, NAGB CAPRIROM facilitated the creation of breed nucleuses for Saanen, Apline, Murciana Granadina and Nubian, implementing thus improvement programs for local breeds by an infusion of genetic material from specialised breeds, with the support of reproduction biotechnologies, namely artificial insemination. The main technical instrument of distribution of the genetic material in the amelioration and perfecting of the goat productive lines is artificial insemination. NAGB CAPRIROM has the technical support and the specialists to support this activity, with an experience of over 800 artificial inseminations only between 2014 and 2015. NAGB CAPRIROM also participated in numerous scientific research projects that approached goat reproduction biotechnologies, systems of zootechnical management and offered support to the dissemination of their results.

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## DISPOSAL OF UNUSED MEDICINES RESULTING FROM HOME TREATMENT IN ROMANIA

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**Abstract**. Unused pharmaceutical compounds and their degradation products that are inappropriate disposed of are a source of pollution of the environment with negative impact on human health, as well as on solid and aquatic environment. The present study is an assessment of compliance with the law standards concerning the final disposal of pharmaceutical wastes resulting from home treatment among the population from western part of Romania, namely Bihor County. A total of 739 people were interviewed by using a questionnaire in order to evaluate their health status, the way of using and storing medicines and the disposal methods of the expired, unwanted or unused medication. The results revealed the level of public awareness regarding the legal disposal methods and destruction of the pharmaceutical wastes and the urgent need of public information campaigns (including educational campaigns for implementation of cost-effective and optimum unused pharmaceutical disposal strategies) that are nowadays totally insufficient and could be performed, for example, by the specific staff from the pharmaceus and as well by the presence of informative advertisements in pharmacies. Proper management of pharmaceutical wastes will mitigate the potential of the disposal problems and will have a positive impact on the environmental and human health.

Keywords: expired drugs, medicines disposal, pharmaceutical waste, education.

#### AIMS AND BACKGROUND

Pharmaceuticals acquired by people, by using a prescription or not, are potentially not totally used due to several reasons: the improved patient condition, adverse effects, and changes in dosage of the medicines, death, leading to expired, unwanted or unused medication<sup>1</sup>. Expired medicines belong to the 'chemical and pharmaceutical wastes' category<sup>2</sup> and these must be properly managed so that the risks to human health and the environmental impact are minimised<sup>3,4</sup>. The pharmaceutical waste resulting from home treatment is usually improperly disposed of around the world, which are more likely thrown away in the garbage bin, rinse down in the sink or in the toilet<sup>1,5–10</sup> and then can enter into the aquatic environment<sup>11</sup>.

<sup>\*</sup> For correspondence.

In two European countries, namely Malta and Republic of Ireland, were conducted surveys in 2012 to monitor the disposal of pharmaceutical wastes and excess medication and less than 10% of respondents returned them to pharmacies<sup>12</sup>.

The current legislation in Romania provided by the Law No 211/2011 on the waste regime<sup>13</sup> and the Order of the Minister of Health no. 1226/2012 established the technical rules on the management of wastes from medical activities and designated by the National Public Health Institute (a public institution with legal status, subordinated to the Ministry of Health) as the authority responsible for managing the national database regarding wastes from medical activities and approving on-site collection by separation on established categories of wastes from medical activities.

These regulations concerned only the specialised units ('pharmacies, drug stores, approved establishments for sale or distribution of medicines and pharmaceutical products; production units, warehousing and storage of medicines and biological products, pharmaceutical research institutes, preclinical units of universities and colleges of pharmacy, the National Agency for Medicines and Medical Devices') which are the main producers of pharmaceutical wastes.

Expired, unwanted or unused medicines resulting from home treatment represent a modest component of pharmaceutical wastes. Their elimination is regulated by the Order No 119/2014, which established the following: 'Expired medicines derived from the population will be submitted to the nearest pharmacy or pharmaceutical point in order to be disposed of and further destroyed by incineration'<sup>14</sup>. When they are managed improperly, unused or expired medicines reach the environment through water, soil and air, becoming a source of pollution to the environment and therefore a risk factor for the public health<sup>15,16</sup>. Thus, monitoring the way of management of them is particularly important<sup>10</sup>. This study is an assessment of compliance with the law standards concerning the final disposal of expired, unused, unwanted drugs resulting from treatment at home in Romania. The main objective consists in assessing the management of these types of wastes, but also focuses on the health of the population and on the education in this field, providing an overview of pharmaceutical waste management in Romania.

#### EXPERIMENTAL

The objective of this study was to ascertain information regarding the use, method of acquired and disposal of unused, expired or unwanted medicines in western part of Romania.

This study was conducted over a period of 12 months and was based on a completion of a questionnaire that assessed personal data, socio-economic indicators, health status, the use of medicines and final disposal of the pharmaceutical wastes resulted from home care. The questionnaires were distributed to 850 people, over 18 years of age, in five pharmacies (three urban and two rural pharmacies), located in Bihor county, during 2014/2015. 79 questionnaires were eliminated due to refusal/ omitting of the patients to answer to all questions and these were excluded from the analysis. 771 questionnaires were completed and remained to be analysed in this study. The personnel from these pharmacies informed their clients that they may return the unused, expired or unwanted medicines in pharmacies, where they will be collected and sent for incineration. All personal data were confidential. An outline of the questionnaire and the probably answers are presented in Table 1.

<b>1</b> 1	
Question	Possible answers
How often do you use medicines?	1. Daily; 2. Several times/week; 3. Once/ week; 4. Once/month
For what type of disease do you use to take medicines?	1. Chronic; 2. Acute; 3. Seasonal
How do you buy the medicines?	1. As indicated by a specialist (doctor/phar- macist); 2. Self-medication
What do you do with the medication if the treatment has changed, the medicine expired or you do not want to use it anymore?	<ol> <li>Throw it away in the garbage bin;</li> <li>I give it to acquaintances/relatives;</li> <li>I return it to the pharmacy</li> </ol>
What do you do with used sanitary waste (needles, syringes, swabs, vials, etc.)?	1. Throw it away in garbage bin; 2. Throw it anywhere
Do you consider that unused, expired or unwanted medicines and medical waste should be disposed of in special containers?	1. Yes; 2. No
Gender	1. Male; 2. Female
Area of origin	1. Urban; 2.Rural
Age (in completed years)	Numerical answer given
Educational level	1. Primary education; 2. Secondary educa- tion; 3. Higher education

Table 1. Content of the questionnaire: questions and possible answers given to participants

For statistical processing of the data SPSS 19 program was used.

## **RESULTS AND DISCUSSION**

In this study were analysed 771 questionnaires received from 47.2% male and 52.8% female given in five pharmacies in Bihor County. Informed consent was obtained from all individual participants included in the study. Most of the re-

spondents were in the age group 35–60 years (58.2%), 24.8% were over 60 years and 17.00% were in the age group 18–35 years.

We emphasised that the group of participants interviewed is representative for the population of Bihor County, in terms of demographic data. The participants lived in cities (61.3%) and in rural areas (38.7%). Educational levels are medium, 44.2% respondents have secondary school attended, 38.7% have elementary school followed, and 17.1% respondents have higher education. The frequency of use of pharmaceutical products is shown in Table 2.

Frequency	No	%
Daily	371	48.1
Several times/week	112	14.5
Once/week	11	1.4
Once/month	6	0.8
Several times/year	43	5.6
Once/year	52	6.7
I do not use drugs	176	22.8

Table 2. Frequency of use of the medicinal products

We noted that 48.1% of respondents are using medicines daily and nearly 22.8% do not use any medicine.

During the interview period, from 595 respondents who reported to take medicines, 77.5% had medication for chronic diseases, 5.4% referred to medicines taken for acute diseases, and 17.1% of participants are taken medicines for seasonal diseases (Table 3).

**Table 3**. Distribution of cases according to the diseases for which medicines are used by participants, in this study

Disease	No	%
Chronic	461	77.5
Acute	32	5.4
Seasonal	102	17.1

Self-medication was reported to be employed by 19.0% of respondents (Table 4), most of them reported to use medication for acute or seasonal diseases. When they were asked 'how the expired, unused, or unwanted medicines are disposed' of 95.3% of respondents thrown the medicines in the garbage, 4.0% gave the medicines to other people and only 0.7% returned to the pharmacy, as is shown in Table 5.

Table 4. Shale of Self-medication		
How do you buy the medicines?	No	%
Self-medication	157	20.4
Prescription on medication	614	79.6
Table 5. Methods used to dispose the medicines           Expired, unused or unwanted medicines	No	%
Throw away in the garbage bin	567	95.3
Throw away to other places	24	4.0
Return to a pharmacy	4	0.7

Table 4. Share of self-medication

The information which may educate the public and the legal regulations should be welcomed as civic behaviour and as well have to be implemented to avoid many accidents that occur due to actually disposal of pharmaceutical waste anywhere.

Regarding the need for special containers for disposing of unused/expired pharmaceuticals or waste facilities in public places, such as pharmacies, and malls, more than 80% of respondents agreed with this (Table 6).

Table 6. Need for special containers for disposing of unused/expired pharmaceuticals

Pharmaceutical products need special containers	No	%
Yes	623	80.8
No	148	19.2

In the last months, in Bihor and Arad counties were interviewed the pharmacists in ten, respectively in five different pharmacies, about their program of 'taking back' the expired/unused medicines from patients. All of them confirmed the availability and the active implementation of it, as it is an unequivocal obligation of the pharmacies in Romania to accept medicinal wastes for disposal. Just in one pharmacy in Oradea, Bihor County, was exposed a written information and also the pharmacists presented to patients the possibility that the pharmacists can take back medicinal wastes to be appropriate disposed of.

The pharmaceutical wastes management is an important feature regarding the human health and has a direct impact on environmental protection<sup>6,15,17,18</sup> especially following the increasing use of medicines by people around the world in the last decades. The methods used by respondents for the disposal of pharmaceutical wastes were evaluated by employing different survey instruments around the world (telephone questioning, written or mailed questionnaires, online surveys, interviews with patients)<sup>5,9</sup>. Usually, the most common method to dispose of the unused medicines is into the garbage, as it was reported in United Kingdom<sup>19</sup>, Kuwait<sup>20</sup>, Ghana<sup>21</sup>, New Zeeland<sup>5</sup>, Serbia<sup>8</sup>, Brasil<sup>22</sup>, Mexico<sup>23</sup>, toilet or sink in United States of America<sup>24</sup> and stored at homes<sup>25</sup> and/or returned to pharmacies in Sweden<sup>26</sup>. In the last years aroused the high concern about the proposals and

implementation of strategies for a proper management of pharmaceutical wastes in Europe and in several publications have been reported about these methods and about the public impact regarding this issue<sup>27</sup>. For example, in Croatia the pharmacists are obliged to collect unused pharmaceuticals from patients and to pay for their disposal<sup>28</sup>. A conclusion of the study was that the amount of collected of unused pharmaceuticals in Zagreb was below the European average annually. Therefore the authors suggested that more advertisement about the methodology of collection has to be performed to increase the public awareness in order to proper dispose of medicinal wastes and perhaps to eliminate the financial obligations of the pharmacies to pay for the disposal service<sup>28</sup>.

The present study was performed to evaluate the Romanian citizens about their health status, the use of medicines and the methodologies of disposal of unused, unwanted or expired pharmaceuticals. Majority of respondents, more than 95%, thrown the medicines in the garbage, as is shown in Table 3. Majority of the respondents, regardless of gender, age or education, during the interview, were not aware about a different management of this type of waste, compared to other household wastes that is well established and used both in urban and in rural areas. Therefore, most of the respondents have thrown the medicinal wastes into the garbage bin. This result has a consequence on environmental safety risk to human health as in the last years the yearly sales of medicines increased around the world<sup>29</sup>. The information which may educate the public and the legal regulations should be welcomed as civic behaviour<sup>30</sup> and as well have to be implemented to avoid many accidents that occur due to actually disposal of pharmaceutical waste anywhere.

Following the analysis of data obtained, there is a modest interest of the population and also a low level of information about how the pharmaceutical wastes should be disposed of/used later. Therefore, it is mandatory to perform more information campaigns to educate the population about a secure storage and as well about safe and proper medication waste disposal, mainly in order to reduce the risk of accidental medication exposure and toxicity especially for children and pets. One alternative to attain the excess<sup>31</sup> of medication at patient homes is to reduce the prescription of quantity of medicines prescribed especially for as-needed orders<sup>32,33</sup>.

A recent study conducted after a public education campaign in Bihor County, showed a significant increase to 87.3% in 6 months of the number of patients who have returned the expired and/or unused drugs to pharmacies<sup>34</sup>. This showed that the pharmacists have a wide audience and they can actively be involved in the education of patients regarding the use and disposal of expired/unwanted medicines.

Our opinion is that a permanent program of public awareness has to be implemented for the safe and secure disposal of the pharmaceutical wastes, and one simple and cheap example is to present in each pharmacy a written advertisement about the possibility to return the medicinal waste in the pharmacies. Therefore, the implementation of special purpose containers for the assembly of the pharmaceutical residues, their location in pharmacy buildings is useful and necessary, especially given by the high increase in the consumption of medication in recent years, in Romania.

#### CONCLUSIONS

It is necessarily required a proper management program of disposal of pharmaceutical wastes from home treatment in Romania. This can be achieved primarily through proper information of population for a suitable safe and secure disposal of medicinal wastes and by increasing the interest of public on the protection of the environment. It was noticed that if the optimum information about the proper method of disposal of unused/unwanted medicines is performed, especially by pharmacists who are in near contact with the patients, the population is open to accept and to use this disposal practices of pharmaceutical wastes. Also, in order to facilitate the recovery of the sanitary wastes it would be very useful to implement their collection in proper containers in pharmacies and in shopping malls, considering that in all of these commercial centres exists a pharmacy, and the number of clients is continuously growing.

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## IMPROVEMENT TO GREEN IN-COUNTRY TOURISM CONDITIONS: A CASE OF THE CZECH RECREATION LAKE PASTVINY

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**Abstract**. To improve quality of waters in reservoirs used for in-country sustainable recreation, namely those recognised as European bathing waters, is one of the current and near-future problems of relevant policies. A model for an ex-ante assessment and improvement of current policies in the field has been developed and is currently being tested for typical practical situations. The model serves to find cost-effective solutions in complex situations where there is a potential for the so-called multiple coalition projects and it introduces economic aspects to early stages of decision making. The paper brings an application of the model to the recreation Lake Pastviny, which is located in east Bohemia. The analysis indicated a potential saving of annualised abatement costs of 6% in the case that half of the municipalities located in the lake watershed would join certain coalitions, i.e. they would build joint wastewater treatment plants and the rest of the municipalities would stay with building individual wastewater treatment plants.

Keywords: sustainable recreation, water pollution reduction, environmental protection.

## AIMS AND BACKGROUND

The most pressing problems of water pollution in major river basins have already been solved (not only in the Czech Republic) by building wastewater treatment plants (WWTPs) in municipalities over 2000 inhabitants<sup>1</sup>. There are still important problems requiring solutions in the field of wastewater treatment, which are appearing on the horizon including Central and Eastern European countries. To improve quality of waters in reservoirs used for in-country sustainable recreation is one of them. This is particularly important for countries and regions with a long distance to a sea. Serious attention to creating and improving conditions for recreation is paid by the EU. Relevant for our paper is EC Bathing Water Directive<sup>2</sup>, which concerns the quality of bathing water. Information about the quality

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of these waters is published periodically by the European Environmental Agency; the latest update is available for 2015 (Ref. 3).

The interest of scholarly literature reflects the importance of the topic in practical policies. A number of authors analyse and point out the complexity of the issue of eutrophication of surface water from the natural science perspective<sup>4–7</sup>. Another bundle of the literature deals with the economic context of the problem<sup>8–10</sup> and others.

A model for an ex-ante assessment and improvement of current planning and policies in the field (having mostly the form of various water basin plans) is being developed and currently tested for typical practical situations by authors of this paper. The model serves: (i) to improve understanding of the multidisciplinary aspects of the problems and their solutions and contribute to finding cost-effective solutions in complex situations where there is a potential for multiple (so-called coalition) projects; (ii) to test various planned policy designs when such solutions could be achieved in situations with information asymmetry between state authorities and polluters-investors when a financial contribution is being negotiated<sup>11</sup>. It is typically relatively costly to reduce pollution generated by small polluters and financial support from governments (at various levels) is important. The paper aims to present an application of the model to the recreation lake Pastviny in the Czech Republic.

#### EXPERIMENTAL

*Lake Pastviny – situation description.* Lake Pastviny is located in east Bohemia, by the Polish border. It was built in response to disastrous flooding in the Orlice river basin, occurring in 1840, 1846, 1862 and 1888. The reservoir was constructed in 1933–1938. The watershed area is 181 km<sup>2</sup>, including 70 km<sup>2</sup> in neighbouring Poland (Fig. 1). The hold-back length is 5.7 km; the max. inundated area is 110 ha; the cubic capacity of water held back is 1284 mil m<sup>3</sup>; and the maximum depth is nearly 26 m.

The purpose of the reservoir is partial protection<sup>12,13</sup> of the area situated by the Divoka Orlice river lower downstream from large floods, provision of a minimum residual flow in the river downstream of the hydraulic structure, and energy utilisation of the water outflow from the reservoir in a peak-time hydropower plant. As for recreational uses, Lake Pastviny is significantly used for various forms of recreation and water sports on its surface, as well as sports fishing, and wave flow enhancement in the river downstream for paddling sports. A plan is also in place for controlling the water outflow from the reservoir in the event of water cleanness accidents, clearing the river bed in the event of icing, etc.

As concerns the water quality development, the quality was very good in the early years of the reservoir, as attested by the initial fish association in the bullhead-trout domain. The housing amenities gradually improved with the advent of new settlers in the Czech part of the Lake Pastviny watershed. Water mains were built and contaminated wastewater was increasingly discharged via the gradually constructed sewerage network into watercourses emptying into the Divoka Orlice, thus the water reservoir.

The increasing contamination due to wastewater, particularly in summer, has been caused, among other factors, by increasing recreational development in the near vicinity of the lake with problematic wastewater disposal strategies. The adverse effects of this development were long ignored.

High quantities of the *Aphanizomenon* genus cyanobacteria (3000 individuals/ ml) were identified back in 1960 (Ref. 12). A Soviet Army garrison was stationed in Klasterec nad Orlici after 1968. Poorly treated wastewater introduced large quantities of nutrients to the lake, causing after 1986 development of mighty water bloom and seriously impairing conditions for recreational bathing. The water quality improved significantly with the departure of the garrison after 1989. However, considerable presence of autotrophic organisms (more than 13 000 individuals/ ml) was identified again in the summer season of 1996, including cyanobacteria (genera *Gomphosphaeria* and *Anabaena*).

Pastviny water reservoir is annually included on the list setting out surface water bodies used for bathing (bathing waters) pursuant to European Directive No 2006/7/EC (Ref. 2). In this context, the regular conclusion is that the reservoir poses a risk of cyanobacterial presence and the trend, unless resolved, will probably have a negative tendency. The main unsatisfactory indicator in Pastviny reservoir is total phosphorus. The excessive introduction of this nutrient and its retention result in massive cyanobacterial blooms in the water in summer. The model presented works with the total phosphorus indicator, but certain aggregate water quality indices, for example, could be used if necessary. For an example of such an index<sup>14,15</sup>.

A signification portion of the phosphorus – the one that is best utilised by the autotrophic organisms according to the latest surveys – originates from anthropogenic sources in the form of untreated sewage. Technical solutions have to focus on these in particular, i.e. deal with contamination originating from smaller municipalities scattered alongside a long stretch of the Divoka Orlice river. In addition, increasing numbers of extreme situations (more profuse and shorter rain events, fewer regional rains; more periods of drought) opens a discussion about effects on the reservoir water quality.

The watershed consists mostly of forests and pastures; arable land is represented to a lesser degree. The phosphorus washed out of farmland areas is typically in a form not accessible to cyanobacterial growth; therefore, the above-mentioned point sources – untreated or poorly treated sewage from municipalities – are considered a much more important source of phosphorus. Farming is viewed instead as a major producer of nitrates in waters and a significant source of sediments released by erosion processes in the watershed.

The technical solution to reducing introduction of phosphorus into the reservoir and decelerating its eutrophication consists in construction of new WWTPs and sewerage, and upgrading of existing systems in adjacent municipalities.



**Fig. 1**. Study area – simplified map of Lake Pastviny watershed (Source: Povodí Labe, state enterprise, Hradec Králové)

*Reverse combinatorial auction model.* We used reverse combinatorial auction theory as a theoretical background for the model serving optimal solution quantification. Reverse combinatorial auction is a specific case of combinatorial auction<sup>16,17</sup>. In this case, there is one buyer and several sellers. In our case, the government solving the lake pollution problem and contributing financially to the costs of the projects is the buyer. Municipalities offering building or renovation of WWTPs are

the sellers. The municipalities have a chance to find coalitions, i.e. to build joint WWTPs, if it is more efficient than to build individual WWTPs.

The criterion for optimisation is minimisation of the projects costs. The optimum solution is computed using a mathematical model. More details about the model, including its mathematical description, are reported in Refs 18 and 19.

*Scenario description and data*. A scenario for how to achieve the environmental standards required for bathing water in Lake Pastviny was developed by specialists from the Elbe river basin management enterprise (Povodí Labe, s.p.). Building individual municipal wastewater treatment plants or building joint treatment plants for several municipalities were considered in the scenario description. The scenario suggests installing progressive membrane technology and installing a third stage of cleaning (adsorption filter media, nanotechnologies, or other modern technologies). The total estimated efficiency of phosphorus removal in this scenario is 98%.

The scenario consists of 24 individual projects (i.e. each of the 24 municipalities builds its own WWTP) and 131 multiple-coalition projects (i.e. situations in which two or more municipalities build joint wastewater treatment plants). Since the number of theoretically possible projects is huge, only acceptable ones were selected by the specialists. These were 32 two-member coalitions, 38 three-member coalitions, 38 four-member coalitions, and 22 five-to-eight-member coalitions.

The abatement costs in the form of investment and operating costs were assessed by the specialists. Average annual costs (AAC), which convert the investment costs to an annual basis, were calculated based on the data. The same formula as in the case of Vrchlice drinking water reservoir<sup>18</sup> was used with a discount rate of 5%. Table 1 shows the list of municipalities introduced to the computations and it is the list of individual projects at the same time. The list of multiple-member coalitions and the assessed costs are available from the authors.

Municipality	Code	Municipality	Code	
Lasówka near Trčkov	M1	Bartošovice v Orl.hor.	M13	
Lesica	M2	Neratov	M14	
Mostowice near Orl. Záhoří	M3	Orlické Záhoří	M15	
Rudawa	M4	České Petrovice	M16	
Niemojów	M5	Mladkov	M17	
Poniatów near Neratov	M6	Petrovičky	M18	
Čihák	M7	Vlčkovice	M19	
Jedlina	M8	Kunvald	M20	
Klášterec nad Orlicí	M9	Končiny	M21	
Lhotka	M10	Zaječiny	M22	
Zbudov	M11	Nekoř	M23	
Pastviny	M12	Údolí	M24	

Table 1. Individual municipality wastewater treatment projects

RESULTS

The analysis has revealed the following optimum (cost-effective) solution:

(a) 5-member coalition: M9 (Klášterec n.O.) + M10 (Lhotka) + M12 (Pastviny) + M16 (České Petrovice) + M18 (Petrovičky);

(b) 3-member coalition: M5 (Niemojów) + M13 (Bartošovice v O.h.) + M20 (Kunvald);

(c) 2-member coalition: M3 (Mostowice near Orl.Záh.) + M15 (Orlické Záhoří);

(d) 2-member coalition: M11 (Zbudov) + M21 (Končiny);

(e) municipalities with individual WWTP: M1, M2, M4, M6, M7, M8, M14, M17, M19, M22, M23, M24.

Costs (AAC):

Optimum (cost-effective) solution: 45 083 thousand CZK (= 1670 thousand EUR)

Individual projects: 47 913 thousand CZK (= 1775 thousand EUR)

The municipalities and the optimum coalitions are given in Fig. 1.

#### DISCUSSION AND CONCLUSIONS

When looking at the computed results, we can recognise that, to achieve the theoretically optimal solution, 12 municipalities should build joint WWTPs. There is one group of five municipalities, one group of three municipalities, and two groups of municipalities are supposed to create coalitions of two members. The rest of the municipalities should build their own individual WWTPs.

When comparing the costs of individual solutions (i.e. the situation where each of the 24 municipalities would build its own individual WWTP), the saving is 6% of the annualised abatement costs.

In the space between the optimum solution and the individual solutions there is high number of feasible coalition structures. For a support of decision making we therefore propose sensitivity analysis based on analysing coalition structures for specific cost levels. This means that the space between costs for individual projects and costs for the first best solution is divided to several levels corresponding to policy decisions about potential (politically acceptable) deviation of the practical program from the (theoretical) first best solution.

In our case we work with 4 costs levels. Level 1 is defined as the cost for the first best (optimal) solution, level 4 as the cost for individual projects. Other levels are always about 20% higher than the previous level, where the second level is the politically acceptable one.

The solutions for the cost levels are computed from the basic model<sup>18</sup> adding a constraint:

$$\sum_{h=1}^{m} \sum_{C \subseteq R} c_h(C) y_h(C) \ge L_i$$
(1)

where  $L_i$  are the cost levels, i = 0, 1, 2, 3;  $L_0$  – the cost level for individual projects;  $L_3$  – the cost level for optimal solution. The difference  $L_0 - L_3$  is divided to three parts and cost levels are gradually reduced from the level  $L_0$  to  $L_3$  to see the progress in coalition structures.

The results of the costs levels computation are shown in Table 2. We can see that the number of multiple-member coalitions decreases with the increase of the cost levels.

Level	Costs (ths.CZK)	Structure description	Saving AAC (%)
1st level = optimum solution	45.083 u-	See the results presented above	6
2nd level	46.027	<ul> <li>4-member coalition: M9+M12+M13+M20</li> <li>2-member coalition: M3+M15</li> <li>2-member coalition: M2+M5</li> <li>2-member coalition: M17+M19</li> <li>14 individual projects: M1, M4, M6, M7, M8, M10, M11, M14, M16, M18, M21, M22, M23, M24</li> </ul>	4
3rd level	46.980	3-member coalition: M11+M20+M21 3-member coalition: M12+M17+M19 2-member coalition: M9+M10 2-member coalition: M16+M18 14 individual projects: M1, M2, M3, M4, M5, M6, M7, M8, M13, M14, M15, M22, M23, M24	2
4th level = in dividual pro ects solution		24 municipalities M1-M24 (see Table 1)	0

Table 2. Structures computed for the selected costs levels

In addition, it follows from the discussion of the above results for Pastviny reservoir (Personal communication with experts from Povodí Labe, s.p.) that implementation of measures aimed at reducing the introduction of phosphorus should proceed from that part of the watershed that closely surrounds the water reservoir. The introduction of phosphorus from the upper, more distant part of the watershed is currently less significant, but it would increase gradually with development in the area if the situation was not handled, and would negate the results of the whole process. It must be noted that the ecological efficiency has to be very high in order to successfully resolve the reservoir water eutrophication issue.

Learning about the (theoretical) optimum solution, i.e. achievement of the target water status typically required by legislation and decision-making entities themselves, in the least costly way<sup>20,21</sup> is undoubtedly beneficial. In practice, however, it is very often a matter of not implementing very ineffective options. There, too, a comparison with the hypothetically most effective option may help.

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## DEVELOPMENT OF THE CULTURAL HERITAGE TOURISM BY REHABILITATING THE SANCRAI CASTLE, ALBA COUNTY

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**Abstract**. This paper approaches the possibilities of rehabilitation and the changes of using the old castles in Transylvania. The study refers to the Sancrai Castle (Banffy Castle), located in Sancrai, Aiud, Alba County. The building has undergone great internal and external transformations, which most likely deformed the initial appearance, but especially led to major degradation, due to the lack of professionalism in interventions. The castle restoration and re-functionality were run by the Alba County Council heritage experts, who paid special attention to the authentic elements that survived the 'historic storms'. They highlighted details that have been preserved untouched throughout the years and have opened new spaces for the public, such as the attic, which became today exhibition area, unused before the rehabilitation. A detail of the original architecture of the castle was preserved on the staircase leading into an octagonal tower. It has a wrought iron railing of an extremely elaborated floral design, very decorative, authentic and representative of the Art Nouveau style. This element is found in the new logo of the Sancrai Castle Cultural Centre, a symbol that highlights the local tradition and promotes a feature which is dedicated to elegance, culture and excellence.

Keywords: Sancrai Castle, restoration and re-functionality, cultural heritage tourism.

## AIMS AND BACKGROUND

Tourism and culture have always been closely related in Europe, as Europe has always been an important destination for those attracted to its rich culture and historical legacies. Moreover, European cultural heritage is one of the oldest and most important heritages producing tourism. Cultural tourism can be linked to the motivation for access cultural goals such as travels for art festivals or other cultural sites or events, as well as trails for the study. In a broader sense, any form of tourism can be defined as cultural tourism if it manages to satisfy the basic human need for diversity<sup>1</sup>, tending to increase the individual cultural awareness, discovering new knowledge, experiences and encounters. Cultural tourism offers the opportunity for intercultural dialogue through the discovery and appreciation of

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cultural diversity. In Transvlvania, this form of tourism increases from one year to the next, through the diversity of cultural objectives available to the tourists visiting our area. A landmark, worthy to consider is Sancrai Castle (Banffy Castle). It is located in the village of Sancrai, having the height regime of basement + ground floor + attic, namely, basement + ground floor + mansard after rehabilitation, and it is one of the Alba county historic monuments. The Castle belonged to Kemeny family and was built in 1805. In 1890 it lapsed to the Banffy family, which has led to its expansion in two steps. Built after a Renaissance model, perfectly symmetrical towards the access axis, the Castle has gone through an initial stage of changes in 1890, when the South-western body and its tower were built, which are of a late Baroque stylistic expression, slightly eclectic, with neoclassical touches. In 1947, the building became asylum for children with disabilities, and it underwent internal and external transformations, which most likely deformed the initial appearance, but mostly led to major degradation, due to the lack of professionalism in intervention. This paper aims to present the castle rehabilitation technology<sup>2</sup> (removing the moisture from the castle walls and basement foundations). The castle restoration and re-functionality are run by the investment beneficiary, which is Alba County Council, respectively the general designer, S.C. Megavox Project S.R.L. Cluj Napoca. The beneficiary of the investment pays special attention to the authentic items that survived the 'historical storms'.

#### EXPERIMENTAL

The general objectives of the intervention works on the Castle are: removal of moisture from the walls and foundations, reconstruction of its interior and exterior finishes and restoration of historical woodwork, replacement of destroyed woodwork by carpentry replicas, interventions on the ceiling over the first floor, replacement of the degraded structural elements, replacement of the roof covering, and redesign of equipment. These were the issues aimed to during the restoration works at the castle of Sancrai. The castle is currently functional. The cultural goods, the historic monuments and the modern buildings are ultimately more or less porous materials, more or less chemically active materials<sup>3-6</sup>, being the support of water migration as salt solutions. The danger of any kind of porous material to degrade while being covered by salt solutions is a widely known natural phenomenon. On the occasion of water evaporation, the crystallisation of salts contained involves an increase in the volume of pores and, as a result, the progressive destruction of the materials, from the surface to their depth. At the same time, humidity higher than 3–5%, considerably reduces the thermal insulating properties of various materials, stimulates the development of certain vegetable organisms (Merulius lacrimans, etc.) or of bacteria, which make the buildings affected unliveable in. In the fight against the moisture penetration in construction, the causes, the effects and the

methods of control should always be considered in close interdependence<sup>2</sup>. For determining the degree of humidity, the Control Company moisture-meter has been used, in four points on different heights ranging from 20 cm from the basement limestone level up to 1.60 m.

## **RESULTS AND DISCUSSION**

#### CAUSES

The causes which have led to excessive moisture in the castle basement walls are different:

- partial degradation of the installation of storm water discharge (gutters and down-spouts) (Fig. 1);

- the lack of vertical waterproofing on the exterior walls of the basement;

- through spouts, the storm water from the roof is discharged on the building and at its base (Fig. 1);

- poor ventilation of the underground space (Fig. 2).



Fig. 1. Degradation of the installation of storm water discharge



Fig. 2. Poor ventilation of the underground

#### EFFECTS

The effects of excessive moisture in the walls and of the fact that no measures have been taken to combat the humidity can be seen in Figs 3 and 4. Figure 3 illustrates the chemical effects (efflorescent), while Fig. 4 shows the biological effects (black crust). The two images were taken in various locations of the castle's basement, the wall deterioration is different due to the wall's finish, namely due to the chemical substances contained by the water that passes through the wall.



Fig. 3. Chemical effects

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Fig. 4. Biological effects

During the Castle rehabilitation, the walls moisture was determined through measurements made in some areas where the humidity was more obvious. The measurements were carried out in four points on different heights ranging from 20 cm from the basement limestone level up to 1.60 m.

The values determined were centralised in Table 1. At the same time, the atmospheric humidity in the basement was established with the hygrometer. The values recorded are very high (68%), the determination inner temperature being of 17°C.

Н	Measurements point			
(cm)	1	2	3	4
20	10.9	20.3	15.9	17.1
40	19.7	18.7	19.6	15.7
60	15.9	19.4	20.8	16.8
80	20.5	19.7	18.9	17.1
100	16.2	18.1	19.2	19.2
120	15.6	16.9	15.5	14.3
140	15.2	18.3	14.3	15.7
160	16.2	15.9	14.1	14.9

Table 1. Walls humidity (W, %)

The above may lead to the conclusion that the humidity in the Castle walls determines a very high humidity in the basement as well. The humidity values of the basement walls show no constancy by height (H) (the humidity values usually decrease from bottom to the top) featuring very large fluctuations as measured values. This is influenced by the fact that the structural walls in the basement are made of rubble work on the exterior and of full brick on the inside, while the foundations are continuous under the structural walls and are made of rubble stone.

Maintaining this humidity in the walls of the castle's basement leads to the degradation of the materials they are built of (which could have repercussions on the building stability), creates a very high atmospheric moisture, stimulates the development of certain vegetable organisms or bacteria that could lead to the failure of the space functionality by considerably reducing the walls' thermal insulation properties. Therefore, it is necessary to apply measures for rooting out the humidity from the castle basement walls.

## MEANS OF INTERVENTION FOR ERADICATION THE HUMIDITY IN THE BASEMENT WALLS

The technical documentation originally stipulated for the chemical method, which consists in waterproofing the walls by different water-repellent solutions that can be introduced into a porous environment in different ways depending on the execution technology<sup>5,6</sup>. This drainage method can encounter difficulties if the product injected does not evenly penetrate the surface we want to waterproof. At the same time, it should be noted that most of the times the walls we want to treat are very old and may have cracks, interior cavities or may be made of different materials and very thick (in our case, brick and stone masonry), or may have air spaces which impede the material injected gravitational or under pressure<sup>5</sup>. This obstruction reduces the material possibility to expand in capillarity and to create a horizontal barrier so that the whole treatment may be compromised. Even the smallest untreated points allow moisture to pass above the horizontal level created by the holes made in the wall. As a result, after a more thorough analysis, we had to abandon this way of intervention and to approach another chemical method, namely the use of special binders that dry the wall without stopping the humidity rise through the wall, but encourage the moisture evaporation through the wall<sup>2</sup>. At the same time, a brick-'breath' floor was executed in the basement, namely a drain trench was dug in the castle's outer perimeter, with vertical insulation7.

*Drainage plaster*. For applying the system of drainage plaster on the basement interior walls, the following steps were completed:

Preparation of the substrate:

- the old plaster is completely removed;
- the joints are widened to 20 m;
- a mean support layer is provided for the new plaster coat;
- the dust and the binder remains are wiped away.

Applying the drainage system by using the HASIT products:

- Hasit Saniergrund drain ground coat;
- Hasit 205 drain spritz;
- Hasit 208 drainage and equalisation plaster;
- Hasit 200 drainage plaster;
- Hasit 212 drain polishing plaster coat;
- Husit PI 203 OKOSIL silica interior paint (Fig. 5).



Fig. 5. Interior rehabilitation

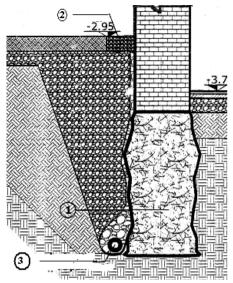
*Brick – 'breath' floors*. They were fitted in the basement on a 20 cm thick sand bed disposed on a capillary breaking layer made of 20 cm thick compacted gravel, of high and medium grain. The interior brickwork was sheathed by an alveoli protective foil that will be installed from the digging level up to the floor level. The brickwork was also protected by a layer of pure shifted gravel, 20 cm wide and 20 cm thick, perimetrically disposed in the rooms<sup>7</sup> (Fig. 6).

*Execution of a drainage ditch around the outer perimeter of the castle with vertical insulation*<sup>2</sup> (*Fig. 6*). A drainage ditch role is to protect the castle from the waters that for various reasons would reach its perimeter area and to steer them towards the septic tank in the vicinity of the castle.

Together with the rehabilitation works, the entire rainwater collection system has been redone, as well as the gutters and the down-spouts.

For the construction of the drainage channel, a ditch was dug perimetral to the castle up to the foundation base. A Hasit 203 foundation plaster layer was applied over the foundation for the support equalisation, in two layers of 14 mm thick. The surface obtained was isolated with two 1 mm layers of Hasit Sockelflex Optiflex flexible mortar. Over the insulating mortar there was positioned a protection foil. A Ø 110-PVC drainage tube perforated at the top was put into the dig and it was protected by a layer of granules sort with a diameter larger than the holes in the drainage tube, respectively, the geo-textile represented in Fig. 6 (Refs 7 and 8). On the exterior of the building there was done a cubic stone – 'breathing' side-walk.

Following the theoretical experience of the methods studied and of the works performed, one may conclude that the success of these works is determined by the strict compliance with the construction technologies, by the choice of the materials and methods to be used according to the specific conditions of each structure.



#### Fig. 6. Execution of drainage

1 – foundation, Hasit 203 foundation plaster, insulating material – flexible mortar, protection foil, large grain sort, geo-textile cloth, coarse gravel, compacted soil; 2 – pure shifted gravel for masonry protection; 3 – drainage pipe  $\emptyset = 110$  mm (PVC tube perforated at the top)



Fig. 7. Final restoration

The high levels of humidity are due to the faulty maintenance of the building, but also to the inappropriate interventions, and not least, due to the postponement of the castle rehabilitation.

Maintaining an excessive moisture leads to the emergence of micro-organisms: bacteria, moulds, with adverse effects on human health. In the areas affected by humidity, higher heat losses occur through walls and the masonry degradation is much more pronounced. All these can lead to the building loss of stability. The intervention methods applied are based on the ventilation of walls and floors, respectively on the castle protection from any rainwater infiltrations. The success of these works (Fig. 7) is determined by the strict compliance with the construction technologies, by the choice of the materials and methods to be used according to the specific conditions of each structure.

### CONCLUSIONS

This paper shows the importance of rehabilitating cultural monuments, old castles and other important cultural objectives. These can be entered in the tourist circuit and can be a huge advantage for the region. The Sancrai Castle (Banffy Castle), located in Sancrai, Aiud, Alba County was rehabilitated by the Alba County Council heritage experts, who paid special attention to the authentic elements that survived the 'historic storms'. They highlighted details that have been preserved untouched throughout the years and have opened new spaces for the public, such as the attic, which became today's exhibition area, unused before the rehabilitation. A detail of the castle original architecture was preserved on the staircase leading into an octagonal tower. It has a wrought iron railing of an extremely elaborated floral design, very decorative, authentic and representative for the Art Nouveau style. This element is found in the new logo of the Sancrai Castle Cultural Centre, a symbol that highlights the local tradition and promotes a feature which is dedicated to elegance, culture and excellence.

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# **STUDY REGARDING THE SOLUTIONS FOR SUSTAINABLE TOURISM DEVELOPMENT IN SIGHISOARA**

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Abstract. Sustainability is one of the most debated aspects of tourist destinations. Given Romanian poor performance in this respect, tourism and sustainability are analysed in a case study covering Sighisoara (Romanian single urban destination listed by UNESCO World Heritage Sites). Several aspects concerning the main issues linked to the destination cultural heritage have been identified and discussed. Considering the significant increase of both tourist supply and demand within this city, various indicators concerning capacity management and tourist flows have been calculated with the purpose of investigating the destination carrying capacity. Given the very high growth rates of the travel and tourism industry in this city, it becomes obvious that authorities must consider developing and adopting sound destination management strategies to ensure the sound development of sustainable tourism. In this respect, after having realised a thorough analysis of the city cultural potential and of its tourist activity and infrastructure, the authors suggest a set of recommendations regarding the adoption the most appropriate destination management strategies for sustainable tourism development in this endangered UNESCO World Heritage Site. The research method relies on case-study writing and consists of a combination of qualitative primary data collection method and of secondary data analyses. Induction and deduction have been used with the purpose of providing a comprehensive image of the destination. The paper most important contribution is the set of destination management measures suggested for the further development of Sighisoara tourism.

*Keywords*: sustainability, tourism, Sighisoara, UNESCO World Heritage Sites, carrying capacity, destination management strategies.

### AIMS AND BACKGROUND

Considering the current state of one of Romanian most important tourist attractions, the inhabited Citadel of Sighisoara, the present paper analyses the perspectives of sustainable tourism development in this urban destination. The paper most important contribution is related to the elaboration of a set of recommendations concerning potential solutions for sustainable tourism development in the endangered UNESCO World Heritage Site Sighisoara. According to the United Nations World Tourism Organisation (UNWTO) (Ref. 1) definition, sustainable tourism implies a balance between the environmental, economic and socio-cultural aspects

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of tourism development in order to grant host-destinations long-term benefits. Further, sustainable tourism<sup>1</sup> should: guarantee the optimal use of environmental resources, with the maintenance of essential ecosystems and the conservation of biodiversity; respect and support socio-cultural authenticity, preserve built and living cultural heritage, and contribute to cross-cultural understanding and tolerance; lead to the achievement of long-term socio-economic benefits and ensure their fair distribution among all community stakeholders (including stable employment and income-earning opportunities, social services and the reduction of poverty). In other words, the UNWTO (Ref. 2) explains that sustainable tourism 'meets the needs of present tourists and host regions while protecting and enhancing opportunities for the future'; this requires to manage all resources in such a way that economic. social and aesthetic needs are fulfilled with the preservation of cultural integrity, essential ecological processes, biological diversity, and life support systems. Further, this raises the need to develop a destination management organisation (DMO) (Ref. 2) with the main role to implement management practices and to create plans designed for controlling tourism development and activity at destination level. This is especially needed in Romania<sup>3-10</sup> due to its very poor performance in terms of sustainability and sustainable tourism<sup>3-12</sup>, and particularly in Sighisoara<sup>13-16</sup>, destination that is today considered endangered, because of the improper manner in which its built-heritage is managed and used. For a better understanding of the destination, Sighisoara tourism is discussed relying on the available statistical data<sup>17</sup>, concluding that due to its significant growth, the adoption and implementation of destination management strategies is compulsory.

## EXPERIMENTAL

Given the subject of this paper, the most appropriate research method is that of case-study analysis. Moreover, the paper consists of a combination of qualitative primary data collection and of secondary data analyses. By collecting and analysing similar data, the same research can be further applied on other destinations from Transylvania, respectively Romania. Even more, comparative international investigations can be also developed based on the present method, with good chances to provide useful examples of good practices at European level. The villages with fortified and citadel churches, located in Sighisoara surrounding Saxon area (in Southern Transylvania) can be some of the beneficiaries of the reproduction of the present research.

## **RESULTS AND DISCUSSION**

From the perspective of its natural and cultural heritage, Romania is an attractive tourist destination but, due to its poor access infrastructure, it still misses to capitalise on its generous resources and to adequately develop its travel and tourism

industry. Moreover, as the data collected in Table 1 reveal, Romania ranks very low in terms of both environmental sustainability and of sustainable travel and tourism development. The most recent Travel and Tourism Competitiveness Report<sup>3</sup> places Romania on the 46th position from among 141 analysed countries for environmental sustainability. The same report<sup>3</sup> reveals that Romania ranks 79th in terms of natural resources, while the extent of its cultural resources and business travel place it on the 49th place worldwide. In the overall tourism competitiveness index the country ranks 66th. It may be concluded that the destination's cultural resources provide valuable sources of tourism competitiveness. Romanian overall travel and tourism competitiveness has improved between 2007 and 2015 by 10 positions but the country has dropped towards the end of the global rankings in the cases of some of the most important sustainability-related indicators: Government prioritisation of sustainable Travel and Tourism (T&T)/Sustainability of T&T industry development (rank 122 of 141 in 2015) or Stringency of environmental regulations (rank 90 of 141 in 2015). This situation is closely related to the poor performance of Romania in two cases: Government prioritisation of the T&T industry (rank 117 of 141 in 2015) and Effectiveness of marketing and branding (rank 120 of 141 in 2015). Slight improvements can be noted in two other cases: Quality of the natural environment (rank 96 of 141 in 2015) and Enforcement of environmental regulation (rank 77 of 141 in 2015); still, these two indicators are far from registering satisfactory values. At European level, Romania scores really low, ranking among the least competitive European destinations, close to the end of the list (32nd of 37 countries for overall competitiveness at European level).

Travel and tourism competitiveness (T&TC)	2015	2013	2011	2009	2008	2007
Travel & Tourism Global Competitiveness	66	68	63	66	69	76
Number of World Heritage sites/cultural sites	34	33	29	26	30	30
Number of World Heritage natural sites	43	45	43	40	39	
Environmental sustainability	46	58	50	37	51	
Government prioritisation of sustainable T&T/ Sustainability of T&T industry development	122	129	118	121	122	115
Stringency of environmental regulation	90	109	71	60	72	74
Effectiveness of marketing and branding	120	123	118	119	118	111
Total number of countries	141	140	139	133	130	124
Romania T&TC at European level	32	35	34	34	32	36

Source: Authors elaboration based on the Travel and Tourism Competitiveness Report<sup>3-9</sup>.

One of the most controversial aspects of Romanian tourism in related to the country branding as a tourist destination. Several (expensive) campaigns<sup>10</sup> have been designed and implemented incoherently over the past two decades but their

effects are far from meeting the allotted budgets. Currently Romania is promoted abroad, except for North America, under the tagline 'Explore the Carpathian Garden!'11. Another slogan is used on the American and Canadian markets, namely: 'Romania. Natural and Cultural'12. Although frequently changed, most branding attempts have concentrated on Romania natural and cultural heritage. Special attention has been granted to the Transylvanian medieval Saxon heritage. Established more than 850 years ago, the Saxon Citadel of Sighisoara was included in the UNESCO World Heritage Sites in 1999. It is the single urban UNESCO heritage site of Romania and it is one of the most attractive destinations of Transylvania. Moreover, it is the only medieval citadel that is still inhabited in South-Eastern Europe. Despite its huge value and tourist potential, the citadel is threatened to be excluded from the UNESCO World Heritage Sites, and to be listed among the world's endangered sites, mainly due to the improper renovation policies and practices. This is an unfortunate consequence of Romanian poor performance in terms of sustainability<sup>3</sup> (Table 1). In the context of the increasing popularity and attractiveness of the World Heritage Site Sighisoara, driven by the desire to prosper quickly, several entrepreneurs and/or owners of properties located in the Citadel have renovated and, even, modified some properties, ignoring the use of appropriate techniques and materials<sup>13,14</sup>. Obviously, this was possible due to the poor implication of the City Hall in the preservation of its highly valuable heritage. Moreover, as the representatives of TransylvaNET<sup>15</sup> point out, Romanian citizens and public authorities must become aware of the great value of the country cultural heritage and, before all, must contribute to its conservation and appropriate valorisation. Despite its rich and highly valuable cultural heritage, when it comes to the management of these resources, Romania ranks last among European nations<sup>13</sup>. Offering useful and valuable examples of problems and good practices, K. Creosteanu<sup>16</sup> synthetises the main aspects related to cultural heritage protection. Thus, cultural heritage must be protected because: it is our home, it proves continuity, and makes us/destinations well-known. Further, it must be protected against: forgetting, irresponsible usage or misuse, and public authorities ignorance and indifference. Owners, inhabitants, tenants and users (shops, enterprises, organisations, etc.), public authorities and institutions which manage, monitor, renovate and save heritage are equally responsible for the appropriate usage of built heritage. Today, many of the heritage buildings located in the downtown areas of Transylvania medieval cities have a double functionality: upfront, most ground-floors at the street-side have become commercial areas (hosting, especially, shops, restaurants, bars and cafes, public institutions, including cultural ones, respectively offices of legal authorities, etc.), while back-yard buildings and upper stories have kept their residential purposes. Some of the main reasons why Romania built heritage suffers are: the nationalization of private properties and businesses (1947–1963), the transformation of individual homes into collective residential buildings and the installation of tenants, the avoidance of taking the responsibility to refurbish the buildings, the lack of financial resources, the lack of interest and knowledge of the inhabitants, the difficult and late process of returning the properties to the entitled owners, the orientation of shopkeepers towards short-term profits, the poor implementation of the existing legal framework by local and national public authorities, etc.

With over 100 buildings that are still inhabited, Sighisoara was listed by UNESCO as an inhabited medieval citadel-town, in 1999, being described<sup>18</sup> as: 'Founded by German craftsmen and merchants known as the Saxons of Transylvania, Sighisoara is a fine example of a small, fortified medieval town which played an important strategic and commercial role on the fringes of central Europe for several centuries.' Two reasons determined the recognition of the site: 'Criterion (iii): Sighisoara is an outstanding testimony to the culture of the Transylvanian Saxons, a culture that is coming to a close after 850 years and will continue to exist only through its architectural and urban monuments. [and] Criterion (v): Sighisoara is an outstanding example of a small fortified city in the border region between the Latin-oriented culture of central Europe and the Byzantine-Orthodox culture of South-Eastern Europe. The apparently unstoppable process of emigration by the Saxons, the social stratum which had formed and upheld the cultural traditions of the region, threatens the survival of their architectural heritage as well'18. The most valuable cultural resources of Sighisoara consist especially of architectural monuments (71 of local interest and 144 of national importance, completed by 14 archaeologic sites, of which 13 are of national importance, and 4 other sites)<sup>19,20</sup>. In fact, these constitute the most significant part of the endangered heritage that must be protected.

The National Institute of Statistics<sup>17</sup> indicates the functioning of 5 museums between 2005 and 2013, with 18 or 19 employees between 2011 and 2013; since 2014, the number of museums has decreased by one, while the number of employees has also declined from 20 in 2014 to 17 in 2015. However, only two museums could be identified in the national database<sup>21</sup>: The County Museum of History<sup>22</sup> (with three sections: The Clock Tower, The Weapons Room, and the Torture Chamber) and N. D. Cocea Memorial House. Commonly, a museum that has more sections, usually, grants its visitors access based on a single ticket, thus, official statistics are going to be treated as referring to only two museums (Fig. 1). Furthermore, as the History Museum is located in the Citadel, being a part of it and due to its offer and particularities<sup>23</sup>, it may be considered to be a lot more attractive to visitors, compared to the N. D. Cocea Memorial House. The main section of the History Museum is located in the Clock Tower, being disposed vertically<sup>23</sup>, visitors going up from one room to another, until they reach the balcony (on the 6th floor), which also offers them a great view over the Citadel and the Lower Town. Around two thirds of the visitors arrive in organised groups<sup>24</sup>.

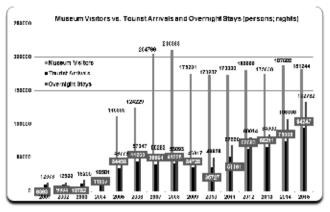
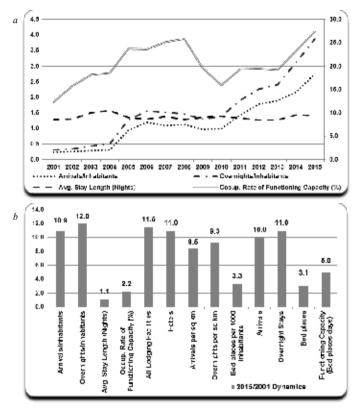


Fig. 1. Museum visitors and tourism activity (arrivals and overnights) in Sighisoara Source: authors elaboration based on statistics<sup>18</sup>

An important part of Sighisoara cultural heritage is provided by the events it hosts<sup>24</sup>. Some of them have a significant contribution to the Citadel tourist activity, being located in-between the medieval walls. Most of the events take place during the summer season: June (German Cultural Days, since 2006; Sighisoara Film Fest, presenting short and very short movies); July (Medieval Sighisoara Festival, organised since 1992, offers: medieval dance, music and theatre shows; Sighisoara is Romanian medieval cultural capital; Proetnica Intercultural Festival, organised between 2001 and 2009; part of The Medieval Sighisoara Festival since 2012; Craft Fair for craft-persons from Transvlvania and Bukovina); August (Academic Music Festival, since 1994, offers: classic concerts of Romanian and international artists and summer music and visual art courses for Romanian and international students); September (Military Band Festival, organised since 2004; Transylvania Hop Fest; Saxons Meeting). Fewer events and of lower tourist and economic impact take place during the late winter and spring season: February-March (Sighisoara Blues Festival, since 2006, promotes blues music); spring (Gaudeamus Cultural-Educational Fest, focuses on chamber music); early summer (National Modern Music Fest for Children and Youth 'The Voice of the Citadel', in May, since 2014; Sighisoara Day, on the 22nd of May, since 2008). The National Folk and Tradition Festival is held at the end of November. Other events of limited interest are organized according to the Romanian and European official calenders<sup>24</sup>.

According to the city hall officials<sup>24</sup>, The Medieval Sighisoara Festival gathers together around 50 000 tourists, reason why one can not understand the authorities decision to suspend the festival in 2016! Over time, due to its more or less authentic atmosphere, Sighisoara has gained an identity of museum-town but, at the same time, in the evening the Citadel is perceived as an abandoned place, because the inhabitants – aiming to protect themselves against the noise of the events, and/or tourists/visitors – have quit living in the rooms up-front and, consequently, do not

turn the lights on anymore. The same perception is also generated by the poor state of many facades of private and public buildings in the Citadel. Good practices<sup>24</sup> have been rewarded: Evangelic Church on the Hill received the Europa Nostra Award (UNESCO) for appropriate renovation; similarly, the team that renovated the House with Deer received the Grigore Ionescu Budesti award.

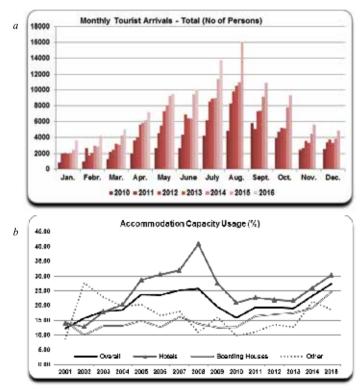


**Fig. 2**. Tourist activity in Sighisoara (*a*) and its growth rates (*b*) Source: authors elaboration based on statistics<sup>18</sup>

Data gathered in Fig. 1 reveal a rather peculiar fact, namely that Sighisoara registers more museum visitors than tourist arrivals and overnight stays. By correlating this situation with the very low length of stay (Figs 2*a* and *b*), one may conclude that Sighisoara is rather a day-visit destination (during circuits) and not a destination itself, although until 2015, compared to 2001, arrivals had increased by 10 times and overnights by 11 times, registering much higher growth rates compared to other urban destinations (Alba Iulia, Brasov, Bucharest, Cluj-Napoca, Sibiu, Timisoara or Tirgu Mures) (Alba Iulia and Tirgu Mures are two Transylvanian smaller cities, that register similar tourism activities; Sibiu is a somewhat larger city and the former European Capital of Culture from 2007 (ECoC 2007),

thus, a relevant urban cultural destination; Cluj-Napoca and Timisoara are two of Romanian largest cities, both important business destinations and also candidates for the ECoC 2021 title; Bucharest is Romanian capital, the country largest city, a major business destination and also a candidate for ECoC 2021).

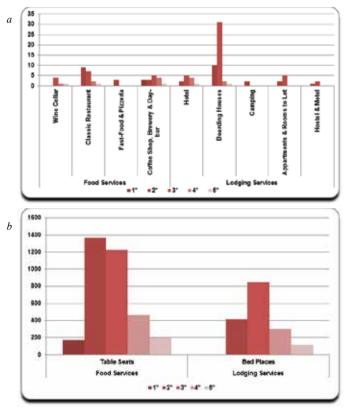
Sighisoara hospitality supply has evolved along with the development of tourist demand. Thus, the destination accommodation capacity has registered a continuously increasing trend beginning since 1990, accelerating after 1999, in terms of numbers of units and of bed places. The supply side is dominated by hotels and boarding houses but tourists clearly prefer hotels. Unfortunately, the average lengths of stay are very short, of around 1.4 nights in 2015, indicating, once again, that Sighisoara is rather a day-visit or a one night stop-over destination mainly for cultural tourism (Figs 3*a* and *b*). Due to its geographic position, in the Southern art of Transylvania and in the centre of Romania, on a major European road (E60, Route), transit tourism is also to be taken into consideration.



**Fig. 3**. Tourist demand: monthly arrivals (*a*) and functioning capacity usage (*b*) Source: authors elaboration based on statistics<sup>18</sup>

Sighisoara tourism is clearly affected by seasonality, tourists preferring to visit it between May and October but with a peak season in July and August. This situ-

ation also occurs due to the planning of cultural events. The low occupancy rates and lengths of stay explain the low contribution of tourism to the local economy<sup>24</sup> (of approximately 6% in the city turnover).



**Fig. 4**. Hospitality supply: types of units (*a*) and capacity by classification (*b*) Source: authors elaboration based on official classification data<sup>25</sup>

Neither the types, nor the qualitative levels of the hospitality services providers are sufficient for the improvement of Sighisoara tourist activity (Figs 4*a* and *b*). According to the City Hall officials<sup>24</sup>, the international foreign markets targeted include: Spain, Japan, Germany, Italy, USA, UK, France and Hungary. Unfortunately there are no detailed statistics concerning international tourism within this destination but, national figures have revealed that foreign visitors seek upscale lodgings and specialised food serving units. Understanding the need to protect the Citadel cultural heritage, local authorities<sup>24</sup> have accessed European funds and have implemented a few projects in this sense (HerO – Heritage as opportunity; FOS Lighting Designers for Historic Urban Landscape; HeritProt – heritage protection against fire; a National Tourist Office was also created) but the major priority that

needs to be addressed is the renovation of the fortifications and of most houses in the Citadel.

# CONCLUSIONS

Given the very high growth rates of the travel and tourism industry in this city, it becomes obvious that authorities must consider developing and adopting sound destination management strategies to ensure the development of sustainable tourism. Currently, the destination is not in danger of being overcrowded but, the increasing tourist flows may eventually generate problems related to its carrying capacity<sup>26</sup>. Moreover, it is a known fact that the development of cultural tourism is closely linked to the modernisation of infrastructure<sup>27</sup>. The next lines include a set of recommendations<sup>10,28</sup> regarding the adoption the most appropriate destination management strategies. The supply-side needs an appropriate management in the sense of further developing according to the needs and expectations of the desired and targeted tourists. Thus, both entrepreneurs and public officials must become aware of the need to come up with an offer of authentic, attractive, diversified, coherent and upscale quality hospitality services. Kitschy designs and products must be banned, and residents must be determined and supported to correctly refurbish their properties. Further, suppliers must develop complimentary services. including the organisation of other types of events (e.g. Christmas markets, Easter fairs and Crop celebrations, educational crafts, traditional cuisine and music and visual art workshops and events for families with children and for school pupils) with the purpose of increasing tourist demand during off-peak seasons.

Establishing networks and developing the cooperation with the surrounding Saxon villages, respectively with other Transylvanian medieval towns, can also contribute to the growth of the destination tourist activity. Even more, international city-to-city<sup>29</sup> networking and cooperation can be considered with other European medieval cities; a successful example in this respect is provided by the Black Sea Tourism Net<sup>30</sup>.

Suppliers must grant a special attention to organised-group tourism and to the cooperation with local travel agencies, which ought to specialise on incoming activities, rather than on outgoing ones, as they currently do.

Moreover, both suppliers and administration must extend their offerings considering the natural heritage of Sighisoara, preserved and promoted by Natura 2000 (Ref. 31), heritage which provides the opportunity to develop nature-based tourism in the area.

Further, given the increasing interest of tourists for the destination but also the problems identified from the perspective of the Citadel residents and their privacy, both public and private actors must work on identifying and establishing areas based on unlimited, limited or restricted public access. In fact, the best technical

solution that can be adopted in this respect is to create a Destination Management Organisation for Sighisoara.

Last but not least, all of the destination stakeholders must consider the development of a tourist brand for Sighisoara and, consequently, the must unite their efforts in this respect. Basically, destination managers must balance several aspects<sup>32</sup>. Thus, marketing efforts should not target demand maximisation but they ought to ensure the local population a high standard of living, simultaneously with highquality services for the destination tourists, both in a good environment. Further, as the preserved natural and cultural heritage represents a key attraction driver for tourists, their promotion must also raise awareness concerning the protection and conservation of the destination assets and values.

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# OPERATIONAL INFLUENCE ON THE ENERGETIC EFFICIENCY OF A GAS COGENERATED OPERATED ELECTRICITY GENERATOR

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**Abstract**. Cogeneration represents a solution for the use of secondary energetic resources considering the thermal-energetic installations. The 'on site' experimental determinations have established the values for the operational parameters for different tasks carried out by the installations. The specific energetic indexes have also been determined, based on the analysis of the energetic balance. The results were materialised through the appointment of the dependence of the energetic efficiency and the cogeneration index on the electric load degree of the generator.

Keywords: diesel engine, cogeneration, energetic efficiency, environmental impact, fossil fuels.

## AIMS AND BACKGROUND

One of the present solutions used to capitalise the secondary heating resources is represented by the cogeneration consisting in the simultaneous supply of electricity and heat. The main advantage of cogeneration is the capitalisation of heat evacuated from industrial installations This installation uses stratified polycarbonate plates that increase noise isolation of the sources up to 30 dB (Ref. 1).

The paper brings forward the analysis of the energetic performances of a cogeneration plant composed of 3 gas operated electricity generation groups, with the electric power of one MW, analysis carried out by experimental determination. The type of installation which was analysed and characterised by the following advantages: it covers a wide range of electric powers, it simultaneously supplies heat and electricity to areas which are out of reach, it reduces the thermal environmental impact making use of the heat emitted by internal combustion engines, and it is built using different modules.

Studies for the assessment of the constructive and functional performances of cogeneration installations powered by internal combustion engines were also carried out by different producers, research institutions and universities. The energetic performances, the analytic relations required for the consideration of the opportunity to adopt a cogeneration method as well as the investment values

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for different maintenance tasks are mentioned in different speciality papers<sup>2</sup>. The performances of different cogeneration systems have been comparatively analysed, insisting upon the solution which uses internal combustion engines, fuelled by gas or other types of biogas.

The energetic and exegetic assessment of the performances of a series of cogeneration plants fitted with steam turbines and respectively with internal combustion engines have been carried out<sup>3</sup>. The obtained results are highlighted through energetic approach (maximum heat performance) and through an exegetic approach (maximum electric performance). The analytic models used to simulate the operation of the cogeneration systems with the definition of the quantity and quality indicators, assessing as well the contribution of the electric and respectively that of the thermal component of the installations, are brought forward<sup>4</sup>. The paper follows the evolution of the energetic performances over a year identifying the thermal-dynamic imperfections determined by the variation of the environmental temperature and the uneven load determined by the succession of the seasons and the exploitation of the industrial energetic system.

#### EXPERIMENTAL

The study deals with a cogeneration plant composed of 3, 1 MW electricity generators, supplied with natural gas foreseen with a Heat recovery steam generator (HRSG). The determined parameters are the following: fuel flow, electric power, steam heating power, hot water heating power, losses with burnt gases, losses with reduced heat water, losses in the electric generator, radiation losses, total losses (engine + generator). The calculations were carried out on a monthly respectively yearly basin in order for the data to relevant<sup>5</sup>. The measuring devices used are actually temperature sensors fitted with data loggers, thermo-hygrometers, ultrasonic liquid flowmeters, Pitot-Prandtl tube for gas pressure and flow, infrared thermal vision camera, Testo 350 gas analyser.

For a real base line the catalogue data of the cogeneration electricity generator group was used (Table 1).

The determination of the significant parameters was carried out based on the analytic relations characteristic to the 1st and the 2nd principle of thermodynamics. The quality and quantity energetic performance indicators (efficiency, cogeneration index, specific fuel consumption, losses percentage, the degree of capitalisation of outlet heat) are graphically presented in the paper.

No	Performance	Measuring	Value
		unit	
1	alternator electric power	kW <sub>e</sub>	1033
2	water thermal power at high and medium temperature	kW	569
3	thermal power of burnt gases up to 110°C	kW	776
4	gas power inserted at low temperature	kW	2752
5	electric efficiency	%	37.5
6	thermal efficiency	%	48.9
7	cogeneration efficiency	%	86.4
8	flow of high temperature water	m³/h	65
9	inlet and outlet temperature of high temperature water	°C	101/105
10	flow of medium temperature water	m³/h	35
11	inlet and outlet temperature of medium temperature water	°C	83/98
12	flow of low temperature water	m³/h	30
13	inlet and outlet temperature of low temperature water	°C	32/35
14	power at the shaft	kW	1070
15	thermal power of high temperature water	$kW_t$	316
16	thermal power of medium temperature water	kW	253
17	thermal power of low temperature water	kW	104
18	thermal power of burnt gases up to 25°C	kW	933
19	power radiated by the thermal engine	kW	76
20	air mass flow	kg/h	5978
21	burnt gases mass flow	kg/h	6187
22	temperature of burnt gases	°C	507
23	gas flow	m³ <sub>N</sub> /h	275
24	inferior combustion value of gas	kJ/m <sup>3</sup> <sub>N</sub>	36000

Table 1. Technical specifications of the analysed electricity generator group

The basic diagram of an electricity cogeneration group powered by an internal combustion engine is presented in Fig. 1.

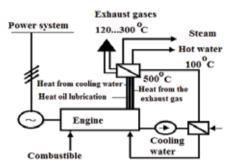


Fig. 1. Basic diagram of an electricity cogeneration group powered by an internal combustion engine

The characteristic indicators of the energetic balance for an internal combustion engine are presented for information in Table 2.

Indicators	Otto	Diesel
Fuel consumption	100	100
Electricity generation αe	35	40
Heat generation αt	50	43
Out of which: burnt gases	20	21
Cooling water	30	22
Total efficiency	85	83
Heating index $-y$ – (of cogeneration),		
$(J_{e}/J_{t}), (kW_{e}/kW_{t}),$	0.6	6-0.93
(kWh/Gcal)	700	-1082

 Table 2. Characteristic indicators of the energetic balance for a system powered by an internal combustion engine

### CASE STUDY

The operating loads of the heating plant, imposed by the technological requirements were explored using hourly experimental determinations, in parallel with the existing monitoring system in order to verify the validity of the indexes of the monitoring system<sup>6</sup>. The monthly and yearly values were taken from the monitoring system. The performance of the internal combustion engines, plate heat exchangers, HRSG and of the cooling installation of the inlet water into the electricity generator group were individually observed.

The lack of heat demand for the technological processes and for heating resulted lead to the shutdown of the plant during summer<sup>7</sup>. Another justification for having shutdown the plant during the warm season was the inefficiency of the water cooling process, which determines the impossibility to load the engine at its nominal capacity.

The determined values of the components of the monthly and yearly energetic balance (electricity, heat, effective energy) are presented in Table 3, while Table 4 presents the energy losses.

Month	Inlet	(%)	Shaft	(%)	Gen-	(%)	Thermal	Effec-	Inlet
	energy		energy		erator		energy	tive	energy
	(MW h)		(MW h)		energy		(MW h)	energy	(MW h)
					(MW h)		(	MW h	)
January	4474.2	100.0	1489.9	33.3	1350.1	30.2	1707.0	38.2	3057.1
February	5116.4	100.0	1728.8	33.8	1569.3	30.7	1930.0	37.7	3499.3
March	5772.2	100.0	1756.8	30.4	1579.5	27.4	2395.0	41.5	3974.5
April	2539.7	100.0	890.4	35.1	797.4	31.4	799.0	31.5	1596.4
May	1516.8	100.0	545.7	36.0	477.6	31.5	348.0	22.9	825.6
September	1094.3	100.0	412.4	37.7	349.6	31.9	108.0	9.9	457.6
October	2246.6	100.0	802.0	35.7	727.5	32.4	763.0	34.0	1490.5
November	2786.7	100.0	960.3	34.5	874.1	31.4	1039.0	37.3	1913.1
December	3661.8	100.0	1271.7	34.7	1163.7	31.8	1402.0	38.3	2565.7
Yearly	29208.4	100.0	9852.0	33.7	8888.7	30.4	10548.0	36.1	19436.7

Table 3. Inlet electricity, heat and effective energy for the monthly and yearly energetic balance

Table 4. Energetic losses

	Engine losses (MW h)	(%)	Genera- tor losses (MW h)	· · ·	losses (MW	(%)	Water losses (MW h)	(%)	tor ra- diation.	(%)	ra- diation.	(%)	CHP index
					h)				losses (MW h)		losses (MW h)		
Jan	2984.3	66.7	139.7	3.1	595.5	13.3	394.2	8.8	115.6	2.6	172.1	3.8	0.8
Feb	3387.7	66.2	159.5	3.1	679.5	13.3	449.8	8.8	132.1	2.6	196.3	3.8	0.8
Mar	4015.4	69.6	177.3	3.1	755.3	13.1	500.0	8.7	146.8	2.5	218.2	3.8	0.7
Apr	1649.2	64.9	93.0	3.7	396.3	15.6	262.4	10.3	77.1	3.0	114.5	4.5	1.0
May	971.0	64.0	68.2	4.5	290.4	19.1	192.3	12.7	56.5	3.7	83.9	5.5	1.4
Sep	682.0	62.3	62.8	5.7	267.5	24.4	177.1	16.2	52.0	4.8	77.3	7.1	3.2
Oct	1444.6	64.3	74.6	3.3	317.7	14.1	210.3	9.4	61.8	2.7	91.8	4.1	1.0
Nov	1826.4	65.5	86.1	3.1	367.0	13.2	243.0	8.7	71.3	2.6	106.1	3.8	0.8
Dec	2390.1	65.3	108.1	3.0	460.6	12.6	304.9	8.3	89.5	2.4	133.1	3.6	0.8
Year	19356.4	66.3	963.3	3.3	4105.8	14.1	2717.9	9.3	798.3	2.7	1186.5	4.1	0.8

#### **RESULTS AND DISCUSSION**

Graphic representations were used in order to facilitate the interpretation of results and to carry out the correlations between the energetic performances. The results obtained and the variations of the significant parameters were graphically presented in the figures hereinafter.

After having analysed Figs 2–6 the following results emerged:

- the operation of the group for reduced loads increases the monthly energetic losses with 6.4% per month, equivalent to 207.7 MWh/month;

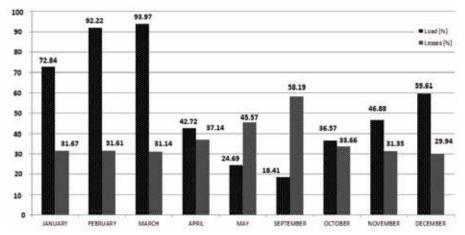


Fig. 2. Variation of total losses related to load of the CHP (combined heat and power) system

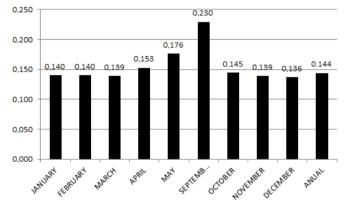


Fig. 3. Specific fuel consumption (m<sup>3</sup>/kWh)

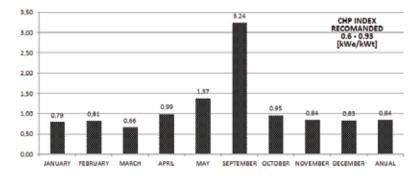


Fig. 4. CHP index

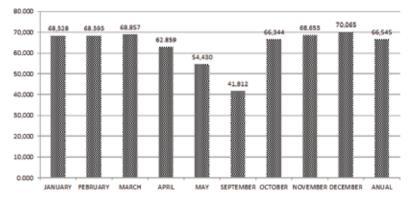


Fig. 5. Global energy efficiency

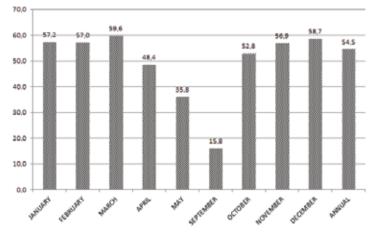


Fig. 6. Efficiency of capitalisation of secondary energy resource (SER)

- corresponding to reduced loads, the specific fuel consumption increases with  $0.0155 \text{ m}^3/\text{kWh}$ ;

- the cogeneration indexes superior to the recommended values, highlight the fact that the group operates as a electricity generation group, the thermal component being therefore insignificant;

- reducing therefore the usage degree of the thermal potential available affects the capacity of the cooling installation, determining the reduction of the load of the thermal engine.

Considering the age of the installation, the operational parameters are situated within the range of values corresponding to similar installations in use.

Two measures are therefore brought forward in order to increase the energetic efficiency:

– ensuring the maintenance which should correspond to the operational period of the installation;

- the complete capitalisation of the thermal potential of the installation by application of the three generation.

The economies realised by the end of one calendar year, considering an optimum load of the three generation group for a period of 10 months/year is estimated to be the following:

 $\Delta E = 207.7 \times 10 = 2077 \text{ MWh/a} = 178.622 \text{ tep/a}.$ 

### CONCLUSIONS

The experimental determinations complemented with analytical determinations highlight the fact that the energetic efficiency, correspondingly the BAT (best available technology), is met by 80% from the nominal load and the operation of the HRSG for a thermal operational efficiency of 90%. As a general conclusion, the use of cogeneration becomes economically and energetically efficient if a 35–50% of the thermal energy demand is ensured.

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# REMOVAL OF Fe, Zn AND Mn IONS FROM ACIDIC MINE DRAINAGE USING HYDROXYAPATITE

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**Abstract**. The present study aimed at evaluating at laboratory-scale the possibility of ability of a synthetic hydroxyapatite in the removal process of metallic ions (Fe, Zn and Mn) from acid mine drainage generated at the abandoned mining perimeter of 'Gura Minei' from Rosia Montana (Alba County, Romania). The water drainage from 'Gura Minei' is acidic (pH 2.9) and contains high concentrations of iron, manganese and zinc. The removal efficiency of metal ions from acid mine drainage by hydroxyapatite was determined at different sorbent dosages and contact times, during batch sorption experiments. The composition of the acid mine drainage from 'Gura Minei' before and after the treatment with hydroxyapatite was determined by X-ray fluorescence spectroscopy. In the investigated experimental conditions, adsorption of heavy metal ions from acid mine drainage by hydroxyapatite lay in the following order: Fe > Zn > Mn. About 99.8, 83.4 and 24.7% of iron, zinc and manganese, respectively, were removed from acid water drainage after 300 min of contact with 1.5 g synthetic hydroxyapatite.

Keywords: acid mine drainage (AMD), heavy metals, sorption, synthetic hydroxyapatite.

# AIMS AND BACKROUND

Acid mine drainage (AMD) is one of the most important environmental challenge facing the mining industry worldwide<sup>1,2</sup>. This type of pollution is commonly generated at the abandoned mines after the pumping was stopped and the water floods the underground sites. It occurs naturally within the environments containing an abundance of sulphide minerals, usually pyrite (FeS<sub>2</sub>) which oxidises and dissolves in contact with water and air.

<sup>\*</sup> For correspondence.

The main sources for AMD occurrence into the environment are waste rock and tailings<sup>3</sup>; underground and open-cast mines; stock and spoil piles and spent heap leach dumps. Generally, the acidic mine drainage is characterised by low pH value, high concentrations of metals (iron is the most common), elevate sulphate level and excessive suspended solids and/or siltation<sup>2</sup>. The release of acid mine water into the environment might cause toxicological effects on aquatic life, damage the ecosystem of receiving rivers and lakes, corrode the metal pipes and break down the concrete structures<sup>4</sup>. Furthermore, taking into consideration the predictions on the future loading of dissolved metals from abandoned mines which advise that sulphide oxidation and release of the dissolved metals into the environment could carry on for decades to centuries, it is evident that appropriate AMD treatments for heavy metals removal are necessary<sup>4</sup>. Usually, the acid mine drainage waters are treated by adding alkaline materials (i.e. CaCO<sub>3</sub>, Ca(OH)<sub>2</sub>, CaO, Na<sub>2</sub>CO<sub>3</sub>, NaOH etc.) to the source of AMD or directly to the polluted stream, in order to neutralise the water and to precipitate the metals as hydroxides. The main disadvantage of this method is the need for continuous operation and maintenance, low reaction rates, and the production of huge amounts of secondary wastes<sup>5</sup>.

Several other methods have been also used for acid mine drainage treatment, including adsorption, biosorption, ion flotation and its variants, ion-exchange, membrane separation, reverse osmosis, electrochemical remediation, solvent extraction and/or adsorption-flotation, precipitation-flotation<sup>6-11</sup>. Nowadays, adsorption is widely accepted in the environmental treatment applications and extensive research has been carried out in the last several years to find materials having high absorption capacity, low water solubility, high stability under reducing and oxidising conditions, low costs and availability in order to be used as sorbents for the removal of metal ions from various wastewaters, including AMD (Ref. 12). In recent years, it was reported that apatite-group minerals with special crystal chemistry characteristics would become the most promising mineral materials for the treatment of wastewater containing fluoride and heavy metals<sup>13</sup>. Among them, hydroxyapatite  $[Ca_{10}(PO_4)_{4}(OH)_{2}]$  which is the main mineral constituent of human tissue (teeth, and bones) appears to be an ideal material for the disposal of long term contaminants from polluted soils or sewage and wastewaters. Several investigations have proved the ability of both, natural and synthetic hydroxyapatites in the removal process of various heavy metals, i.e. Pb, Zn, Cu, Cd, Co, As, Ni, Fe, Mn from solutions<sup>12</sup>. The sorption mechanisms of heavy metals on hydroxyapatite are diverse and mainly include adsorption, ion-exchange, dissolution/precipitation, surface complexation, diffusion into the solid, coprecipitation or precipitation of amorphous phases<sup>14</sup>. However, different sorption mechanism often work together and it is difficult to quantify the relative contribution from each process responsible for the metal uptake. Despite various studies<sup>15</sup> attesting the effectiveness of several types of hydroxyapatite for heavy metal ions from synthetic aqueous solution under

different experimental conditions, no research have been carried out to establish the sorbent ability to eliminate the metals from authentic wastewaters.

The present study aimed at evaluating at laboratory-scale the possibility of using synthetic hydroxyapatite (sHA) (Ref. 16) the removal process of the metallic ions (Fe, Zn and Mn) from the acidic mine drainage generated at the abandoned mining perimeter of 'Gura Minei' from Rosia Montana (Alba County, Romania), under batch sorption experiments. The concentration of metals in the water drainage from 'Gura Minei' before and after the treatment with synthetic hydroxyapatite was determined by X-ray fluorescence spectroscopy.

## EXPERIMENTAL

*Sampling site*. The exploitation 'Gura Minei' gallery is located in Rosia Montana mining perimeter, in the South Apuseni Mountains (Romania). Although the mining operation in Rosia Montana area were closed, currently the mine waters from 'Gura Minei' are flowing unimpeded in the 'Valea Rosiei' creek from nearby and then in Abrud river. The mine water drainage channel is arranged inadequately (Fig. 1) as it is a simple trench dug in the topsoil blankets from the abandoned platform.

Three samples of water drainage were collected from the abandoned mining perimeter of 'Gura Minei' (Fig. 1) and their chemical composition was determined by X-ray fluorescence spectrometry using a Quant'X ARL spectrometer (Thermo Scientific, USA). The water pH was measured on-site using a portable pH meter (Hanna instruments).



Fig. 1. Water drainage at 'Gura Minei' abandoned mining perimeter (23.05.2014, A. Lancranjan)

*Characterisation of sorbent material.* Hydroxyapatite (sHA) was obtained by wet chemical methods (precipitation), from CaCl<sub>2</sub> (Sigma-Aldrich) and Na<sub>2</sub>HPO<sub>4</sub>

(Sigma-Aldrich) as raw materials. Precipitation reactions take place at the room temperature<sup>16</sup>:

 $10 \text{ CaCl}_2 + 6 \text{ Na}_2\text{HPO}_4 + \text{H}_2\text{O} \rightarrow \text{Ca}_{10}(\text{PO}_4)_4(\text{OH})_2 + 12 \text{ NaCl} + 8 \text{ HCl}$ 

The resulted precipitate was maintained in the suspension at the ambient temperature, in the presence of crystallisation admixtures, for 40 h, then filtered, washed with de-ionised water, dried at 110°C and thermally treated at 400°C for 2 h.

The granulation and specific surface area (BET) of the hydroxyapatite used in the present study were 10–70  $\mu$ m and 50 m<sup>2</sup>/g, respectively.

The efficiency of synthetic hydroxyapatite (sHA) in the removal of the metal ions from 'Gura Minei' water drainage was investigated at the laboratory scale, using a batch reactor (250 ml) with continuous stirring at 300 rpm.

Various quantities (0.5, 1 and 1.5 g) of sHA were left in contact with 100 ml AMD solution. Aliquots of supernatant (1.5 ml) were collected at different time intervals (from 15 to 300 min) and the concentration of the metal ions in the aqueous phase was immediately determined by X-ray fluorescence spectrometry. It should be noted that the total sampling volume did not exceed 10% of the initial solution volume. All experiments were conducted at room temperature  $(22 \pm 0.5^{\circ}C)$ , in duplicates to observe the reproducibility of the results and the mean values were used.

The removal efficiency, RE (%) of metallic ions by synthetic hydroxyapatite was calculated using the following equation:

RE (%) = 
$$\frac{c_{\rm i} - c_{\rm f}}{c_{\rm i}} \times 100$$
 (1)

5

1

0.5

where  $c_i$  and  $c_f$  are the concentrations of the metal ions (mg l<sup>-1</sup>) in the initial and final solutions, respectively.

#### RESULTS AND DISCUSSION

Fe (mg  $l^{-1}$ )

 $Mn (mg l^{-1})$ 

 $Zn (mg l^{-1})$ 

Acid mine drainage characterisation. The average chemical composition and pH of the water drainage samples collected from 'Gura Minei' abandoned mining perimeter is presented in Table 1.

abandoned mining perimeter	-	-
Parameter	AMD	Maximum consent limits*
pH	$2.9 \pm 0.1$	6.5-8.5

 $594 \pm 0.5$ 

 $21 \pm 0.2$ 

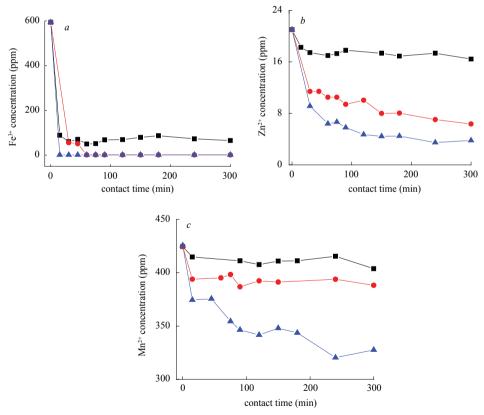
 $424.5\pm0.7$ 

Table 1.	Average	chemical	composition	and pl	H of	water	drainage	collected	from	'Gura	Minei'
abandon	ed mining	g perimete	r								

\*According to Romanian Standard NTPA 001/2002.

From Table 1 it could be observed that the water drainage is strongly acidic and contains significant levels of metal ions (Fe, Mn and Zn), whose concentrations substantially exceed the maximum consent limits established by Romanian Standard NTPA001/2002. It is clear that this water drainage introduces sulphuric acid and toxic metals into the environment that could damage the natural ecosystem, since AMD is disposed without any previous remediation treatment. On-site, the infiltration of the acidic water drainage into the soil which retains heavy metals and, therefore, causes constant pollution could be easily observed.

*AMD treatment using synthetic hydroxyapatite*. The removal of heavy metals from the acidic water drainage collected from 'Gura Minei' abandoned mining perimeter onto sHA samples was investigated at different contact times and sorbent dosages. The variations of iron, manganese and zinc ions concentration during the contact time between 100 ml acid water drainage and various amounts of sHA are illustrated in Fig. 2.

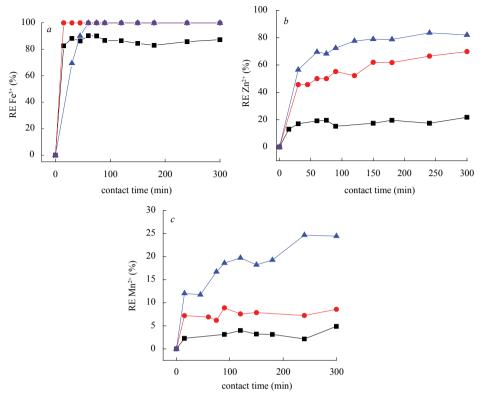


**Fig. 2**. Variation of iron (*a*), zinc (*b*) and manganese (*c*) ions concentration in AMD sample (100 ml) at different dosages of synthetic hydroxyapatite as a function of the contact time ( $\blacksquare$ ) 0.5 g; ( $\bullet$ ) 1 g; ( $\blacktriangle$ ) 1.5 g

It can be observed from Fig. 2 that at a dosage of 0.5 g, the investigated synthetic hydroxyapatite was not able to treat any of the metallic ions from AMD solution to below the legal requirements. Although, the concentration of iron in the AMD solution decreases from the initial values of 594 to 89 ppm during the first 15 min of contact with 0.5 g sHA, the manganese and zinc concentrations remained almost unchanged. At this sHA dosage, further increase of the contact time up to 300 min does not lead to an important enhancement of metal ions removal from AMD solution. However, the concentrations of the metal ions, especially iron and zinc in AMD solution significantly decrease by using higher amounts of adsorbent, when the removal rate is fast and the equilibrium is easily attained. This reveals that the instantaneous and equilibrium sorption capacities of the metal ions from AMD are functions of the sHA dosage. Since there is a higher total surface area at the higher adsorbent concentration, more adsorption sites are available causing higher removal of solution. For instance, the iron concentration in AMD sample decreases to 1 ppm after 60 min of contact with 1 g sHA and this value remains stable throughout the total contact time of 300 min. When an amount of 1.5 g sHA was used, the sorption equilibrium was attained after the first 15 min of contact and the value of the iron concentration in AMD solution was 1 ppm.

In the case of zinc and manganese, at a dosage of 1 g sHA, their concentrations remain at high levels, as could be observed from Figs 2*b*,*c*. An increase of sHA dosage to 1.5 g resulted in substantial improvement of zinc removal. Hence, the zinc concentration in AMD solution decreases from 21 to 9.1 ppm after the first 30 min of contact with sHA, while at the total contact time of 300 min, it reaches the value of 3.5 ppm. Disregarding the dosage of synthetic hydroxyapatite, the manganese concentration remains elevated in AMD solution and only a slight decrease from 424.5 ppm to 320 ppm took place after 300 min of contact with 1.5 g sHA.

From Fig. 2 it seems that the removal process of all metal ions from AMD solution took place in two steps. In the first step, the metal ions uptake is fast, while the in second one it happened slowly and exhibited a subsequent removal until equilibrium was reached. The main reason for the appearance of rapid step could be correlated with the high number of the active sites on the sHA surface at the first stages of adsorption process. The gradual occupancy of these sites might causes an emerging of the lower step<sup>17</sup>. The values of the removal efficiency of the synthetic hydroxyapatite calculated at different contact times, as a function of the adsorbent dosage are presented in Fig. 3.



**Fig. 3**. Effect of sHA dosage on the removal efficiency of the metal ions from AMD:  $(\blacksquare) 0.5 \text{ g}; (\bullet) 1 \text{ g}; (\blacktriangle) 1.5 \text{ g}$ 

As it can be seen in Fig. 3, in the investigated experimental conditions, sHA was able to remove significant amounts of metal ions, especially iron and zinc from the water drainage collected from 'Gura Minei' abandoned mining perimeter. At a sorbent dosage of 0.5 g/100 ml AMD, the iron removal efficiency was 88.1% after 30 min and reached the value of 99.2% by increasing the contact time to 60 min. At a sorbent dosage of 1.5 g, the percent of iron removal was 99.8 and the final concentrations of iron were reduced to levels less than the maximum consent limit for wastewater discharges after only 15 min of contact with sHA. In the case of zinc, the maximum value of the removal efficiency was 21.7% when an amount of 0.5 g sHA was used, but it enhanced to 69.7% by increasing the sorbent dosage to 1 g. The removal efficiency of zinc attains the maximum value of 83.5% after 240 min of contact with 1.5 g sHA. As seen in Fig. 3c, in the investigated experimental conditions, the removal efficiency of manganese is only 24.5% after 300 min of contact with 1.5 g sHA. This low removal rate of manganese from AMD solution may be the result of the weak interaction between Mn<sup>2+</sup> and sHA caused by the fact that the volume of hydrated manganese ions is high and the ions are more likely to be in solution rather than adsorbed. Another possible explanation could be related to the low value of water drainage pH, since it is known that the solution pH greatly affects metal sorption mechanism by apatite and metal–apatite reaction products. In the investigated experimental conditions, adsorption of heavy metal ions from acid mine drainage by synthetic hydroxyapatite lay in the following order: Fe > Zn > Mn. The high removal rate of iron and zinc ions from AMD solution is most probably attributed to the adsorption of metal ions on the solid surface followed by their diffusion into hydroxyapatite structure and the release of cations originally contained within the sorbent. This hypothesis was supported by the increases of the calcium concentration into the solution after the first moments of contact of AMD solution with sHA (results not shown).

# CONCLUSIONS

The present paper examined the effect of a synthetic hydroxyapatite in the removal process of metal ions from the acid water drainage collected at 'Gura Minei' abandoned mining perimeter in Rosia Montana (Romania). The preliminary results indicated that the used synthetic hydroxyapatite was able to remove significant amounts of metal ions, especially iron and zinc from acidic mine drainage. The removal effectiveness of the metal ions by the investigated sorbent is strongly dependent on its applied dosage and contact time with AMD solution. In the investigated experimental conditions, about 99.8, 83.4 and 24.7% of iron, zinc and manganese, respectively, were removed from AMD solution in contact with 1.5 g synthetic hydroxyapatite. The preliminary results showed that synthetic hydroxyapatite has the potential to be used for treating actual acid mine drainage. The rapid sorption rates of iron and zinc on hydroxyapatite allows the consideration of their removal by column filled setup which generally has a short contact time between the polluted solutions and the adsorbent. Further investigations are needed to investigate the mechanisms of adsorption in relation to the ion exchange process and to determine the optimum conditions for the use of synthetic hydroxyapatite for cleaning-up industrial wastewaters. The possibility of metals recovery and the regeneration of the sorbent used for AMD treatment using various eluting agents (i.e. hydrochloric acids, double distilled water, calcium chloride, barium hydroxide) is another issue to be further investigated.

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Clean technologies

# BATCH STUDIES FOR METHYLENE BLUE REMOVAL AND RECOVERY BY UNTREATED COFFEE RESIDUES

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**Abstract**. This paper reports the practicability of using coffee residues to remove widely used basic dyes like methylene blue from wastewaters. The effect of different batch system parameters, namely stirring speed, pH, initial dye concentration and contact time were studied. Moreover, in continuous fixed-bed column systems, the effects of parameters such as bed-depth, flow rate and initial dye concentration were examined. The experimental batch systems data were simulated using: (a) Freundlich, Langmuir and Sips isotherm models and (b) 1st order, 2nd order, and intra-particle kinetic models. The results revealed that the methylene blue is fairly adsorbed on coffee residues. This process could be a low cost technique for the removal of basic dyes from aqueous systems.

Keywords: adsorption, desorption, column, methylene blue, coffee residues, wastewaters.

### AIMS AND BACKGROUND

Many industries, such as paper, plastics, food, printing, leather, cosmetics and textile, use dyes in order to colour their products<sup>1</sup>. In textile industries about 10–15% of the dye gets lost in the effluent during the dyeing process, which are harmful products and may cause cancer epidemics<sup>2,3</sup>. Dyes usually have a synthetic origin and complex aromatic molecular structures which make them more stable and more difficult to biodegrade<sup>1–4</sup>. The industrial runoffs are usually discarded into rivers and lakes, altering the biological stability of surrounding ecosystems<sup>5</sup>. Therefore, removal of dyestuffs from wastewater has received considerable attention over the past decades.

In wastewater treatment, various methods were applied to remove dyes including photocatalytic degradation, membrane separation, chemical oxidations and electrochemical process<sup>6–9</sup>. Among the above mentioned techniques of dye removal, the process of adsorption gives the best results as it can be used to remove different types of colouring materials<sup>10</sup>.

Adsorption onto activated carbon is the most widespread technology for the removal of pollutants from water and wastewaters. The disadvantage of activated

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carbon is its high cost<sup>11</sup>. Hence, it is of pivotal importance thence of low-cost substitute absorbents to replace activated carbons. Various types of untreated biomass have been reported to have a use in dye removal: peanut husk<sup>4</sup>, palm kernel fibre<sup>11</sup>, *Turbinaria turbinate* alga<sup>12</sup>, graphene<sup>13</sup>, defatted jojoba<sup>14</sup> and sugar bet pulp<sup>15</sup>. The sorption efficiency of several adsorbents is presented in Table 1.

Further, numerous pretreated lignocellulosic materials are used to remove dyes in water and wastewater. Pyrolysed date pits<sup>16</sup>, date stones<sup>17</sup> and *Turbinaria turbinate* alga<sup>12</sup> have proved to be effective adsorbents.

Coffee residues and others similar byproducts like coffee husks and coffee grounds have been investigated to remove various dyes and heavy metals from aqueous solutions. Coffee husks have been used for the removal of Cu(II), Cr(VI), Cd(II) and Zn(II) (Ref. 18), coffee grounds for Nylosan Red N-2RBL (Ref. 19) and degreased coffee bean for Malachite Green<sup>20</sup>. Kyzas et al. reported the removal of Remazol Brilliant Blue RN and Basic Blue 3G using coffee residues, focusing on batch processes and not on continuous systems<sup>21</sup>.

In the current study, the adsorption capacities of coffee residues to remove methylene blue (MB) from aqueous solutions were investigated. Both\_batch and continuous fixed-bed-column systems were investigated. As regards the fixedbed-column systems filed with coffee residues, various bed-depths, flow rates and initial dye concentrations were examined for the first time.

#### EXPERIMENTAL

Methylene Blue (CI 52015) is a heterocyclic aromatic chemical compound with the molecular formula  $C_{16}H_{18}N_3SC1$ . It has many uses in a range of different fields, such as biology and chemistry. At room temperature it appears as a solid, odourless, dark green powder, that yields a blue solution when dissolved in water. The hydrated form has 3 molecules of water per molecule of methylene blue. Methylene blue should not be confused with methyl blue, another histology stain, new methylene blue, nor with the methyl violets often used as pH indicators. The dye used herein in batch and column experiments was Methylene Blue ( $C_{16}H_{18}ClN_3S.3H_2O$ , molecular weight = 373.90 × 10<sup>-3</sup> kg mol<sup>-1</sup>) supplied by Sigma-Aldrich. A stock solution was prepared by dissolving a specific amount of MB (humidity 22%) in distilled water. Working solutions were 3–140 mg l<sup>-1</sup>. MB concentrations were determined by measuring the absorbent values in each experiment with an HACH DR4000U UV-vis. spectrophotometer at  $\lambda = 664$  nm.

Coffee residues, a low-cost material acquired from different cafeterias in the city of Piraeus (Greece) were used as adsorbent. These residues were untreated (or modified to improve their adsorptive ability), but just only washed with distilled water to remove dirt and colour, and dried at 105°C for 5 h in a convection oven.

The residues used were in powder form (475–525 m) after sieving. This adsorbent was washed with distilled water and dried at 110°C for 24 h to remove the humidity.

Coffee residues, a low-cost material acquired from different cafeterias in the city of Piraeus (Greece) were used as adsorbent. This adsorbent was washed with distilled water and dried at 110°C for 24 h to remove the humidity. The dye used herein in batch and column experiments was methylene blue ( $C_{16}H_{18}ClN_3S.3H_2O$ , molecular weight = 373.90 10<sup>-3</sup> kg mol<sup>-1</sup>) supplied by Sigma-Aldrich. A stock solution was prepared by dissolving a specific amount of MB (humidity 22%) in distilled water. Working solutions were 3–140 mg l<sup>-1</sup>. MB concentrations were determined by measuring the absorbent values in each experiment with a HACH DR4000U UV-vis. spectrophotometer at  $\lambda = 664$  nm.

Isotherms were obtained from batch experiments. Accurately weighted quantities of approximately 0.5 g coffee residues were transferred to 0.8-1 bottles, where 0.5 l adsorbate solution were added. The temperature was 23°C, the MB initial concentration ranged from  $C_0 = 1.5$  to 150 mg l<sup>-1</sup>. The bottles are sealed and mechanically agitated for a period of 7 days. The 7-day period was determined after optimisation analysis, with agitation periods varying from 4 h to 14 days, to ensure that nearly equilibrium conditions were achieved. The final concentrations were determined. Concentrations (before and after equilibrium) from each bottle represented one point on the adsorption isotherm plots. The pH was near 7.5.

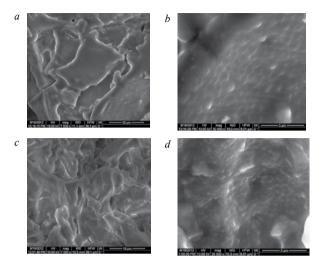
Batch experiments were carried out at stirring speeds from 65 to 664 rpm, pH varied from 1.54 to 12.9, the adsorption temperature varied from 23 to 70°C, initial dye concentration from 3 to 140 mg l<sup>-1</sup> for a maximum contact time up to 95 min. Samples were taken at 5 min intervals and the MB concentration was measured. The reactor, containing V = 11 aqueous solution of dye was placed in a water bath to maintain constant temperature at the desired level.

The study of the coffee residues by scanning electron microscopy (SEM) was conducted at the Institute of Materials Science, National Centre for Scientific Research 'Demokritos', using a FEI INSPECT SEM.

The concentration of output solution was measurement at  $\lambda = 664$  nm and using a HACH DR4000U UV-vis spectrophotometer. Finally, pH measurements were made using a digital pH meter, MultiLab model 540.

### **RESULTS AND DISCUSSION**

The SEM micrographs for coffee residues before and after MB adsorption are presented in Fig. 1. They indicate the swelling effect on the lignocellulosic particles after MB adsorption. The texture of the coffee residues particles after MB adsorption in Fig. 1*d* is rougher comparing to the same material before MB adsorption, presented in Fig. 1*b*. This fact indicates the swelling effect on the lignocellulosic particles after MB adsorption.



**Fig. 1**. SEM analysis for coffee residues (the coffee residues particle in Fig. 1*a* with magnification 7500× shows some pores. The surface texture of this particle is relatively rough as it can be observed in Fig. 1*b* with magnification 30000×. The adsorption conditions were  $C_0 = 140 \text{ mg } l^{-1}$ , t = 95 min,  $m/V = 1 \text{ g } l^{-1}$ , agitation speed = 664 rpm)

Three isotherm models including Freundlich<sup>22</sup>, Langmuir<sup>23</sup> and Sips<sup>24</sup> equations were used to fit the experimental data. The Freundlich isotherm model assumes that the surface of absorbent is heterogeneous and polymolecular layer adsorption takes place. This model can be described by the equation:

$$q = K_{\rm F} C_{\rm e}^{1/n} \tag{1}$$

where *q* is the amount adsorbed per unit mass of the adsorbent (mg g<sup>-1</sup>);  $C_e$  – the equilibrium concentration of MB adsorbed (mg l<sup>-1</sup>);  $K_F$ ((mg g<sup>-1</sup>) (l mg<sup>-1</sup>)<sup>1/n</sup>) – the Freundlich isotherm constant related to adsorption capacity, and *n* – the Freundlich isotherm constants related to adsorption intensity. In cases where the isotherm experimental data approximates the Freundlich equation, the parameters  $K_F$  and *n* can be estimated either by plotting lg *q* versus lg  $C_e$  either by non-linear regression analysis (NLRA). The linear form of the Freundlich isotherm model<sup>22</sup> can be defined by the following equation:

$$\lg q = \lg K_{\rm F} + (1/n) \lg C_{\rm e}.$$
 (2)

The Langmuir (1916) isotherm model<sup>23</sup> is given as:

$$q = \frac{K_{\rm L} q_{\rm m} C_{\rm e}}{1 + K_{\rm L} C_{\rm e}} \tag{3}$$

Moreover, this equation in linearised form is:

$$\frac{1}{q} = \frac{1}{q_{\rm m}} + \frac{1}{K_{\rm L} q_{\rm m}} \frac{1}{C_{\rm e}}$$
(4)

where  $K_{\rm L}$  is the Langmuir constant related to the energy of adsorption (l mg<sup>-1</sup>), and  $q_{\rm m}$  – the amount of MB adsorbed (mg g<sup>-1</sup>) when saturation is attained. In cases where the isotherm experimental data approximates the Langmuir equation, the parameters  $K_{\rm L}$  and  $q_{\rm m}$  can be estimated either by plotting 1/q versus 1/ $C_{\rm e}$  either by NLRA.

Numerous studies incorporate another important parameter,  $R_L$ , namely the separation factor. The value of  $R_L$  indicates the type of the isotherm to be either unfavourable ( $R_L > 1$ ), linear ( $R_L = 1$ ), favourable ( $0 < R_L < 1$ ) or irreversible ( $R_L = 0$ ) and is expressed by the following equation:

$$R_{\rm L} = \frac{1}{1 + K_{\rm L} C_0} \tag{5}$$

where  $C_0$  is the initial dye concentration (mg l<sup>-1</sup>), and  $K_L$  is the Langmuir constant (l mg<sup>-1</sup>).

Sips<sup>24</sup> isotherm model is a combination of the Langmuir<sup>23</sup> and Freundlich<sup>22</sup> isotherm type models and is expected to describe heterogeneous surface better. The Sips equation is presented by:

$$q = \frac{q_{\rm m} (K_{\rm L} C_{\rm e})^{1/n}}{1 + (K_{\rm L} C_{\rm e})^{1/n}}$$
(6)

where  $K_{\rm L}$  (l mg<sup>-1</sup>) is the Langmuir constants;  $q_{\rm m}$  (mg g<sup>-1</sup>) – the amount of MB adsorbed when saturation is attained, and n – the Freundlich constant<sup>22</sup>. These parameters can be estimated by NLRA.

	1	5	1		
Model	$K_{\rm F} (({\rm mg}~{\rm g}^{-1})$	п	$K_{\rm L} ({\rm l}{\rm mg}^{-1})$	$q_{\rm m}({ m mg~g^{-1}})$	SEE
	$(l mg^{-1})^{1/n})$				
Freundlich	3.555	1.612			2.397
Langmuir			0.0230	78.90	2.197
Sips		1.195	0.0111	109.27	2.168

Table 1. Isotherms parameters of Methylene blue adsorption on coffee residues

MB adsorption isotherms experimental data for untreated coffee residues are presented in Fig. 2. The theoretical curves are estimated for the above mentioned three models. The parameter values of these models are shown in Table 1. These parameters were obtained by NLRA. The standard error of estimate (SEE) was calculated in each case as follows:

SEE = 
$$\sqrt{\sum_{i=1}^{n'} (y_i - y_{i, \text{theor}})^2 / (n' - p')}$$
 (7)

where  $y_i$  is the experimental value of the depended variable,  $y_{i,\text{theor}}$  – the theoretical or estimated value of the depended variable; n' – the number of the experimental measurements, and p' – the number of parameters (the difference n - p' being the number of the degrees of freedom).

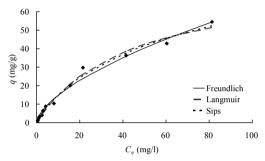


Fig. 2. Isotherm parameters of adsorption of coffee residues

The fitting of the Sips adsorption model to the present data was the most satisfactory for MB adsorption, better than the other two isotherm models, as shown by the corresponding SEE-values given in Table 1. The fitting of the Langmuir adsorption model to the present data was also satisfactory but to a lesser degree than the Sips model. The  $R_L$  values were found to be 0.236–0.935, i.e.  $0 < R_L < 1$ for all MB concentrations  $C_0$  in the range of 3–140 mg l<sup>-1</sup>.

In order to make adsorption mechanism more clear, the adsorption data were fitted using the pseudo-first order, the pseudo-second order and the intra-particle diffusion kinetic model.

The Lagergren<sup>25</sup> non-linear pseudo-first order equation is given as follows:

$$q - q_t = q \ e^{-kt} \tag{8}$$

where q and  $q_t (\text{mg g}^{-1})$  are the amounts of MB dye adsorbed per unit mass of the adsorbent at equilibrium time  $(t \to \infty)$  and adsorption time t (min), respectively, while  $k (\text{min}^{-1})$  – the pseudo-first order rate constant for the adsorption process. Moreover:

$$q = (C_0 - C_e) V/m \text{ and } q_t = (C_0 - C) V/m$$
 (9)

where C,  $C_0$ ,  $C_e$  (mg l<sup>-1</sup>) are the concentrations of MB in the bulk solution at time t, 0, and  $\infty$ , respectively, while m (g) – the weight of the adsorbent used, and V

(ml) – the solution volume. Further modification of equation (9) in logarithmic form gives:

$$\ln\left(q-q_{t}\right) = \ln q - kt. \tag{10}$$

The pseudo-second order<sup>26</sup> equilibrium adsorption model equation is given as:

$$q_t = q - (q^{-1} + k_2 t)^{-1} \text{ or } q_t = q - \frac{1}{1/q + k_2 t}$$
 (11)

where  $k_2$  (min<sup>-1</sup>) is the rate constant of second order adsorption.

The intra-particle diffusion model based on the theory proposed by Weber and Morris<sup>27</sup> is expressed as:

$$q_{\rm t} = c + k_{\rm p} t^{1/2} \tag{12}$$

where  $k_p (\text{mg} (\text{g min}^{0.5})^{-1})$  is the intra-particle diffusion rate constant and  $c (\text{mg g}^{-1}) - a$  constant related to the thickness of boundary. A value of *c* close to zero indicates that diffusion is the only controlling step of the adsorption process.

The stirring speed was studied between 65 and 664 rpm at constant dye concentration 14 mg l<sup>-1</sup>, pH equals 7 and stirring time of 95 min. The correlation between the first-order rate constant *k* for the MB absorption by coffee residues is shown in Fig. 3. The rate constant is generally increasing with stirring speed up to 200 rpm were a level-off is reached.

$C_0 \;({ m mg}\;{ m l}^{-{ m l}})$	140	75.3	26.3	11.3	7.8	3.0
First order						
k	0.0183	0.0227	0.0211	0.0113	0.0107	0.0176
$q_{\rm e}$	28.78	22.26	11.11	5.79	4.98	1.82
SEE	1.2827	0.9917	0.7150	0.1271	0.0515	0.0243
Second order						
$k_2$	0.000376	0.000604	0.001139	0.000796	0.000823	0.004587
$q_{\rm e}$	37.81	30.99	15.42	9.27	8.18	2.74
SEE	1.1818	0.8594	0.6563	0.1198	0.0488	0.0209
Intra-particle diff	usion					
с	-1.05	-0.70	-0.29	-0.49	-0.49	-0.16
k <sub>p</sub>	2.3371	2.1586	1.0365	0.4275	0.3647	0.1706
SEE	0.9196	0.6165	0.5137	0.1945	0.1613	0.0535

Table 2. Kinetic parameters of Methylene blue adsorption on coffee residues

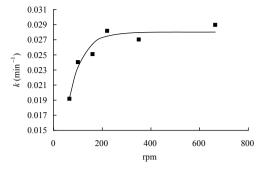


Fig. 3. Effect of stirring speed

According to the literature, the pH of the aqueous solution is one of the most important parameters of the adsorption process. The effect of the initial pH on the MB adsorption onto coffee residues is illustrated in Fig. 4. The range of pH studied was from 2 to 12. As can be seen the rate constant increases linearly by pH increasing.

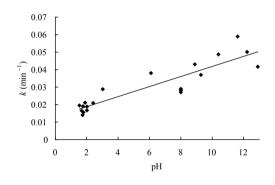


Fig. 4. Effect of pH

The effect of initial MB concentration on adsorption on coffee residues is shown in Fig. 5. The kinetic parameters of the above mentioned three kinetic models were estimated using NLRA and are presented in Table 2. As can be seen, the SEE values are lower for the intra-particle diffusion kinetic model indicating better fitting to the experimental data. In addition the pseudo-second order kinetic model had better fitting that the pseudo-first one.

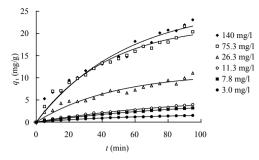


Fig. 5. Effect of initial MB concentration

The temperature variation from 20 to 70°C on MB uptake by coffee residues revealed that increase in temperature of the process enhanced better adsorption of MB from bulk solution. The conditions under which they became the experiments with different temperature were pH = 8, the initial concentration was 14 mg l<sup>-1</sup>, the stirring speed was 600 rpm and the dose of coffee residues was 1 g. Increase in temperature increased entropy of the system which yielded more chances of adsorption. The change in adsorption rate constant *k* as a function of adsorption temperatures is presented in Fig. 6.

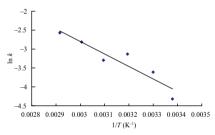


Fig. 6. Effect of temperature

The rate constant k of the Lagergren model follows the Arrhenius law:

$$k = p \exp\left(-E/RT\right) \tag{13}$$

The activation energy E (kJ mol<sup>-1</sup>) for the adsorption of MB on untreated coffee residues can be estimated by linear regression of lnk on 1/T. This activation energy was found to be 27.4 kJ mol<sup>-1</sup> or 6.57 kcal mol<sup>-1</sup>. The frequency factor of equation (13) was p = 1236 min<sup>-1</sup>. Consequently, the intra-particle diffusion, which is a physical process, is probably the controlling step of the adsorption process.

Our study is a research on the adsorption capacity of the coffee residue and its application to industrial scale. There is object for further research to determine the coffee residues quantity by region distribution and corresponding exploitation by industrial scale unit. The expressions used to calculate the parameter K and N

values after having performed NLRA are  $K = r/C_i$  and  $N = u \ln A/(xK) = C_i u \ln A/(xr)$ . The effluent dye solution volume V (in L) is V=Qt, where Q is the dye solution flow rate. According to the Bohart and Adams model<sup>28</sup>, the experimental data of the N and K values are presented in Table 3. The SEE-values are shown in the same table.

Table 3. Column	· · ·		2		<b>^</b>		CEE
	$C_{i}$	$Q_{\downarrow}$	п	N	K	$q_0$	SEE
	(mg/l)	(ml/min)		(mg/l)		(mg/g)	
x = 15  cm							
Bohart-Adams	165	20	2.000	42798	0.000080	96.90	2.3110
Clark	165	20	1.612	40522	0.000060	91.76	2.3053
Bohart-Adams	130	40	2.000	29317	0.001400	66.39	0.4876
Clark	130	40	1.612	26317	0.000100	59.59	0.6402
Bohart-Adams	150	70	2.000	31482	0.000250	71.29	5.8235
Clark	150	70	1.612	28382	0.000220	64.27	5.1515
Bohart-Adams	145	80	2.000	24745	0.000210	56.03	7.5229
Clark	145	80	1.612	20673	0.000180	46.80	6.3513
Bohart-Adams	800	20	2.000	46166	0.000020	104.54	38.2861
Clark	800	20	1.612	33869	0.000010	76.69	32.5900
Bohart-Adams	1600	10	2.000	30224	0.000010	68.44	102.8607
Clark	1600	10	1.612	22024	0.000010	49.87	93.8242
Bohart-Adams	1600	20	2.000	36641	0.000020	82.97	103.0998
Clark	1600	20	1.612	25017	0.000020	56.64	90.4492
x = 25  cm							
Bohart-Adams	150	70	2.000	38290	0.000160	86.65	4.0545
Clark	150	70	1.612	35501	0.000130	80.34	3.7795
Bohart-Adams	800	20	2.000	21788	2.25E-05	49.30	62.5900
Clark	800	20	1.612	16007	1.98E-05	36.20	56.0000
Bohart-Adams	550	40	2.000	32847	8.61E-05	74.30	17.6150
Clark	550	40	1.612	30028	7.51E-05	68.00	14.6410

Table 3. Column models parameters of methylene blue adsorption on coffee residues

The MB adsorption capacity for various lignocellulosic materials found in the literature was compared to the coffee residues adsorption capacity estimated in the present work (see Table 4). The adsorption capacity of the coffee residue was better than the adsorption capacity of other waste biomass such as peanut husk<sup>14</sup>, *Turbinaria turbinate alga*<sup>12</sup>, and wheat straw<sup>29–33</sup>. The adsorption capacities of various coffee residues/byproducts found in the literature are shown in Table 5. The coffee residues adsorption capacity found in the present work was close to the average of the Freundlich and Langmuir capacities, i.e.  $K_{\rm F}$  and  $q_{\rm m}$ , respectively, reported in the literature.

Adsorbents	Pretreatment	K <sub>F</sub>	п	$q_{\mathrm{m}}$	K	Refer-
		$((mg g^{-1}) (l mg^{-1})^{1/n})$		$(mg g^{-1})$		ence
1	2	3	4	5	6	7
Activated Car- bon fibres	chemical vapour deposi- tion modified		1.460	478.00	0.3750	38
shell	carbonised, burn activated	40.850	8.090	212.77	0.2690	39
Coffee residues	_	3.555	1.612	78.90	0.0230	in this work
Commercially activated carbon		_	-	370.40	1.0500	19
Cotton stalk	_	50.440	6.150	147.06	0.0249	5
Cotton stalk	sulphuric acid treated	202.210	1.810	555.56	0.6207	5
Cotton stalk	Phosphoric acid treated	157.910	16.150	222.22	14.516	5
Date pits acti- vated carbon	pyrolysed, FeCl <sub>3</sub> acti- vated	74.871	4.670	249.46	0.0830	16
Date stones activated carbon	pyrolysed, ZnCl <sub>2</sub> acti- vated	71.190	3.440	369.38	0.0429	17
<i>Enteromorpha</i> <i>prolifera</i> acti- vated carbon	ZnCl <sub>2</sub> activated	236.230	3.720	270.27	1.2300	13
Graphene	_	90.920	5.710	153.85	1.4400	13
Jojoba (defat- ted)	-	0.132	1.054	167.00	0.0009	14
Palm kernel fiber	_	8.670	1.770	95.40	0.0317	14
Peanut husk	_	16.650	3.270	72.13	0.0850	14
Pine sawdust	autohydro- lysed	15.680	2.582	88.02	0.1072	41
Pistachio hull	_	112.300	4.200	389.00	0.0960	39
Posidonia oceanica (L.) dead leaves activated carbon	carbonised, ZnCl <sub>2</sub> acti- vated	112.120		217.390		39
Sugar beet pulp	-	12.520	1.990	244.600	0.0149	15
Sugar beet pulp		2.777	10.128	714.290	0.0039	41

Table 4. Methylene blue adsorption capacity for various lignocellulosic materials

to be continued

Continuation of Table 4

e communication of 1						
1	2	3	4	5	6	7
Tea (rejected)	NaOH modi- fied	40.870	2.110	242.110	0.130	40
<i>Turbinaria tur-</i> <i>binate</i> alga	_	14.000	2.310	63.000	0.183	12
<i>Turbinaria tur- binate</i> alga	pyrolysed	40.000	3.030	163.000	0.104	12
<i>Turbinaria tur- binate</i> alga	pyrolysed, physically activated	105.000	3.630	411.000	0.083	12
<i>Turbinaria tur- binate</i> alga	pyrolysed, chemically activated	137.000	5.670	345.000	0.136	12
Wheat straw	acid hydro- lysed	10.950	1.010	20.410	0.663	33
Wheat straw	3-chloro-2-hy- droxypropyl trimethylam- monium chlo- ride modified	30.120	3.252	135.300	174.700	34

Table 5. Adsorption capacity of various coffee residues/byproducts

Adsorbents	Adsorbates	$K_{\rm F}(({ m mg} { m g}^{-1})) ({ m l} { m mg}^{-1}) { m l}/n)$	n	$q_{ m m} \ ({ m mg}{ m g}^{-1})$	$\frac{K_{\rm L}}{(\rm l~mg^{-1})}$	Refer- ence
Coffee grounds chemically acti- vated carbon	nylosan red N-2RBL	_	_	367	0.077	19
Coffee grounds chemically acti- vated carbon	methylene blue	_	_	181.80	0.109	19
Coffee husk-based activated carbon	remazol brilliant orange 3R dye	3.747	3.608	66.76	0.401	35
Coffee husks	methylene blue	2.567	1.290	90.09	0.0226	18
Coffee grounds	methylene blue	51.926	3.300	18.73	0.2687	36
Coffee bean (degreased)	malachite green	2.030	0.511	55.30	0.0935	20
Coffee residues	remazol brilliant blue RN	11.840	2.530	175.00	0.009	21
Coffee residues	basic blue 3G	36.610	3.120	240.00	0.041	21
Coffee husks	Cu(II)	3.702	5.010	7.496	0.4232	18
Coffee husks	Cd(II)	3.188	5.050	6.854	0.3024	18
Coffee husks	Zn(II)	2.721	6.070	5.565	0.2238	18
Coffee husks	Cr(VI)	2.353	3.650	6.961	0.1805	18
Coffee residues	methylene blue	3.555	1.612	78.900	0.0230	in this work

The present study is a part of continues involvement of our Research group in the study of wastewater treatment using original and modified (pretreated) lignocellulosic biomass as potential adsorbents. The Langmuir capacity is estimated as  $q_m =$  38.7 mg g<sup>-1</sup> for pine sawdust, significantly lower comparing to  $q_m =$  78.9 mg g<sup>-1</sup> for the coffee residues studied in the present work.

# CONCLUSIONS

The use of coffee residues is an innovative technique using waste biomass from urban and rural areas in and Industrial Ecology framework. Also, that is an index of the sensitivity of citizens for the rational management of waste and their contribution to sustainable development. The present work proves the viability of using coffee residues to remove basic dyes like MB from wastewaters. The effect of different system parameters on (a) batch and (b) continuous fixed-bed column systems were studied. The experimental systems data were simulated using the most commonly used isotherm and kinetic models. The continuous fixed-bed column results proved that the MB is practically adsorbed on coffee residues giving maximum Bohart and Adams capacity  $N = 46\ 166\ mg\ l^{-1}$  or  $q_0 = 104.5\ mg\ g^{-1}$  for bed-depth 15 cm, initial dye concentration 800 mg l<sup>-1</sup> and flow rate 20 ml min<sup>-1</sup>. These results provide evidence for suggesting this low cost residue for the removal of basic dyes in industrial scale applications.

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Environmental legislation

# INTERNATIONAL LIABILITY FOR ENVIRONMENTAL DAMAGE. CHALLENGES AND PROSPECTS

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Abstract. The environment protection has become a constant concern and interest of the international community with regard to environment issues, thus placing this concept within the major preoccupations of the international community, putting emphasis on the relationship between economic growth, population welfare and the degree of the planet pollution, showing that in present days the environment and its protection are the core imperatives of international community. This study starts from a concrete case of breaching environment protection rules, with major potential impact on international law and international relations. Social and economical aspects regarding the fraudulent approach towards pollution norms are also analysed, together with the impact of these emissions on environment and public health.

Keywords: climate change, fraud, socio-economical impact, international responsibility, due-diligence.

### AIMS AND BACKGROUND

Despite the existence of numerous political declarations and international multilateral conventions in the field of environmental protection, they apply mainly on preventing and reducing pollution and do not contain at present, besides some punctual elements, specific rules to determine how liability and accountability of States incur under public international law for issues that affect the environment and how to handle such unavoidable conflicts, as well as to address such concrete issues that may arise in the event of harm produced to the environment<sup>1</sup>.

We intend to analyse in this study the concept of international liability for harms produced to the environment<sup>2</sup> as well as the possibility of applying the viability and effects of the measures to be taken and can be taken in cases such as 'Dieselgate', which appears to affect, through a Domino effect, both the harmony of international relations (universally and at European level), as well as the global labor market, with adverse effects not only on the environment but on an entire economic system. Finally, we intend to analyse the economic and social impact of the infringement of rules of international law that can affect on short-term not only the environment but also employment and competitiveness, two EU important pillars.

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This study contains an analysis of key international conventions on the protection of the environment with implications for the liability under public and private international. For the purposes of this study, relatively recent international literature and relevant international jurisprudence were consulted. The conclusions in this study took into account the recent reports by the EU official institutions, but also of public international law entities, banks or media.

## CONCEPT OF INTERNATIONAL LIABILITY

Liability is the corollary of legal systems, the *sine qua non* condition for operation of social balance. Although at international level the concept of State liability is one of the oldest regulatory elements in jurisprudence solution of interstates disputes, at present there is no legal instrument agreed by the international community meant to legislate States accountability mechanisms, this gap being justified by the concept of State sovereignty.

Therefore, for a long time, liability for harms produced to the environment was made punctually, having an eminently national environmental character, the repairing process for damages incurred being made under the rules of tort or contract breach liability, the adverse effect being the reduced application of liability, made only towards individuals and specific entities thus leading, consequently, towards the lack of consistent environmental protection measures by *a priori* accountability of the main actors involved in environmental protection at international level.

Consequently, for environmental protection purposes, seen as a collective good of the planet, mechanisms that can trigger liability process even before the a damage occurred needed to be created.

However, regarding the responsibility of States, the international community has witnessed a jurisprudence conflict between two international judicial bodies, conflict confirming the need for a clear set of rules and tools in this area.

### ENVIRONMENTAL IMPACTS ON THE SOCIOECONOMIC SYSTEM

International community is currently facing a dilemma regarding the concept of international responsibility, as doctrine and jurisprudence are divided regarding State liability for acts committed by non-state agents or persons, particularly where damage occurs as a result of the violation of national and international regulations by private individuals without any formal link with the concerned State.

Hense, the mechanism of accountability of States for offenses committed by private persons raising serious problems in terms of public international law, as international courts showed excessive precautions when liability of a State was about to be engaged, in the absence of a specific positive law instrument regulating the concept of international liability for actions committed by persons with no formal link with the State concerned. Therefore, given that economic and social development and the globalisation process are prerequisites which could affect the environment, endangering both the ecological balance and the proper conduct of international relations, an analysis of how solutions can be found both for responsibility for the damage caused and also for the accountability of other potential attempts against the environment is required, together with the impact that such a cross-border phenomenon may have on the economic system.

For instance, according to a recent Credit Suisse report, it is possible that the Dieselgate scandal could bring significant harm to the German car maker Volkswagen, amounting between 23 and 78 billion euro, i.e. up to 60% more than the cost of environmental disaster that resulted from the oil spill in the Gulf of Mexico in 2010.

As an exapmle, the Erste Group Research division of Romania considers that the negative impact of the Volkswagen scandal in Romania would reduce exports by 2.9 and 1% of GDP only decreased exports of spare parts to Germany, as only two days after the fraud recognition by the German car-maker – more than 11 million cars potentially affected, its market capitalisation fell by almost 35% (26.450 million euros) (Table 1).

It is likely that not only the German producer could lose its reputation, but this scandal could affect more actors, as this loss of reputation will decrease capitalization of machinery industry with direct impact on sales and the whole society will react in case of fraud of this caliber, especially when talking about the second largest German car manufacturer.

As a result, general population could be directly affected by unemployment, not only in Germany, and the decrease in car sales will affect the entire chain of supply, generating that an entire technology standard hardly gained by one of the market leaders, will be lost.

In another vein, one should not lose sight of the fact that some users have been sold cars with a certain degree of pollution declared. In other words, another possible consequence is the impact that pollution from these vehicles has on environment and human health<sup>2</sup>. If it is assumed that the vehicles delivered between 10 and 40 times more pollutants for around 11 million cars, we could find out that the environmental impact is significant against the background in which considerable sums by other users to reduce pollution and environmental protection.

According to Eurostat, EU-28 allocates significant direct expenses are designed to prevent, reduce and eliminate pollution activities or any other source of environmental degradation (Fig. 1). These expenditures can be analysed by studying three components: public sector, industry and public and private enterprises specialised in environmental services<sup>3</sup>.

	Index		Earnings 2015e							
		perfo	performance (%)		index	points	growth (%)		P/E 2015e	
			since							
		31/08	30/06	31/12	current	31/12	current	31/12	current	31/12
DAX 30	9.613	-6.3	-12.2	-2.0	806.9	779.7	14.1	10.2	11.9	12.6
MDAX	19.186	-2.5	-2.2	13.3	1038.0	1053.0	14.1	13.9	18.5	16.1
Euro Stoxx 50	3.080	-5.8	-10.1	-2.1	232.9	242.2	7.3	9.9	13.2	13.0
S&P 500	1.393	-1.7	-6.0	-5.8	117.7	124.7	1.2	7.6	16.5	16.5
Source: Com	merzhan	k resear	ch I/R/	F/S						

Table 1. DAX under intense pressure due to the VW emissions scandal

Source: Commerzbank research, I/B/E/S.

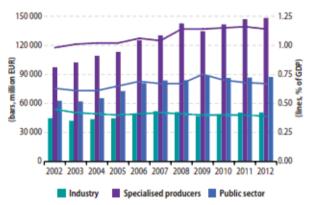
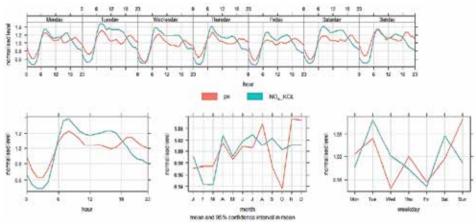


Fig. 1. Total expenses on environment protection Source: Energy, transport and environment indicators, ed. Eurostat Pocketbooks, 2014, p. 245

Data collected in recent years by the Institute for Transport Studies of the University of Leeds reveal that the problem of NO<sub>x</sub> is widespread in the world, and legislation has done little to solve the problem, given that the majority of fleets companies have diesel engines (Fig. 2). These issues are correlated with economic aspects of success in business for diesel automobile manufacturers, but such a decision is closely linked with tackling broader global warming.



**Fig. 2**. PNC and NO<sub>x</sub> time variations at Marylebone Road 2005–2012 Source: Institute for Transport Studies – University of Leeds (http://www.its.leeds.ac.uk/)

Interpretation of graphs shows that diesel engines with direct injection generated significant fuel savings relative to gasoline engines, but increased emissions through emission of particles ultra-fine 'with more than 80% of particles emitted diesel that size falling in the range of around 0.1 pm in diameter'.

Also, data collected by the University indicate that large amounts of particles are made up of organic carbon and poly-cyclic aromatic hydrocarbons, known for their carcinogenic agents.

# EFFECTIVE CONTROL VERSUS GLOBAL CONTROL AND STATES STRICT LIABILITY

Although the institution of liability is a fundamental one for every legal system, the international community has been hit by a series of difficulties in establishing clear legal instruments governing the responsibility of States because of the need to reconcile responsibility with the concept of sovereignty, which makes impossible the use of force in interstate relations.

However, the UN approach towards the concept of State responsibility has allowed a clear distinction between the rules of primary concern that refer to the obligations of States and the secondary rules that relate to liability arising from non-compliance with the primary obligations.

Although a draft of articles on the responsibility of States for internationally wrongful acts was proposed to the UN General Assembly, no multilateral convention to that effect was adopted, but the UN General Assembly recognised the importance of such an instrument and took note that some international jurisprudence related to some of the proposed articles, which imply a mechanism consisting of two components, the unlawful act<sup>4</sup> and the content of the State international responsibility.

The international wrongful act as part of liability presupposes the existence of three mandatory elements which are the State conduct (both positive, through a well-determined action, and negative, by omission<sup>5</sup>). In this latter case, the ICJ noted that international responsibility of Albania was engaged due to the lack of information of the international community about the existence of mines in the Canal (liability by omission), admitting as well the responsibility of the United Kingdom for the action of de-mining the Channel, achieved in Albanian territorial waters without the consent of the coastal State (liability for action).

Therefore, to imputability and breach of a provision of international law two more components can be added, i.e. the existence of a prejudice and a causal link between the wrongful act and the damage occurred. The content of the responsibility, on the other hand, consists of a series of obligations of the responsible State, such as the respect of international law, the end of all acts that generate(ed) liability and the obligation to repair.

However, in the absence of an international instrument adopted by most International community members, we believe now that the mechanism of States international responsibility is governed by a set of principles derived from the international jurisprudence. In 1986, in a decision concerning military and paramilitary activities in Nicaragua<sup>6</sup>, the International Court of Justice established the principle of effective control by the State as condition to engage its international responsibility for acts committed by unofficial State entities, as responsibility can only be considered if a State ordered the respective unlawful acts or had the control of those who commits such unlawful acts.

Basically, this element is supported by the concept of sovereignty, which implies that the State has ultimate control on activities on its territory or performed by its citizens and, consequently, is responsible for any action committed by them.

However, another UN international court, the Permanent Court for the former Yugoslavia established another principle, namely that of global control, with a much wider scope than the actual effective control, allowing thus State *a priori* accountability<sup>7</sup>, as States may act only through individuals which they coordinate both under the personal and the territorial jurisdiction. Still, given the concept of imputability foreshadowed in the UN draft of articles and the relevant decisions of the International Court of Justice, State liability engagement mode will be different.

Therefore, unquestionably in a case such as Dieselgate, although international responsibility of Germany can not be engaged for alleged misconduct by its own officials, as long as those who are guilty *a priori* of the alleged facts are not State officials, this responsibility may nevertheless be engaged by the fact that Germany did not fulfill an obligation of due diligence in environment protection, which was established by the 1992 New York Convention on Climate Change, which

stipulates under art. 2 pt. 3 that '*The Parties should take precautionary measures to anticipate, prevent or minimize the causes of climate change and mitigate its adverse effects*' and under art. 4.2, which stipulates that the developed States commit themselves to adopt national policies and take measures on the mitigation of climate change and thus failed to ensure proper implementation of control measures that could reveal illegal practices of offenders. A 2010 decision of the International Court of Justice in the case regarding the paper pulp factory on the Uruguay River supports this approach.

Today, a new concept of responsibility seems to be the most appropriate in relation to the environment, which is the strict liability concept<sup>8</sup>. However, doctrine and jurisprudence have not yet reached a consensus on this principle<sup>9</sup>, given that this concept can be considered one of an extraordinary nature.

However, the concept of strict liability can be considered as a starting point in developing the concept of international responsibility in the environment field, as it was confirmed as such by the Council of Europe Convention on civil liability for damages resulting from activities considered dangerous to the environment, signed in Lugano on June 21, 1993.

Thus, aiming to ensure the implementation of a mechanism for adequate repair for damages from dangerous activities to the environment, the Convention has a wide field of application, ensuring that this principle applies to all significant risks caused to man and environment (except for nuclear substances or transport activities, covered by other international instruments with European and universal vocation).

In this case, the strict liability regime established by Article 6 of the Convention imposes on States the obligation to ensure that holders of dangerous activities to the environment have the ability to pay for any damage caused by those activities, establishing thus a control mechanism to be set up by States in order to consequently establish the possibility of State liability where this control was not done adequately.

#### CONCLUSIONS

Currently, the fact that accountability for internationally wrongful acts only applies to States, as subjects of international law, no longer meets the needs of the current world characterised both by interaction and globalisation and by the fact that individual actions can create international damages.

In this context, new international law rules are needed, in which individuals should be perceived more as direct international law subjects, thus adapting the actual international law in ways still untested and allowing the strengthening of international environment law, whose rules can engage the responsibility of both States and of individuals for acts committed against the environment throughout rules applicable *erga omnes* to the world, as well as of a repressive or cooperative *modus operandi* that does not depend only on breach of legal rules in order to be implemented.

On the other hand, we believe that it is absolutely necessary, by imposing a special tax, the establishment of a guarantee fund managed by an international institution established under the auspices of the United Nations for serious violations of environment and cover any damage which may incur as a result of international illegal or wrongful acts.

A critical assessment of environmental law indicates that the international community efforts to address climate change are currently counterproductive. There is a certain style of policymaking that emphasise more on economic efficiency and less on maintaining a balance in the environment and health areas.

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Environmental legislation

# VALIDATION IN A NOVEL LAVENDER HARVESTER FOR OIL PRODUCTION

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**Abstract**. This study focuses on the comparison of a new (prototype) lavender harvester, for oil production, to current harvesting methods, hand harvesting and a conventional cutter bar harvester. Following the identification in the work rate and the produced oil quantity and quality, an economical analysis was conducted to demonstrate the cost effectiveness of each harvest method. The proposed method showed that the total cost per litre of oil produced was reduced by £40.90 and £15.30 compared with hand harvest and a conventional harvester, respectively. These prices include the cost of transport assuming that the harvested field was 110 miles (175 km) away from the distillery. It was shown also that the most cost effective operation was not achieved at maximum yield but at a set up in which the machine was capable of harvesting the maximum flower head with the minimum stem percentage.

Keywords: lavender harvester, flower removal, economic analysis.

# AIMS AND BACKGROUND

The introduction of a new method which reduces the total costs in a lavender cultivation for oil production was investigated. Considering the new demands in mechanical lavender harvest for oil production a prototype lavender harvester has been developed employing the detachment technique developed for the harvesting of cereals<sup>1-6</sup>. The harvester works in a unique way for this crop by removing the flower heads in the field, leaving the majority of the stems intact using a rotating drum attached with stripping elements. The applied technique found beneficial for this plant because the most refined lavender oil comes from the flower only as British pharmacopeia suggests<sup>7</sup>.

A prototype lavender harvester was designed, manufactured and optimised<sup>8</sup>. For the validation of the new lavender harvester an economic analysis was established. An economic analysis was conducted to compare the production cost for each harvesting method considering the harvest operating cost. To determine the operating costs, findings of work rate, crop mass and oil yield. This study focuses on the comparison of a new (prototype) lavender harvester for oil production, to

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current harvesting methods, hand harvesting and a conventional cutter bar harvester (CLIER).

# EXPERIMENTAL

The test to compare the work rate, the mass harvest yield and the oil yield for each method was conducted at Yalding area in South England. The hand harvest, prototype, and CLIER harvest methods were evaluated in a plot of 336 m<sup>2</sup>. The same plot was used to conduct the test for the evaluation of the prototype harvester. The total field area was 5.6 ha. The plants of *Lavandula angustifolia Folgate* cultivar were in the 3rd year of growth. The average bush height and width was 0.75 and 0.9 m, respectively with a mean density of 436 plants per m<sup>2</sup>. The mean moisture content was found at 65.9% w.b. The row width was 1.83 m (6 ft). For the hand harvest one adult person was used to harvest the chosen plot areas (Fig. 1). Figure 2 shows the prototype machine during test trials. Figure 3 shows the conventional CLIER harvester



Fig. 1. Hand harvest



Fig. 2. Prototype harvester



Fig. 3. CLIER harvester mounted on a tractor

The conventional harvester (CLIER) was a portable one row harvester machine, and was mounted on the right hand side of the tractor. The tractor was a John Deere 2wheel drive 90 hp. The CLIER machine had a pick up and a cutter bar mechanism with two adjustable lifters at the front. The stalks were gathered by two chains equipped with rubber ridges which guide the crop into the cutter bar where a second cutting mechanism is used to chop and throw the crop into a container on the rear of the machine.

Part of the research was to determine the work rate and the yield in terms of harvest mass and oil yield. For the comparison of the 3 harvest methods a 2 block completely randomised design was used. Four rows (two in each block) were used (Table 1). The total number of treatments examined was four; the hand harvest, the CLIER and two different settings for qualitative and quantitative harvest for

the Prototype. The two examined settings for the Prototype achieved for best oil quality at 310 rpm and for maximum oil yield at 510 rpm drum speed. The higher percentage in harvested flower heads achieved at first set leading in a qualitative harvest and the maximum amount of plant material (flower and stems) was harvested at the second set leading in a quantitative harvest. Four different treatments were replicated twice resulting in a total of 8 measurements.

Harvest method	Row length (m)	Row width (m)	Distance between rows		• Mean harvested mass from	1 2
		. ,	(m)	(s)	each plot	mass)
					(kg)	
Hand harvest	10	0.9	1.83	1940	9.05	0.77
Prototype quality (310)	10	0.9	1.83	27	4.60	1.20
Prototype quantity (510)	10	0.9	1.83	57	6.05	0.97
CLIER	10	0.9	1.83	10	7.35	0.58

Table 1. Data collected from Yalding field trials

Then an economic analysis was conducted to compare the production cost for each harvesting method considering the harvest operating cost<sup>9</sup>. To determine the operating costs, findings of work rate, crop mass and oil yield. The annual costs for each method were divided into 2 categories, namely the fixed and the variable costs. The fixed costs consisted of depreciation and insurance. The variable costs included the fuel and oil, repair, maintenance and labour.

To calculate the hand harvest method the annual cost of the work rate and the cost per hour labour work was used. The hours needed to harvest by hand one hectare was measured during tests and found 539 h. John Nix indicates that each hour of field work costs of field work costs  $\pounds 4.5$  (Ref. 10).

To calculate the fixed costs an estimation of the prototype harvester price was required. The value of £ 15 000 was used. The value was estimated considering the materials price value (£ 5000) and the labour (£ 10000) to build the machine. For this method no insurance was calculated because the prototype was a test machine. For the running costs of the prototype the fuel consumption was measured during harvest and found to be 4.5 l/ha of petrol. For depreciation a 10% straight line reduction of the capital cost was used. For repairs and maintenance 1.5% of the capital cost were used. Two workers were needed to conduct the harvest using the prototype machine.

For the CLIER harvester the total annual cost was a combination of two machines. First was the tractor and second the mounted harvester. To evaluate the total mean annual cost of using a tractor for this operation a 100 hp tractor was used as a basis as these are common available in UK farming systems. The mean hourly cost was multiplied by the harvest duration<sup>10</sup>. The data reference used relates to a 4 wheel drive tractor as these are more commonly available at this engine size. To calculate the annual cost of the mounted harvester a capital cost of £ 20 000 was used<sup>11</sup>. For repairs and maintenance 1.5% of the capital cost was calculated and added to the final cost. One operator was needed to conduct the harvest. For the calculations the harvesters were assumed to work for 10 days (80 h) per season.

To conduct the comparison of the produced oil cost from the 3 different harvest methods a scenario, based on real data supplied from Alec Hunter from a 0.8 ha plot area considering the 2002 harvest season were used<sup>12</sup>. The data used to construct the analysis are shown below:

• Distillery unit: Botanix Ltd. (110 miles (176 km) away from the harvest field);

- Lorry hire cost: £100/day;
- Lorry fuel cost: £0.36/mile (£0.22/km);
- Distillation cost at Botanix distillery = £95/chamber.

Mass of material transported: 1460 kg and filled 1 1/2 chambers  $\approx$  1000 kg/ chamber

# **RESULTS AND DISCUSSION**

The performance results of each method are presented in Table 2. Especially the last two columns demonstrate the calculated produced oil quantity for each harvest method expressed in volume per area and volume per mass harvested plant material.

Harvest method	Forward velocity	Work rate (ha/h)	Harvest mass	Produced	oil quantity
	(km/h)	(110/11)	(kg/ha)	(l/ha)	(l/1000 kg)
Hand harvest	0.0098	0.0018	8145.8	65.91	8.09
Prototype quality (310)	1.3	0.23	4140.4	51.89	12.53
Prototype quantity (510)	0.63	0.11	5445.5	54.45	10.00
CLIER	2.26	0.37	6615.6	40.34	6.09

 Table 2. Performance of the different harvest methods

Table 3 shows the operational costs for each harvest method regarding the fixed and the running costs.

Hand harves	st	The hours needed to harvest by hand 1 ha were measured
		during tests and found as 539 h. John Nix (2004) indicates that each hour of field work costs £4.5 So, the total cost would be: 539 h/ha × £4.5 = £2425.5
Prototype	Prototype quality	Depreciation: £1500/80 h (harvest window) = £18.75/h Repairs and maintenance: 1.5% (of the capital cost) £225/80 h (harvest window) = £2.81/h Fuel and oil: 4.5 l/14 h (harvest time) = $0.32$ l/h 0.32 l/h × £0.78/l = £0.25/h Labour: £4.5/h × 2 workers = £9.00/h Total: £30.81/h £30.81/h/0.23 ha/h (work rate) = £133.9 /ha
	Prototype quantity	Depreciation: £1500/80 h (harvest window) = £18.75/h Repairs and maintenance: 1.5% (of the capital cost) £225/80 h (harvest window) = .£2.81/h Fuel and oil: 4.5 l/14 h (harvest time) = $0.32$ l/h 0.32 l/h × £0.78/l = £0.25/h Labour: £4.5/h × 2 workers = £9.00/h Total: £30.81 /h £30.81 /h (work rate) = £280.00 /ha
CLIER	Tractor	From John Nix (2004) tractor 100 hp 2 wheel drive operation $cost = \pounds 11.23/h$
	Mounted harvester	Depreciation: £2000/80 h (harvest window) = £25.00/h Repairs and maintenance: 1.5% (of the capital cost) £450/80 h (harvest window) = £3.75/h Fuel and oil: £0.00 /h Labour: £4.5/h × 1 worker = £4.50/h Total: £44.48 /h 44.48 £/h/0.37 ha/h (work rate) = £120.21/ha

 Table 3. Harvest operation costs calculation for 3 harvest methods (based on 10 days (80 h) harvest work per season)

Table 4 shows the final costs for each harvest method regarding the cost per ha.

**Table 4**. Determination on produced oil cost (for the calculations the running hours of the distillery was taken as 12 hours per day for peak harvest season)

was taken as 12 hour	s per day for peak harvest season)
Hand harvest	Distillation: 8145.8 kg/1000 kg (of 1 chamber) = 8.14 chambers $\approx 8$
ē	a runs × 95 $\pounds$ /run = $\pounds$ 760
Oil yield: 65.91 L/ha	Transportation: 8145.8 kg/1460 kg (max van fill) = 5.58 van fills (110 miles) $\approx$ 6 trips/3 trips per day = 2 days hire $\times 100 \text{ £/day} = \text{\pounds}200$ (66 mile) $\approx$ 6 trips/6 trips per day = 1.0 days hire $\times 100 \text{ \pounds/day} = \text{\pounds}100$ (1 mile) $\approx$ 6 trips/6 trips per day = 1.0 day hire $\times 100 \text{ \pounds/day} = \text{\pounds}100$ Fuel: (110 miles) 40 £/trip $\times$ 6 trips = £240 (66 miles)66 miles $\times$ 0.36 £/mile = 24 £/trip $\times$ 6 trips = £144 (1mile) £10 Total cost = distillation + lorry hire + fuel (1 mile distance to still) TC = D + LH + F = 760 + 100 + 10 = £870 (66 miles distance to still) TC = D + LH + F = 760 + 100 + 144 = £1004 (110 miles distance to still) TC = D + LH + F = 760 + 200 + 240 = £1200
Prototype quality Mass: 4140.4 kg/ha	Distillation: 4140.4 kg/1000 kg (of 1 chamber) = 4.14 chambers $\approx$ 4 a runs $\times$ 95 £/run = £380
Oil yield:	Transportation: 4140.4 kg/1460 kg (max van fill) = 2.8 van fills
51.89 l/ha	(110 miles) $\approx 3$ trips/3 trips per day = 1.0 day hire $\times 100 \text{ f/day} = \text{\pounds}100$ (1 mile) $\approx 3$ trips/6 trips per day = 0.5 $\approx 1$ day hire $\times 100 \text{ f/day} = \text{\pounds}100$ Fuel: (110 miles) 40 $\text{\pounds/trip} \times 3$ trips = $\text{\pounds}120$ (1 mile) $\text{\pounds}10$ Total cost = distillation + lorry hire + fuel (0 miles distance to still): TC = D + LH + F = 380 + 100 + 10 = \text{\pounds}480 (110 miles distance to still): TC = D + LH + F = 380 + 100 + 120 = $\text{\pounds}600$
	Distillation: 5445.5 kg/1000 kg (of 1 chamber) = 5.44 chambers $\approx$ 5 a runs $\times$ 95 £/run = £475
Oil yield: 54.45 l/ha	Transportation: 5445.5 kg/1460 kg (max van fill) = 3.72 van fills (110 miles) $\approx$ 4 trips/3 trips per day = 1.33 $\approx$ 2 days hire $\times$ 100 £/ day = £200
	$(66 \text{ miles}) \approx 4 \text{ trips/6 trips per day} = 0.66 \approx 1.0 \text{ days hire} \times 100 \text{ \pounds/}$ day = £100
	$(1 \text{ mile}) \approx 4 \text{ trips/6 trips per day} = 0.66 \approx 1 \text{ day hire} \times 100 \text{ \pounds/day} = $ £100
	Fuel: (110 miles) $40\pounds$ /trip × 4 trips = £160 (66 miles) 6 miles × 0.36 $\pounds$ /mile = 24 $\pounds$ /trip × 4 trips = £96 (0 miles) £10
	Total cost = distillation + lorry hire + fuel $(TC = D + LH + F)$
	(0 miles distance to still): $TC = D + LH + F = 475 + 100 + 10 = \text{\pounds}585$ (66 miles distance to still): $TC = D + LH + F = 475 + 100 + 96 = \text{\pounds}671$ (110 miles distance to still): $TC = D + LH + F = 475 + 200 + 160 = \text{\pounds}835$
	to be continued

to be continued

#### Continuation of Table 4

CLIER	Distillation: 6615.6 kg/1000 kg (of 1 chamber) = 6.61 chambers $\approx 7$
Mass: 6615.6 kg/ł	ha runs $\times$ 95 £/run = £665
Oil yield:	Transportation: 6615.6 kg/1460 kg (max van fill) = 4.53 van fills
40.34 l/ha	$(110 \text{ miles}) \approx 5 \text{ trips/3 trips per day} = 1.66 \approx 2.0 \text{ days hire} \times 100 \text{ \pounds/}$ day = £200
	(66 miles) $\approx$ 5 trips/6 trips per day = 0.83 $\approx$ 1.0 days hire $\times$ 100 £/ day = £100
	$(1 \text{ mile}) \approx 5 \text{ trips/6 trips per day} = 0.83 \approx 1.0 \text{ days hire} \times 100 \text{ £/day}$ = £100
	Fuel: (110 miles) 40 $\pounds$ /trip × 5 trips = $\pounds$ 200 (66 miles) 66 miles × 0.36 $\pounds$ /mile = 24 $\pounds$ /trip × 5 trips = $\pounds$ 120 (0 miles) $\pounds$ 10
	Total cost = distillation + lorry hire + fuel ( $TC=D + LH + F$ )
	(0 miles distance to still): $TC = D + LH + F = 665 + 100 + 10 = \text{\pounds} 775$
	(66 miles distance to still): TC=D + LH + F = $665 + 100 + 24 = \text{\pounds} 885$
	(110 miles distance to still): $TC = D + LH + F = 665 + 200 + 200 =$
	£1065

Table 5 shows the final costs for each harvest method regarding the cost per ha and cost per litre of produced oil.

Harvest method		from the llery	Harvest costs (£/ha)	tion and	Total cost (£/ha)	(£/l of
	(miles)	(km)	_	distillation costs (£/ha)		prod. oil)
Hand harvest	1	1.6	2425.5	870	3295.5	50.0
	66	105.6	2425.5	1004	3429.5	52.0
	110	176.0	2425.5	1200	3625.5	55.0
Prototype quality	1	1.6	133.9	490	623.9	12.0
	110	176.0	133.9	600	733.9	14.1
Prototype quantity	1	1.6	280.0	585	865.0	15.9
	66	105.6	280.0	671	951.0	17.5
	110	176.0	280.0	835	1115	20.5
CLIER	1	1.6	120.2	775	895.2	22.2
	66	105.6	120.2	885	1005.2	24.9
	110	176.0	120.2	1065	1185.2	29.4

Table 5. Overall total costs for each harvest method

The results show that the proposed method produced the minimum overall costs. Especially the prototype 'quality' setting up which gave the lowest cost results ( $\pounds$ 623.9/ha) (1 mile distance from the distillery (D.T.DSTL)) and  $\pounds$ 733.9/ha total cost (110 miles D.T.DSTL). The minimum production oil yield then must be 12.5 l/ha for the 1 mile D.T.DSTL and 15.7 l/ha for the 110 miles D.T.DSTL to cover the production costs (if a  $\pounds$ 50/L take into account as market price<sup>11</sup>). On the con-

trary, the maximum total costs gave the hand harvest method with £3295.5/ha (1 mile D.T.DSTL) and £3625.5/ha (110 miles D.T.DSTL). Therefore, the minimum production in oil yield for this method must be 65.9 l/ha for the 1 mile D.T.STL and 72.5 l/ha to cover the total costs. The distance has a great influence in to the total cost. The costs per litre of produced oil shown the effectiveness of the proposed harvest method compared to methods currently in use.

A comparison between the two mechanised harvest methods, Prototype 'quality' and CLIER harvester showed that the prototype needs approximately half the costs to harvest the same field area for the given scenario. The prototype only harvests the flower and leaves the most of the stem. This provides the opportunity to conduct a second pass at a late stage with the purpose of harvesting late blooming flowers. This would only be economic if 1891 kg/ha of flowers remaining after the first pass.

Although the CLIER harvester appears to have an advantage due to faster working speed the economic analysis shows that the prototype 'quality' setting was the most cost efficient harvest method to use for the given scenario examined.

#### CONCLUSIONS

Lower overall operational costs of the proposed harvest method compared to the existing conventional harvesting methods. The total cost per litre of the oil produced was reduced from £55.00 and £29.40 for the hand harvest and conventional mechanised harvest methods respectively to £14.10. These prices include the cost of transport assuming that the harvested field was 110 miles (175 km) away from the distillery. It was shown also that the most cost effective operation was not achieved at maximum yield but at a set up in which the machine was capable of harvesting the maximum flower head with the minimum stem percentage.

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# USE OF GEOPROCESSING TOOLS IN HYDROLOGICAL AND GEOMORPHOLOGICAL ANALYSES FOR THE 'FRUMOASA' LAKE AREA OF THE HARGHITA COUNTY, ROMANIA

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**Abstract**. The objective of this study is to develop a geographic information system which integrates a lot of information and various types of data required for hydrographic and geomorphologic analyses, applicable in the sphere of hydrology and geomorphology. The case study will show how to extract watershed boundaries and determine which basins have higher possibility of flooding by analysis of its shape. The raw data (primary database) will be obtained through vectorisation from which different maps will be gain by analysing the geographic characteristics of the studied area. The paper focuses on pre-meet potential disasters (floods), providing support in data collection and processing by transposing the real geographical problems in acomputer-assisted modelling.

*Keywords*: Geographic Information System (GIS), Digital Elevation Model (DEM), geomorphology, hydrology.

# AIMS AND BACKGROUND

The aim of the study is to implement geographic informational system (GIS) technology in landscape modeling (studying the topographical surface) by numerical methods, having application in hydrology and geomorphology.

The fundamental objective of this study is to preventany disasters and predict in a very clear manner which are the areas with the highest potential risk in the case of floods, focusing on achieving a geographic informational system which integrates all kind of information, all types of data that are needed for hydrological analyses from the automatic extraction of the river basins, watersheds, as well as finding those that have the highest risk of flooding, till study the gradients and orientations of the slopes.

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Using GIS there is the possibility to conduct analyses and correlation of great complexity, which can not be achieved with classical techniques<sup>1</sup>, having in mind that the action of the natural factors is dependent on the local conditions<sup>2</sup>.

Basically floods have a strong impact on environment as well as on local economies<sup>3</sup> and the reason which supports the use of GIS techniques in hydrology is that it leads to achieve the goal in a definitely better manner, being no other practical methods of achieving those objectives.

The softwares used for the study was the ArcGis package, more accurate the *ArcMap* 10.1 program with extensions and *ArcScene* 10.1.

#### EXPERIMENTAL

The study area is located on the territorial administrative unit surface of Frumoasa village, to the East towards the Frumoasa country side, from Harghita County, Romania.

Layers	Entity	Contained spatial elements	Attributes
Level curves	polyline	level curves	elevation
Hydrography	polyline	hydrographic network from the surface	name and type
Residential areas	polygon	limits of the residential areas	name
Lake	polygon	water storage lake of Frumoasa	name

Table 1. Primary graphical database structure

To obtain the digital elevation model (DEM), we vectorise topographic map (which was spatially referenced) at scale 1:25 000 with the increments between contour lines of 10 m and for the remainder graphic database (rivers, creeks, lake, countrysides), we used topographic maps at scale 1:5000 and orthophotomap up to date.

Primary database was established by the followings layers (organised multilevel), which are presented in Table 1.

The first step in the implementation of the database it was represented by procuring cartographic materials, while the second step it was to convert the informations from the analogic raster support to vector format through vectorial entities as points, lines and polygons.

For this study in order to obtain a better set of data in the shortest possible time we used the *ArcMap* 10.1 program with ArcScan extension. To use this extension first the analog rasters maps must be converted into \*.*bmp* format.

The working mode it was an interactive digitising, using the *Vectorisation Trace* function within *ArcScan* extension, which requires the user intervention only in certain cases (e.g. intersections or interruptions of the lines).

This process accomplishes the primary database and from now starts the creating of the derived database.

To understand the steps taken in this study we make a logical schema (Fig. 1) which shows the chaining-mode of the steps that were made.

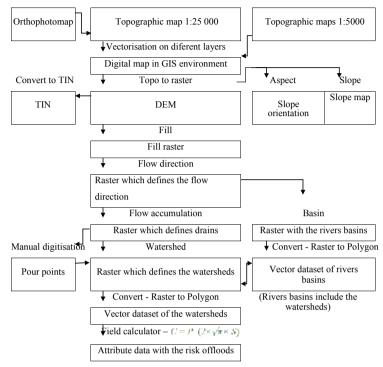


Fig. 1. Schematic of workflow

To view the terrain in 3D and performe the hydrological analyses, must be made the digital elevation model (DEM). This was done using the Topo to Raster function from 3D Analyst tools of ArcMap 10.1 program (Fig. 2).

If we have available the digital elevation model in raster format, we can choose to view it in 3D, but first is required to convert the digital elevation model (DEM) into a TIN (Triangulated Irregular Network) structure using the *3D Analyst* tools. The 3D visualisation of the TIN that was created in previous step it must be done in the *ArcScene* program.

Derived database there was obtained after applying various analyses functions which lead to the exploitation of the DEM obtained in previous phases.

The first analysis was related to hydrography of the area, *ArcMap* offering for users a wide range of functions (Fig. 3) that can be accessed from the main toolbar of the Spatial Analyst.

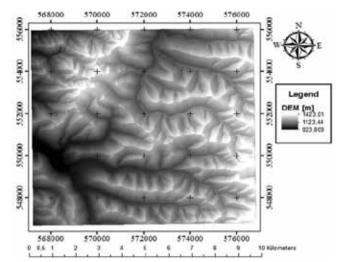
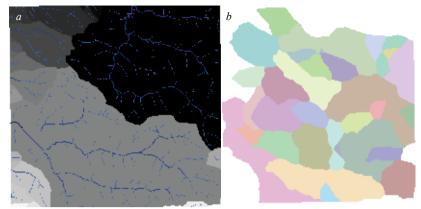


Fig. 2. Digital elevation model in Stereografic 1970 reference system



Fig. 3. Hydrology toolbar

The functions that were used for creating the derived raster database structures, as well as their application order it was: fill – filling depressions; flow direction – determining the direction of drainage water on the slope; flow accumulation – leak accumulation; basin – determining boundary of the rivers basins (hydrographic basins) (Fig. 4*a*); watershed – determining the watersheds (Fig. 4*b*), using the raster obtained after perform the flow accumulation function and from the digitised points where were recorded the highest accumulations of water, those points are located in the nodes of the hydrographic network and they are named 'pour points' – each digitised point will be the lowest pixel of his watershed.



**Fig. 4**. Hydrographic basins (left) -a, watersheds (right) -b

Having the watersheds in raster format, all that remains is to convert them into polygons entities and to calculate their surfaces. To be differentiated between them, the watersheds are labeled with their unique ID (Fig. 5).

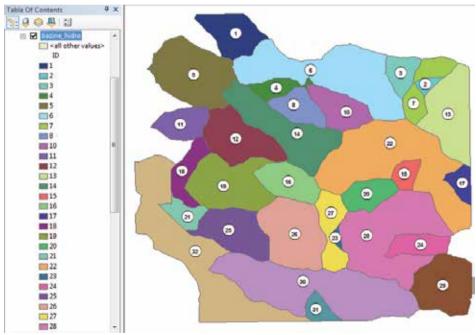


Fig. 5. Identifiers of watersheds

As Bilasco describes in his Ph.D. thesis entitled 'Implementation of GIS in Flood Modeling on Slopes' (2008, Cluj Napoca, Romania)<sup>4</sup> the form of watersheds influences the regime of floods, by the way how are located the tributaries rivers

and creeks in plan. If the watershed shape is closer to a circular one then the flood will be more concentrated. The coefficient of circularity characterised the watersheds circular shape, taking into account the area of the surface and its perimeter<sup>4</sup>.

$$C = \frac{P}{2 (\pi S)^{1/2}}$$
(1)

where C is the coefficient of circularity  $(C \ge 1)$ ; P – watershed perimeter (km); S – watershed area (km<sup>2</sup>).

As the coefficient of circularity is closer to one, the risk of a possible floods is in increase.

Using the *Field Calculator* function from the attributes table of the layer that contains the watersheds, we determined the circularity coefficients of the watersheds by introducing the calculation formula submitted in the relationship one.

Having the values calculated for perimeter (km) and area (km<sup>2</sup>) we determined the circularity coefficients for all watersheds (Fig. 6).

F	ID	Shape *	ID	GRIDC	Sup km	Perim km	Coef circ	
	0	Polygon	1	29	1,75	5,77	1,23	
	1		2	26	0,23	2,42	1,42	
	2	Polygon	3	24	0,85	4,01	1,23	
	3	Polygon	4	22	0,67	5,2	1,79	
	4	Polygon	5	30	3,59	8,15	1,21	
	5		6	23	5,53	13,86	1,66	
	6	Polygon	7	25	1,19	6,49	1,68	
	7	Polygon	8	20	1,17	4,76	1,24	
	8	Polygon	10	21	1,5	5,46	1,26	
	9	Polygon	11	31	1,23	4,5	1,15	
	10	Polygon	12	15	2,54	7,04	1,25	
	11	Polygon	13	28	2,96	7,9	1,29	
	12	Polygon	14	19	3,42	9,89	1,51	
	13	Polygon	15	18	0,55	2,95	1,13	
	14		16	14	1,58	5,14	1,15	
	15	Polygon	17	17	0,56	3,4	1,28	
	16	Polygon	18	11	1,06	5,87	1,61	
	17	Polygon	19	12	3,1	8,82	1,41	
	18		20	9	1,15	4,36	1,15	
	19		21	13	0,7	4,24	1,43	
	20		22	16	6,76	15,99	1,74	
	21	Polygon	23	7	0,14	2,12	1,62	
	22		24	5	1,02	4,82	1,35	
	23		25	1	2,28	6,87	1,28	
-	24		26	10	3,26	7,13	1,11	
	25		27	8	1,36	6,65	1,61	
	26		28	6	5,32	13,49	1,65	
	27	Polygon	29	4	2,57	6,22	1,1	
	28	Polygon	30	2	6,58	14,89	1,64	
	29		31	3	0,53	3,23	1,25	
_	30	Polygon	32	0	6,48	19,38	2,15	

Fig. 6. Attributes table for the *bazine\_hidro* layer

The gradients of slopes was determined using the Slope function, available for users under the Spatial Analyst Tools extension, Surface tools. The tilt of the slopes was expressed in sexagesimal degrees, using the DEM obtained from the level curves (Fig. 7).

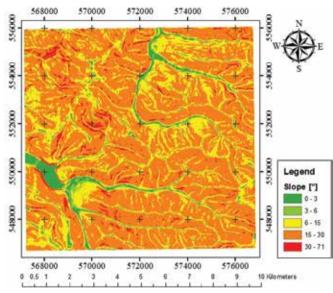


Fig. 7. Average tilt of the slopes in Stereografic 1970 reference system

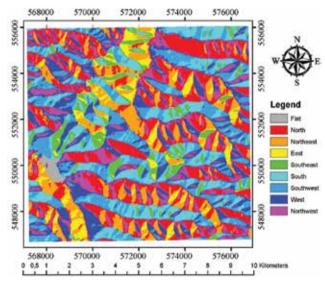


Fig. 8. Slopes orientation in Stereografic 1970 reference system

Slopes orientation is shown in Fig. 8 and it was obtained in a similar way, by applying the *Aspect function* from the *Surface tools* on the DEM obtained through vectorisation.

# **RESULTS AND DISCUSSION**

Analysing the coefficient of circularity we could conclude that the watersheds with the highest risk of floods are with the small values of circularity coefficient (close to the value one and which has a circular shape as shown in Fig. 9*a*), namely 11, 15, 16, 20, 26, 29 while those watersheds whose value of circularity coefficient was bigger (which does not have the geometrical shape close to a circle – see waters hed geometry identified with ID 32 in Fig. 9*b*), the possibility of occurrence floods is less and those watersheds are: 4, 6, 7, 22, 28, 32.

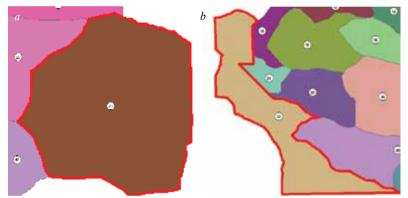


Fig. 9. Circularity coefficient: minimum -a, maximum -b

The coefficient of circularity is the radius of the circle in which the watershead is inscribed. Radius is measured from the gravity centre of the watershead.

The map of slopes and the map of slopes orientations are part of the elevation data, which can be used to perform a variety of cartographic analyses<sup>5</sup>.

Looking at the map of the slopes it can be observed that it is a predominantly mountainous area with an average inclination of slopes situated between the range of 15° and 30°, which was obvious on the topographic map also, given the short planimetric distance between the level curves lines.

## CONCLUSIONS

This study presents theoretical and experimental information for the implementation of a geographic information system (GIS) in order to provide standard products as maps and statistical tables. After storing informations inside of a GIS and applying on them several sets of analyses, is ensuring achieve of the objectives in a categorically superior manner than traditional techniques.

If obtaining methods of the initial data were reasonable than surely the spatial data derived that results after applying the analyses will be also accurate.

Above all was prepared the primary database by vectorisation, and then for the second step we generated the DEM, using elevation data which was attached to the graphic entities. The creation of the DEM was the most important step of the work, because it will serve as input for all future analyses. After applying all the necessary functions that are required for hydrological analyses we obtained the watersheds, followed by calculation of the geometric features thereof (perimeter and area), afterwards we applied the relationship one to find the circularity coefficient of all watersheds, coefficient that is an indicator of the possibility risk of floods.

Given the previously announced things, we consider that the work brings many contributions in terms of how to pickup, process, analyse the information with the purpose of investigating and inventorying the current situation and we consider that the application can be used in further developments.

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# IMPACT OF TOBACCO SMOKE EXPOSURE ON ASTHMA COPD-LIKE PATIENTS

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Abstract. Chronic obstructive pulmonary disease (COPD) is still representing an unknown respiratory disease among smokers. Recently, a greater concern occurs in defining asthma COPD-like cases, including adult onset asthma and asthma-COPD overlap syndrome (ACOS). COPD and asthma are both complex obstructive lung diseases with genetic, smoking and occupational environment interactions. Asthmatic smokers with non-fully reversible airway obstruction are mimicking COPD. A retrospective study was conducted, from January 2010 to 31 March 2016, among asthmatics hospitalised (n = 675) in Constanta Clinical Pneumophthisiology Hospital, Romania. All patients with decreased Tiffeneau-Pinelli index <0.7 were included in study group (n = 31). Data of personal history, type of tobacco smoke exposure (TSE), asthma onset were collected from medical files. Clinical, radiological and lung function were assessed. Three categories of inpatients were identified: 8 former and non-smokers with asthma early onset (before 40-year old) and non-fully reversible airway obstruction; 20 smokers with adult-onset asthma cases progressive FEV1 decline, and 3 cases with ACOS, having a previous history of asthma and smoking  $\geq 10$  pack-year. In conclusion, asthma COPD-like is influenced by active TSE and frequent severe exacerbations are influenced by both active and passive TSE.

Keywords: tobacco-smoke exposure, asthma, adult-onset, chronic obstructive pulmonary disease.

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## AIMS AND BACKGROUND

We are passing through a long-epidemic tobacco smoking era with an overhelming human health problem induced by the emergence of chronic obstructive lung diseases (OLD). Cigarette smoking exposure is considered a 'threat from the first days of life' for people<sup>1</sup>. The impact of smoking exposure in developing chronic obstructive pulmonary disease (COPD) and lung cancer is more suggestive and sufficient than asthma. The consequences of tobacco environmental air pollution on involuntary smokers are still debating. In the last decades of the XX century, it was implemented the concept of 'environmental tobacco smoke' (ETS) and researchers showed the harm of second hand smoking (SHS) on the health of involuntary smokers, by decreasing lung function, but relationship between passive smoking and the incidence of COPD is considered less sufficient studied<sup>2</sup>.

Before the XXth century, asthma was frequently associated with breathless in younger people but, in the last few decades, COPD and adult-onset asthma (AOA) became newer entities. COPD is, now, better understanding as a tobacco smoking-related disease, but its phenotypes or components like emphysema and chronic bronchitis have been previously described by Badham (1814) and Läennec (1821), covering a long medical history of almost 200 years<sup>3</sup>. The condition of asthma coexisting with COPD was, firstly, described as asthmatic bronchitis and, later, AOA became new concept frequently confused with COPD. According to Dutch hypothesis, both asthma and COPD were considered in the 60s as 'a different expression of a single disease', having a common genetic origin<sup>4</sup>. Several decades after, in 1995, the guidelines of American Thoracic Society included a new overlap syndrome named Asthma-COPD Overlap Syndrome (ACOS) and, in the last years, more definitions and algorithmic approaches of it appeared<sup>5</sup>. ACOS is a very challenging concept and researchers are still debating the entity versus phenotype dogma<sup>6,7</sup>. Prevalence of ACOS is underestimated because of the similarities between asthma and COPD diagnosis, a 'complex matrix of airway disease' influenced by the guidelines and criteria of diagnosis, representing 20% of patients diagnosed with OLD (Ref. 7). Tobacco smoke and occupational exposure are considered important risk factors of AOA, which is difficult to be distinguished from COPD, especially in older people<sup>8</sup>.

#### EXPERIMENTAL

The aim of this study was to assess the impact of tobacco smoke exposure (TSE) on defining asthma in adults hospitalised for exacerbations, with an analysis of active TSE versus passive TSE (or environmental tobacco smoke exposure). Study design consisted in a retrospective descriptive analysis of data collected from medical files of asthmatic inpatients aged  $\geq$  40 year-old. The setting of the study was Pulmonary Clinic of Constanta Clinical Pneumophthisiology Hospital,

Romania. The interval of time optimal for study was from January 1st 2010 to March 31st 2016. The target study population referred asthmatic inpatients, aged  $\geq$  40 years (n = 675). Inclusion criteria was persistent decreased Tiffeneau-Pinelli index [post-bronchodilator ratio of force expiratory volume in 1 second (FEV1)/ forced vital capacity (FVC): FEV1/FVC]<0.70. Study group consisted of 31 subjects with asthma mimicking COPD. Clinical and socio-demographic data were collected from medical files. Personal history of atopy including inpatientreported allergic reactions to food, drugs, and/or allergic rhinitis, atopic dermatitis was recorded. Personal history of asthma diagnosis before or after the age of 40, severe exacerbations with hospitalisation in the last 12 months, therapy, persistent airflow obstruction, were evaluated in all medical files. Personal history of  $\geq 10$ pack-years of smoking divided the group of smoker subjects in active or former smokers. Supplementary specific data about TSE among non- or former smokers were collected for a pertinent analysis by a telephone interview of all consenting participants (non-smoker subjects, and former smokers). Occupational risk factors of work-related asthma were assessed, too. ACOS diagnosis was determined according to the 2015 joint of Global Initiative for Asthma (GINA) and Global Initiative for Chronic Obstructive Lung Disease (GOLD) guidelines<sup>9</sup>. Spirometric measures of FEV1 and FVC and ratio FEV1/FVC were performed. FEV1 reversibility was calculated with following required specifications: normal values of FEV1/ FVC pre or postbronchodilator excluded ACOS; FEV1  $\leq$  80% of predicted value was compatible with the severity of airflow limitation; if FEV1  $\ge$  80% of predicted value diagnosis of mild ACOS was considered9. The result of bronchodilator test performed after inhalation of a complete dose of 400 mcg of Salbutamol was considered according to the joint of GINA/GOLD guidelines: positive if FEV1 increased  $\geq$  12% and  $\geq$  200 ml; very positive if more than 400 ml and 15% of FEV1 elevation were noticed on two more occasions; being more suggestive for asthma or ACOS and less for COPD diagnosis9. Severe exacerbations were monitored and counted for every case during the whole interval of study. Frequent exacerbator (FE) was considered any subject with  $\geq 2$  exacerbations by every 12 consecutive months. Pulmonary emphysema was investigated by imaging techniques. Comorbidities were assessed and tuberculosis-like findings on the chest X-ray were considered for active or inactive tuberculosis disease.

Statistical analysis of data was performed by SPSS version 19 and EPI Info. Health outcomes of study subjects were assessed in order to measure the impact of TSE on asthma COPD-like patterns and severity of the disease through exacerbations. Categorical variables were evaluated by chi-square test, continuous variables by ANOVA test and *t*-test for comparison between groups, considering p < 0.005 as significant.

#### RESULTS

675 adults of white ethnicity, diagnosed with asthma, aged over 40 year-old were included in target population, 641 of them had uncontrolled asthma (95%), and 31 subjects mean aged  $59.13 \pm 9.670$  years (limits: 40–83 years) had persistent airflow obstruction with ratio FEV1/FVC < 0.70. Prevalence of asthma mimicking COPD was 5% (n = 31/675). Gender ratio was 1.07/1 in the favour of women (W = 16/M = 15). History of asthma was estimated from 1 to 42 years, with a median of 10 years and a mean of  $14.32 \pm 13.867$  years. Obesity was associated 5 times more frequent in women versus men (n = 5/16; 31.25% versus n = 1/15; 6.66%), coexisting with cardiovascular diseases in 60% (n = 3/5). Occupational exposure was assessed in a few cases (n = 6), with predominance of exposure to cement and asbestos fibers (n = 3), and no one case of diagnosed work-related asthma. Prevalence of active TSE was 51.61% (n = 16/31). Of this cohort study, 22.6% (n = 7/31) were current (active) smokers, 29% (n = 9/31) former smokers and 48.4% (n = 15/31) non smokers. Active smokers reported a history of 44.57  $\pm$ 34.244 pack year (limits from 16 to 120) during their whole lifetime. This mean of pack years of smoking was significantly 2 times greater than in former smokers  $(22.00 \pm 23.016 \text{ with limits from 5 to 80})$  (F = 39.436; p < 0.0001) (Fig. 1). Active smoking was almost 2 times greater in men (n = 10/15; 66%) than in women (n =6/16; 37.5%), in contrast with environmental tobacco smoke exposure, which was 2 times higher in women than in men. (Figs 2a, b) The rate of quitting smoking was 58% (n = 4/7) and, after smoking cessation, exposure to passive smoking was reported by 8 cases, mostly women married to a smoker (n = 6).

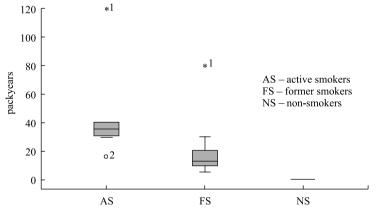


Fig. 1. Tobacco active smoke exposure of cases measured in pack years of smoking

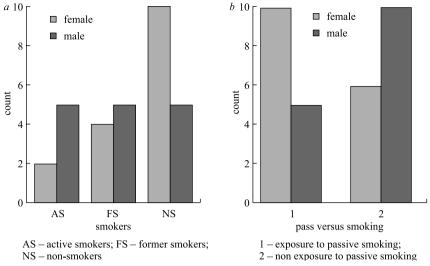


Fig. 2. Exposure to tobacco active (a) and passive (b) smoking among cases (n = 31) by gender

Three categories of asthmatics with tobacco smoke exposure were identified (Fig. 3):

(1) Group 1 of non or former first hand smokers with early onset asthma (before 40 year-old) and the longest history of disease (mean of  $27.67 \pm 5.859$  years; limits: 21-32 years), having a pattern of non-fully reversible airway obstruction (n = 8/31; 25.80%);

(2) Group 2 of AOA active or passive smokers with a shorter history of asthma (mean  $2.40 \pm 2.074$  years; limits: 1–6 years), and a progressive FEV1 decline (n = 20/31; 64.52%),

(3) Group 3 of ACOS cases with a previous long history of asthma (mean of  $27.50 \pm 20.056$  years; limits: 13-42 years), and  $\ge 10$  pack-year cigarette smoking (n = 3/31; 9.68%).

History of personal atopy including inpatient-reported allergic reactions to food, drugs, and/or allergic rhinitis, atopic dermatitis was mostly recorded in group 1 of cases (n = 2/8; 25%). Persistent fixed airflow obstruction was noticed in 17 patients, mostly current and former smokers (n = 11/16; 68.75%) versus nonsmokers (n = 6/15; 40%) (OR = 3.3 (0.7527–14.4687);  $\chi^2 = 2.5005$ ; p < 0.2). In group 3, ACOS diagnosis was facilitated by the worsened symptoms, an increased rate of recurrent hospitalisations and rapid disease progression to the decline of FEV1. Less emphysema was diagnosed in adult-onset asthmatics (n = 2/20; 10%). Lung cancer was diagnosed in a third of ACOS inpatients (n = 1/3, 33.33%) with heavy tobacco smoking exposure. Pulmonary Tuberculosis scars were noticed in 10 patients. Bronchiectasis with *Pseudomonas aeruginosa* recurrent infection were reported in a non smoker patient exposed to second hand smoking by her husband.

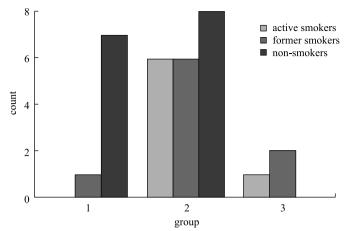


Fig. 3. Categories of cases with asthma COPD-like by tobacco smoke exposure

High risk of hospitalisation for  $\geq 2$  recurrent exacerbations in the last consecutive 12 months was noticed in 58% of subjects (n = 18/31). The mean rate of exacerbations was  $4.48 \pm 4.202$ . The frequency of observed cumulative exacerbations was influenced by TSE and longer history of asthma (Pearson  $\chi^2 = 59.288$ for 36 degree of freedom; p = 0.009). The mean of exacerbations among ETS exposed cases was greater versus non exposed ones (5.80  $\pm$  4.945 versus 3.25  $\pm$ 3.022), but without statistical significance (F = 3.045; p = 0.092). More frequent exacerbators (FE) were identified in group 1 (n = 5/8; 62.5%) and 3 (n = 2/3; 67%) than 2 (n = 7/20; 35%). Distribution of FE by tobacco smoke exposure, revealed a significant higher proportion of nonsmokers in group 1, current and former smokers in group 2 and less FE among active smokers of group 3 (Pearson  $\gamma^2$  = 10.524; 4 df; p = 0.032). Passive smoking or environmental tobacco smoke (ETS) exposure influenced exacerbations among former and non smokers. The influence of ETS exposure in the occurence of FE pattern among grouped cases revealed a significant statistical higher risk among cases from group 1, and lower in group 1 and 3 (Pearson  $\chi^2 = 9.551$ ; 2 df; p = 0.008).

The therapy was different according to the stage of asthma severity and the lack of the symptoms control, with no significant differences between groups (F = 0.372; p = 0.693). All inpatients with exacerbations received supplementary systemic corticoids and antibiotics for every episode of exacerbation. Triple therapy with long-acting beta agonist (LAMA) + long-acting beta agonist (LABA) + inhaled corticosteroids (ICS) was recommended in more than a half of patients (n = 17/31; 54.8%), but patients treated with fixed combinations LABA+ICS (n = 6/31; 19.4%) had the lowest mean of exacerbations ( $1.17 \pm 1.835$ ) (Table 1).

Therapy/exacerbations	Mean	Ν	Std. deviation
LABA+ICS	1.17	6	1.835
LABA+ICS+LTRA	8.00	3	4.359
LAMA+ICS+Theofiline	5.00	1	_
LAMA+LABA+ICS	4.94	17	4.507
LAMA+LABA+ICS+LTRA	4.75	4	3.775
Total	4.48	31	4.202

Table 1. Mean of exacerbations in asthmatic cases at different treatments

LABA – long-acting beta agonist; ICS – inhaled corticosteroids; LAMA – long-acting muscarinic antagonist; LTRA – leukotriene receptor antagonist.

Former smokers and non-smokers experienced more exacerbations than active smokers (4.67 ± 4.123, 5.00 ± 4.943 versus  $3.14 \pm 2.410$ ; F = 0.461; p = 0.635) (Fig. 4). The mean of exacerbations was significantly greater in group 1 (9.38 ± 3.583; limits: 3–15) versus 3 (4.33 ± 5.132; limits: 0–10) and 2 (2.55 ± 2.523; limits: 0–8) (F = 14.147; p < 0.0001). (Fig. 5).

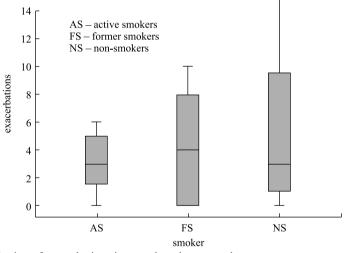
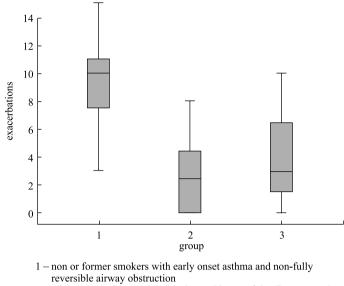


Fig. 4. Distribution of exacerbations in cases by tobacco smoke exposure



- 2 adult-onset asthma cases with shorter history of the disease ± active or passive tobacco smoke exposure, and a progressive FEV1 decline
- 3 asthma overlapping COPD (ACOS) cases with a previous long history of asthma and smoking ≥ 10 pack-years

Fig. 5. Distribution of exacerbations among groups of patients

#### DISCUSSION

Traditional differences in diagnosing asthma versus COPD generally consist in age (younger for asthmatics, older for COPD patients), pattern of inflammation (more eosinophilic inflammation in asthma, and neutrophilic type in COPD), airway limitation (reversible in asthma and progressive and persistent irreversibility in COPD), FEV1 decline (elevated in COPD) (Ref. 6). The natural history of adult-onset asthma (AOA) and ACOS is incompletely known. Physicians are confounded in their current medical practice with a preliminary stage of AOA and COPD knowledge. Previous medical history of asthma in COPD-like patients is still under debate and physicians are awareness about common features of asthma and COPD: increased variability of airflow, bronchial hyper-responsiveness (BHR), severe respiratory symptoms, a more rapid decline in FEV1 and incompletely reversibility of airflow obstruction<sup>7-9</sup>. Work-related lung conditions included asthma<sup>10</sup> and COPD and why not ACOS itself because climatic changes, ETS- and/or work-related ACOS can be a real concern and needs further identification. Adults with asthma are more predisposed to experience irreversible airflow limitation as COPD patients influenced by tobacco smoke11. There are more ACOS cases reported in older people, including lung cancer<sup>12</sup>, increasing progressively with age<sup>13</sup>. Tobacco smoke, including environmental exposure, is a major asthma trigger and its damage produced on the respiratory airways is permanent<sup>14,15</sup>. Smoking morbidity and mortality all over the world is an overwhelming reality but how much exposure to smoke (active or secondhand smoking) or environmental air pollution can increase the risk of COPD overlapping (ACOS) is insufficiently known. ETS may contribute to the origin of ACOS among asthmatics, and smoker asthmatics can gain more pathological similarities to COPD. ETS exposure is considered an aggravating risk factor for asthmatics, increasing the severity of disease, but, surprisingly substantial evidence about ETS effects among adults with asthma is limited<sup>15</sup>. The main component of ETS is the inhaled side-stream smoke which is 4 times more dangerous than the mainstream smoke inhaled by first-hand smokers, containing carcinogens, more toxic substances per gram of total particulate matter<sup>16</sup>. Tobacco smoke exposure influences acute broncho constriction and bronchial hyper-responsiveness (BHR) in asthmatics<sup>17</sup>. Researchers demonstrated BHR is associated not only with ACOS, but with a more rapid FEV1 decline smoking-related<sup>18,19</sup>. Clinical and genetic features of ACOS are still poorly studied<sup>19</sup>. ACOS is a common clinical issue in older people, more than a new entity<sup>13,20,21</sup>. Patients with ACOS are more predisposed to develop exacerbations, so new consensus and guidelines are important for the early recognition of its clinical features, diagnosis, management and assessment of symptoms and disease control<sup>22</sup>. Because of narrow spirometric definition of ACOS, Rhee C.K. tried to characterise several phenotypes of ACOS with different clinical features, prognosis and therapeutic options<sup>23</sup>. Asthma is generally considered a treatment - responsive disease but one main issue of asthma control is tobacco smoke exposure (TSE). In our study case, LABA + ICS fixed combination had more benefit in decreasing the frequency of exacerbations among cases with asthma COPD-like, and, from the perspective of TSE, 3 phenotypes of FE were determined: nonsmokers of group 1 (early onset and longer history of asthma) with a higher risk of FE induced by passive TSE; AOA cases of group 2 with a high risk of exacerbation induced by continuous active TSE; and ACOS former smokers with high risk of exacerbation ETS-related. Clinical implications of TSE can be avoided by supportive smoke free policies of the public opinion and public health authorities. Smoking must be considered a crucial mechanism involved in the phenotyping of asthma, having a bad prognosis of disease. Quitting smoking is recommended as a method of improving the health, quality of life.

## CONCLUSIONS

Asthma COPD-like is influenced by active tobacco smoking exposure and frequent severe exacerbations are influenced by both active and passive TSE. Active smoking tobacco exposure induced a markedly increased risk of  $\geq 2$  exacerbations in the last 12 months among the phenotype of adult-onset asthmatics. Environmental tobacco smoke exposure or second-hand smoking was a particularly risk condition of exacerbations among ACOS former smokers and nonsmokers with an early onset of bronchial asthma. All categories of patients shared features of a few exacerbations if treated with fixed combination of long-acting bronchodilators and inhaled corticosteroids.

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# ROMANIAN ASBESTOS INDUSTRY, OCCUPATIONAL EXPOSURE AND DELAYED DIAGNOSIS OF A MALIGNANT PLEURAL MESOTHELIOMA – CASE REPORT AND LITERATURE REVIEW

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**Abstract**. Asbestos fibre cumulative exposure is influenced by industrialisation, environmental conditions of pollution, may vary in magnitude in different area of the world. The latency of mesothelioma may extent to more than 40 years after the first year of exposure to asbestos fibres. A late onset of malignant pleural mesothelioma (MPM) is reported in a 68 year-old, non smoker, white woman, with prolonged ignored respiratory symptoms. She worked as a lab chemist in a heavy cement manufacturing and asbestos industry, placed in Medgidia, a town of South Eastern Romania, from 1964 to 1990. She was diagnosed with lung fibrosis asbestos related in 1986 and, 29 years later, in 2015, a recurrent left pleural effusion. Since first year of employment, 51 years of retention time of asbestos fibres were estimated. Persistent painful left chest and progressive dyspnea represented clues for a high suspicion of a pleural asbestos related disease. Morphologic assessment of a left pleural mass, facilitated by videothoracoscopy surgery and biopsy, and immunochemistry exam of pleural samples revealed MPM. In conclusion, a 29 years lag time from the onset of lung fibrosis asbestos related (1986) to the MPM delayed diagnosis (2015) were noticed, with 6 months survival.

Keywords: asbestos industry, pleural asbestosis related disease, malignant pleural mesothelioma.

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### AIMS AND BACKGROUND

The exposure of general population to air pollution is a reality of our times. The impact of respirable air pollutants is huge not only on the environment and climate but also on human health. Pollution of the air is greater in industrialised countries and developing countries and air quality assessment of basic and specific pollutants is difficult to be routinely performed<sup>1</sup>. Outdoor air pollution with asbestos from natural sources has been analysed, in Romania, by A.M.Moldoveanu and author evaluations show that 'general population is exposed to low levels of asbestos primarly by inhalation', but the concentrations of asbestos fibres in Romania are ranging from below 0.1 ng/m<sup>3</sup> in rural areas to over 100 ng/m<sup>3</sup> in urban areas near specific industrial sources', such as asbestos mines and factories<sup>2</sup>. Asbestos fibres cumulative exposure is influenced by industrial development, environmental conditions of pollution, and may vary in magnitude in different area of the world and researchers considere that asbestos related diseases are strongly dose dependent<sup>3</sup>. The delayed diagnosis of asbestosis is influenced by many years of latency from the first exposure to asbestos fibres till the first respiratory symptoms and later clinical recognition of the disease, especially in patients with pleural involvement. Pleural disorders asbestos related may coexist or not with lung involvement as fibrosis and/ or bronchogenic carcinoma<sup>3</sup>. Lung cancer is frequently associated with secondary malignant pleural effusions, but, in carcinoma asbestos-related even bilateral diffuse pleural thickening can be diagnosed. Mesothelioma is usually a primary malignancy of the pleura and/or pericardium, or peritoneum. Its association with lung cancer is reported mainly in asbestos occupational exposed individuals<sup>4,5</sup>. Additional occupational interview for asbestos fibres is important to be assessed in all past exposed workers. Amphibole fibres of asbestos, known as straight fibres, are more likely to induce the occurrence of mesothelioma than chrysotile ones (curly fibres)<sup>6</sup>. The onset of the disease needs a lag time of occurence between 25 to 40 vears, even longer<sup>3</sup>. The incidence of mesothelioma is lower among females (2 per 1 million annually cases) and 5 times greater among males<sup>3</sup>.

Categories of malignant pleural mesothelioma (MPM) are characterised by heterogenous phenotypes. Histopathologically, they are clasified into epithelioid (50%), sarcomatous or mesenchymal or fibrous (16%) and mixed type (34%), and positive histologic diagnosis by paraaminosalicylic acid staining is considered difficult because of the similarities between epithelial mesothelioma and adenocarcinoma metastases in pleural area<sup>3,7,8</sup>. Prognosis is influenced by the morphologic subtype of MPM, being better for epithelial type<sup>3,7,9</sup>. The morphologic and immunohistochemical (IHC) assessement of malignant pleural lesions must be done particularly in patients with mesothelioma asbestos related<sup>10–12</sup>.

## CASE REPORT

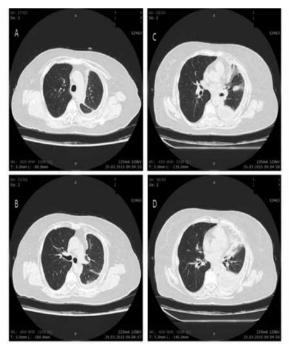
We are presenting a case report of a 67 year-old white woman, non smoker, diagnosed with lung fibrosis asbestosis related in 1986, and delayed MPM in March 2015. Occupational asbestos exposure and 51 years of asbestos fibres retention time were evaluated from 1964, the 1st year of employment in a heavy industry of cement and asbestos from Medgidia, Romania, till 2015, and showed 23 years of exposure to dust with cement, and blue and white asbestos fibres. The exposure to crocidolite, as blue asbestos fibres, and chrysolite, as white asbestos fibres was considered from 1964 to 1990, excluding 3 years of patient retirement from 1986 to 1988.

Patient first admission in Pulmonology Clinic of Constanta, Romania, was in December 22, 2014, for left persistent chest pain, accompanied by progressive chronic dyspnea, and important loss in weight (13 kg in the last 2 months). Computed tomography (CT) scan of the chest performed in December 11, 2014, revealed bilateral bronchiectasis in lower lobes and lingula, diffuse interstitial fibrosis and no pleural involvement. After 11 days, chest radiograph, ultrasound examination and left thoracentesis confirmed a left small pleural effusion (Fig. 1), with an exudative hemorrhagic lymphocyte-rich fluid, a mild increased level of glucose and adenosine deaminase (ADA) (48 U/l versus normal level of 33 U/l). Smears and cultures of both sputum and pleural fluid were negative for *Mycobacterium tuberculosis* and/or other germs. After 14 days, spontaneous resolution of pleural fluid was noticed.

In January 2015, a moderate restrictive ventilatory defect was diagnosed by lung function testing and, two months later, in March 2015, patient became pale with gradual alteration of general condition, chronic dry cough, progressive dyspnea and extremely painful left chest pain. For recurrent moderate left pleuresy and lung fibrosis asbestos-related, contrast enhanced CT chest was performed again and revealed bilateral circumferential nodular pleural thickening with relapsed left small pleural effusion (Fig. 2).



**Fig. 1**. Chest X-ray exam, March 23, 2015 (performed in a white woman, 68 year-old, a former worker in a cement and asbestos industry, showing lung fibrosis, left pleural effusion, and bilateral nodular area of thickening pleura)



**Fig. 2**. Chest CT exam, March 25, 2015 (performed in a white woman, 68 year-old, a former worker in a cement and asbestos industry, showing lung fibrosis, left pleural effusion, and bilateral nodular area of thickening pleura)

Left thoracentesis showed an exudative serohemorrhagic pleural fluid with erythrocytes, neutrophils, lymphocytes, and eosinophils detected by May Grunwald Giemsa (MGM) staining. Left videothoracic surgery and pleural biopsy were performed.

Morphologic examination of collected parietal pleural samples by hematoxylin-eosin (HE) staining revealed with tumoral proliferation of acinar (glandular) epithelioid mesothelioma in a mass of fibrous myxoid tissue, with hialin and hemorragique areas was sugestive for epithelioid pattern of MPM (Fig. 3). Several fragments of pleura were infiltrated by tumoral cells with proliferative biphasic character, sarcomatoid and epithelioid, with a storiform pattern of spindle cells, epithelial like papillary tumors, cells with eosinophilic cytoplasm and foci of sarcomatous components (Fig. 3).

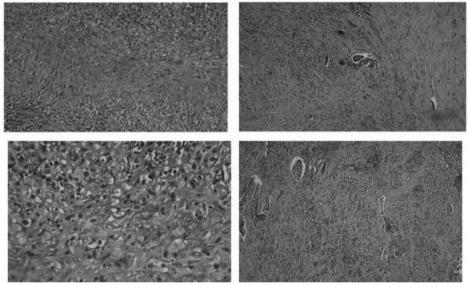


Fig. 3. Biphasic malignant mesothelioma with epithelioid tumor and foci of sarcomatous components with frequent mitosis, clusters of epithelioid cells, round enlarged nuclei and proeminent nucleoli, revealed by morphologic exam of pleural parietal samples

Immunohistochemical (IHC) tests were also performed for providing the differential diagnostic criteria between mesothelioma and pleural metastases of adenocarcinoma and supported the diagnosis of MPM diffuse biphasic subtype. Mesothelioma markers (calretinin, mezotelina) were positive. Adenocarcinoma markers (carcinoembrionic antigen (CEA) and thyroid transcription factor 1 (TTF-1)) were negative.

### **RESULTS AND DISCUSSION**

The effects of asbestos exposure were analysed in the last century by the Romanian Institute of Public Health Bucharest in the late 1970s and 1980s and several years later, in 1996, the Romanian Health Ministery and Occupational Medicine Regulations established a limit for exposure to air-borne asbestos fibres in the occupational atmospheres to be < 1 fibre/cm<sup>3</sup>, because in 1980, in Romanian areas with asbestos cement production, the level of occupational exposure was ranging from 3.5 to 5 fibres/cm<sup>3</sup> (Ref. 13). In the last century, Banat area of Romania was more representative for asbestos mining versus Gorj and Constanta which were mostly involved in heavy industrial production of asbestos and cement product categories. Today, asbestos mining and industry are only of historical interest but patients with asbestos related diseases are on going.

Medgidia industry of cement and asbestos production was placed in Constanta county in the South Eastern part of Romania and, after 1990, its activity had been gradually stopped till 2007. Local industry of Medgidia developed after the second war world by producing terracotta and roof tile kilns. Cement manufacturing factory was later developed into a more complex industry for producing binders and asbestos cement products. Medgidia was a city with an important high environmental pollution of the respirable air because of its industrial source. After 1990, several factories of Medgidia were closed down and others were reorganized, and became privatised.

Diagnosis of pleural asbestos related diseases must be suspected in older age categories of patients with chronic occupational exposure and identified diffuse pleural thickening with significant restrictive ventilatory impairment, circumscribed or localized pleural plaques with or without calcifications, round atelectasis as an unique form of pleural thickening localised on the posterior face of lower lobes, pleural bloody fluid, pleural exudate with predominance of polymorphonuclear lymphocytic or eosinophil cells<sup>3,7</sup>. Diffuse pleural fibrosis has no clearly defined margins, involves both visceral and parietal pleura, consists in a deposit of collagen and hyperplasia of mesothelial cells<sup>7</sup>. Mesothelioma can be localized and asymptomatic with no pleural effusion versus diffuse type which is always a malignant pleural disorder, accompanied by persistent chest pain with pleural effusion or multiple small nodules which extend from parietal to visceral pleura, penetrate into the lung and pericardium.<sup>7</sup> Mesothelioma is declared an occupational disease in Romania<sup>13</sup>. It is less reported in females<sup>3</sup>.

We reported a case in a 68 year-old woman with a progressive asbestosis disease in the absence of continued asbestos exposure after 1990. Patient medical history included allergic rhinitis and bronchial asthma since 1985, blood hypertension since 2000, with no personal and/or familial history of tuberculosis or cancer. For progressive dyspnea and left chest pain, a chest CT scanning was recommended in December 11, 2014, providing almost inconclusive data of a diffuse lung fibrosis, and bronchiectasis with no abnormal pleura. MPM of left hemithorax was 3 months later suspected. Final diagnosis was not in the favour of an well differentiated epitheliod type of MPM. Prognosis is influenced by the morphologic subtype of mesothelioma, being worser for the biphasic type. In our study case reported, patient survived only 6 months.

The delayed diagnosis of asbestosis is ussually influenced by many years of latency from the first exposure to asbestos fibres to the first respiratory symptoms and final recognition of the disease, especially in the pleural involvement.

Reported case of an asbestos and cement fibres exposed female worker is referring to an old carrier of a previous lung fibrosis and provided diagnosis of asbestosis since 1986. Although her asbestos related disease was firstly diagnosed 29 years ago, based on diffuse intestitial lung fibrosis, it was not reported at that time because of Romanian communist political issues. Later, the diagnosis of asbestosis was ignored both by the patient, general practitioner and other physicians. Her persistent respiratory symptoms associated with restrictive ventilatory impairment, and recurent left pleural effusion were very lately suggestive for a pleural involvement asbestos-related. Misunderstanding the occupational etiology of persistent breathing problems and prolonged left chest pain was influenced by her medical history of asthma, bronchiectasis, blood hypertension, and postponed the diagnosis of asbestosis disease for almost 3 decades: 29 years lag time since the first evaluation of lung fibrosis (1986) and obvious clinical presentation of the pleural asbestos-related disease.

Statistics showed that more than 50% of patients diagnosed with benign pleural asbestos disorders have a left chest side predominance<sup>3</sup>. In presented case report, one year of chronic chest pain preceded left pleural involvement. Chronic chest pain could be a suggestive feature of a pleural involvement with diffuse pleural thickening<sup>14</sup>. Unexplained pleural effusion, even if mesothelial cells are not present in pleural fluid, must rise the suspicion of MPM, mainly in individuals with persistent chest pain<sup>15</sup>. Pleural lesions were better observed in April 2015, having a prolonged lag time of occurence to 51 years, longer than 40 years generally accepted<sup>3</sup>. CT plays an important role in providing morphologic appearance of MPM, but magnetic resonance imaging (MRI) is considered superior<sup>16</sup>. Histologic features of MPM patterns consist in cells with different form (polygonal, oval or cuboidal) or spindle type<sup>17</sup>.

The approach to malignant pleural lesions must included IHC assessment because electron microscopy has its own limits induced by the heterogenity of MPM and overlapping adenocarcinoma. Immunohistochemical testing with at least 2 mesothelial and 2 carcinoma markers are recommended by International Mesothelioma Interest Group for distinguishing reactive mesothelial hyperplasia from epithelioid mesothelioma, fibrous pleuresy from desmoplastic variant of sarcomatoid mesothelioma, and both of other malignant pleural lesions<sup>17</sup>. Malignant mesothelioma is generally identified by antibodies against calretinin, mesothelin and vimentin, but calretinin is considered a higher sensitive marker than mesotheline<sup>18</sup>. Correlation between positive markers of MPM with no reactivity to TTF-1 and CEA is a very strong immunohistochemical issue for the positive diagnosis of MPM (Refs 11 and 12). In conclusion, immunohistochemical (IHC) examination is a very usefull method for certain identification of MPM.

The histopathologic findings and immunohistochemical tests supported in the reported case the diagnosis of MPM diffuse subtype biphasic epithelial and sarcomatoid classified by World Health Organisation (WHO) in International Classification of Diseases (ICD) for a better epidemiological surveillance with the code ICD -O (WHO): 9053/3 (Ref. 19).

Prognosis of patients is influenced by morphologic type, being better for epithelial mesothelioma, which is the most common type, and worse for sarcomatoid type<sup>3,7,9</sup>. Human MPM is considered one of the most aggressive tumors and mixed tumors have more unfavourable prognosis. In this reported case of biphasic type of MPM, the outcome of the survival limited to extra 6 months after diagnosis was determined.

Human activity in an industrialised era are a continuos source of pollutants<sup>20</sup>. Even immigration phenomenon can cause a transboundary pollution of waters with asbestos<sup>21</sup>. Preventive maintenance of industrial building is a new concept, very important for limiting air pollution and avoiding occupational exposure<sup>22</sup>.

#### CONCLUSIONS

Most patients are unaware and/or uninformed about their occupational exposure to asbestos, avoiding to describe their professional history. Assessment of asbestos exposure on human health is important to be sistematically performed in all former workers. Delayed diagnosis of malignant pleural mesothelioma asbestos related was revealed by the recurrence of a small pleural effusion in an old woman, former worker in a heavy cement and asbestos industrial area of Medgidia, Constanta county, Romania. Prolonged asbestos fibres retention time of 51 years was associated with a biphasic type of mesothelioma and worse prognosis.

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Public health – environmental medicine

# COMPARATIVE HEPATIC EVALUATION AND THE LIPID PROFILE IN DANUBE DELTA PATIENTS WITH FISH-BASED DIETS

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Abstract. According to literature data, the normal values of biochemical parameters in blood vary by sex, age, geographical region and type of diet. The aim of this study was to analyse the benefits of a fish-based diet among the population of Sulina Town, in the Danube Delta, by performing a comparative hepatic evaluation and the lipid profile of patients. It is acknowledged that a fish-based diet is rich in high quality protein, it is an important source of Omega 3, vitamins and minerals, it helps to lower blood pressure and decrease levels of triglycerides, it regulates total cholesterol, HDL- high-density lipoproteins ('good' cholesterol) and LDL- low-density lipoproteins ('bad' cholesterol) values and total serum lipids. The batch of patients underwent a set of biochemical tests in the RoutineMed Laboratory of Sulina. The novelty of the research is represented by the geographic area covered, as the Danube Delta had no medical analysis laboratory until 2010, when RoutineMed Laboratory was opened in Sulina. Blood samples were collected from 200 patients for the evaluation of the liver parameters: aspartate aminotransferase (AST), alanine aminotransferase (ALT), total lipids, total cholesterol, high-density lipoprotein (HDL) cholesterol, low-density lipoprotein (LDL) cholesterol, and triglycerides. Both women and men were involved in the research and patients were grouped into age ranges: 20-40 years, 40-60 years, > 60 years. The study included 200 patients, 97 male (48.5%) and 103 were female (51.5%), who declared they eat fish or fish-based products at least once a week. The values obtained were statistically analysed using the SPSS v. 20 software and then compared to the ranges considered normal for these parameters. The results obtained showed that patients with a fish-based diet seem to be healthier than those with a diet in which fish meat is scarce, as their blood biochemical parameters values are closer to normal, which leads to the conclusion that including fish and fish products in people regular diet is beneficial in preventing lipid metabolism disorders and preserving the overall health of the liver.

*Keywords*: lipid profile, aspartate aminotransferase (AST), alanine aminotransferase (ALT), highdensity lipoproteins (HDL), low-density lipoproteins (LDL), triglycerides, fish.

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## AIMS AND BACKGROUND

The aim of this study was to analyse the benefits of fish diet in the Danube Delta, Sulina population. Based on literature data, normal values of biochemical parameters vary by gender, age, diet and geographical region and we wanted to determine if the fish-based diet really helps maintain normal levels of lipids and other analytes at the biochemical level<sup>1–3</sup>. The nutrition and endocrine diseases or the screening of healthy people are made trough sets of laboratory analyses such as: glucose, triglycerides, lipoproteins, ionogram, total calcium, cholesterol, transaminases, proteins, phosphorus, magnesium, hemoglobin, iron<sup>4</sup>. The liver is the most important seat of degradation and synthesis of lipids metabolism, but the main role of liver is in glucose metabolism<sup>5</sup>. Transaminases are cellular enzymes that act exclusively at intracellular level. Alanine aminotransferase (ALT) is found in the cytoplasm, aspartate aminotransferase (AST) both in the cytoplasm and mitochondria<sup>6</sup>.

The Danube River ecosystem is dominated by fish (about 160 species, of which 75 are freshwater) forming local and migratory fish fauna<sup>7,8</sup>. Fishing is one of the oldest occupations of the Danube Delta population and still represents an important economic activity engaging more than half of the active Delta population<sup>9</sup>. Thus, fish represents a major source of food for local inhabitants. The consumption of fish and fish products in Romania is estimated at 6.3 kg/inhabitant/year (Ref. 10). However, there are indications that the consumption is increasing, mainly due to the health benefits associated with the consumption of fish.

### EXPERIMENTAL

The novelty of the research is represented by analysed geographic area. The Danube Delta had no medical analysis laboratory until 2010, when the RoutineMed Sulina laboratory was opened. Patients were subjected to a set of biochemical tests in the RoutineMed Sulina laboratory. They all declared they eat fish or fish-based products at least once a week. Tests were performed on 200 patients for evaluation of the liver: AST, ALT, AST/ALT, total lipids, total cholesterol, HDL cholesterol, LDL cholesterol, LDL/HDL, total cholesterol/HDL, triglycerides. Blood was collected and analysed during September 2013–May 2016. For serum biochemistry the following measuring instruments were used: Rotofix 32 A centrifuges, SAP-PHIRE 350 automatic biochemistry unit and Audit Diagnostics reagents<sup>11</sup>. The experimental data were processed using IBM SPSS Statistics 20. The procedures used were: descriptive statistics (characterisation variables discrete and continuous defined in the database), graphs, statistical tests parametric (*t*-test to compare the average of two independent samples, *t*-test to compare the average of a sample value specified test One-Way ANOVA), correlation analysis<sup>12–15</sup>.

#### **RESULTS AND DISCUSSION**

The study included 200 patients, 97 were males (48.5%) and 103 were females (51.5%) (Fig. 1). The distribution by age groups was: 53 were in the age range (20–40) years (26.5%), 79 were in the age range (40–60) years (39.5%) and 68 were in the age range (60–...) years (34.0%). Of the 200 patients included in the study, 97 were males (48.5%): 19 men (19.6%) were aged (20–40) years, 40 men (41.2%) were aged (40–60) years and 38 men (39.2%) were aged (60–...) years; 103 patients were females (51.5%): 34 women (33.0%) were aged (20–40) years, 39 women (37.9%) were aged (40–60) years and 30 women (29.1%) were aged (60–...) years (Fig. 2).

The determination of cholesterol and lipid metabolic disorders assesses their status, the risk of atherosclerosis, coronary stenosis and myocardial infarction.

HDL cholesterol is a lipoprotein group synthesised and secreted by hepatocytes. It is recommended to be checked regularly, in order to accurately assess the risk of developing atherosclerosis. HDL has an important role in cholesterol metabolism, participating in its transportation from extrahepatic tissues to the liver for excretion and catabolic<sup>16</sup>. LDL cholesterol is involved in the transport of cholesterol to tissues, mainly in the arterial system, which explains the high incidence of atherosclerosis and coronary heart disease in patients with elevated serum levels of this lipoprotein<sup>17</sup>. So, LDL determination is specific for predicting cardiovascular risk and establishing therapeutic decision<sup>18</sup>.

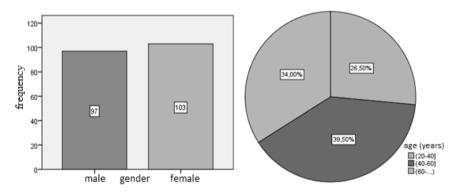


Fig. 1. Patient distribution by age and gender

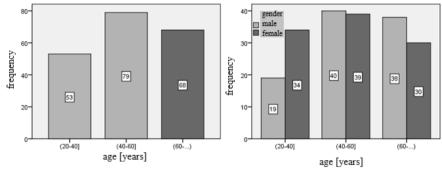


Fig. 2. Patient distribution by age groups

Average value of total lipids – males. For the group (20–40) years (N = 19) the average of total lipids is  $M_{(20-40)} = 450.42 \text{ mg/dl}$  and standard deviation  $\text{SD}_{(20-40)} = 69.37 \text{ mg/dl}$ ; in group (40–60) years (N = 40) the average value of the total lipids is  $M_{(40-60)} = 499.45 \text{ mg/dl}$  and the standard deviation  $\text{SD}_{(40-60)} = 123.40 \text{ mg/dl}$ ; in the group (60–...) years (N = 38) the average value of the total lipids is  $M_{(60-...)} = 458.68 \text{ mg/dl}$  and the standard deviation  $\text{SD}_{(60-...)} = 100.56 \text{ mg/dl}$ . Average values of total lipids measured at the three age groups showed no significant differences (F = 2.02;  $p = 0.138 > \alpha = 0.05$ ) (Fig. 3*a*).

Average value of total lipids – females. For the group (20–40) years (N = 34) the average of total lipids is  $M_{(20-40)} = 412.23 \text{ mg/dl}$  and standard deviation  $\text{SD}_{(20-40)} = 74.03 \text{ mg/dl}$ ; in group (40–60) years (N = 39) the average value of the total lipids is  $M_{(40-60)} = 468.15 \text{ mg/dl}$  and the standard deviation  $\text{SD}_{(40-60)} = 92.76 \text{ mg/dl}$ ; in the group (60–...) years (N = 30) the average value of the total lipids is  $M_{(60-...)} = 497.36 \text{ mg/dl}$  and the standard deviation  $\text{SD}_{(60-...)} = 75.24 \text{ mg/dl}$ . Mean total lipids measured at three age groups differ significantly for at least two of the age groups considered (F = 9.05;  $p < 0.001 < \alpha = 0.05$ ) (Fig. 3*a*).

Average value of total cholesterol – males. For the group (20–40) years (N= 19) the average total cholesterol is  $M_{(20-40)}$  = 177.73 mg/dl and standard deviation SD<sub>(20-40)</sub> = 33.41 mg/dl; in group (40–60) years (N = 40) the mean value of total cholesterol is  $M_{(40-60)}$  = 182.65 mg/dl and the standard deviation SD<sub>(40-60)</sub> = 36.03 mg/dl; in the group (60–...) years (N = 38) total cholesterol is an average value of M<sub>(60-...)</sub> = 178.07 mg/dl and the standard deviation SD<sub>(60-...)</sub> = 38.93 mg/dl. The mean total cholesterol measured at the three age groups showed no significant differences (F = 0.192; p = 0.826 >  $\alpha$  = 0.05) (Fig. 3b).

Average value of total cholesterol – females. For the group (20–40) years (N = 34) the average total cholesterol is  $M_{(20-40)} = 175.35$  mg/dl and standard deviation  $SD_{(20-40)} = 34.09$  mg/dl; in group (40–60) years (N = 39) the average value of total cholesterol is  $M_{(40-60)} = 192.30$  mg/dl and the standard deviation  $SD_{(40-60)} = 31.74$  mg/

dl; in the group (60–...) years (N = 30) the average value of total cholesterol is  $M_{(60-...)} = 203.66$  mg/dl and the standard deviation SD<sub>(60-...)</sub> = 27.28 mg/dl. Mean values of total cholesterol measured at three age groups differ significantly for at least two of the age groups considered (F = 6.66;  $p = 0.002 < \alpha = 0.05$ ) (Fig. 3*b*).

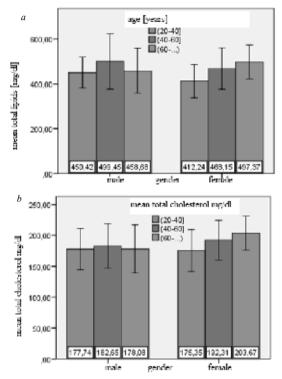


Fig. 3. Average value of the total lipids (a) and total cholesterol (b) in males and females by age groups

Average value of HDL cholesterol – males. For the group (20–40) years (N = 19) the mean HDL cholesterol is  $M_{(20-40)} = 50.10$  mg/dl and standard deviation  $SD_{(20-40)} = 11.15$  mg/dl; in group (40–60) years (N = 40) the average HDL cholesterol is  $M_{(40-60)} = 47.90$  mg/dl and the standard deviation  $SD_{(40-60)} = 12.29$  mg/dl; in the group (60–...) years (N = 38) the average HDL cholesterol is  $M_{(60-...)} = 51$  mg/dl and the standard deviation  $SD_{(60-...)} = 12.17$  mg/dl. The mean values of HDL cholesterol measured at three age groups showed no significant differences (F = 0.672;  $p = 0.513 > \alpha = 0.05$ ) (Fig. 4*a*).

Average value of HDL cholesterol – females. For the group (20–40) years (N = 34) the mean HDL cholesterol is  $M_{(20-40)}$  = 50.17 mg/dl and standard deviation SD<sub>(20-40)</sub> = 12.78 mg/dl; for the group (40–60) years (N = 39) the average HDL cholesterol is  $M_{(40-60)}$  = 55.53 mg/dl and the standard deviation SD<sub>(40-60)</sub> = 11.48 mg/dl; in the

group (60–...) years (N = 30) the average HDL cholesterol is  $M_{(60-...)} = 59.16$  mg/dl and the standard deviation SD<sub>(60-...)</sub> = 11.86 mg/dl. The mean values of HDL cholesterol measured at three age groups differ significantly for at least two of the age groups considered (F = 4.55;  $p = 0.013 < \alpha = 0.05$ ) (Fig. 4*a*).

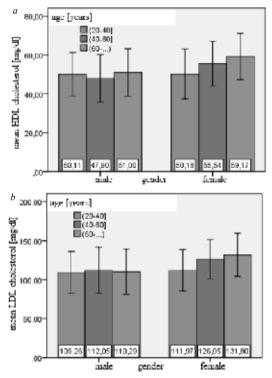


Fig. 4. Average value of the HDL (a) and LDL (b) cholesterol in males and females by age groups

Average value of LDL cholesterol – males. For the group (20–40) years (N = 19) the mean LDL cholesterol is  $M_{(20-40)} = 109.26$  mg/dl and the standard deviation SD<sub>(20-40)</sub> = 26.97 mg/dl; in group (40–60) years (N = 40) LDL cholesterol is  $M_{(40-60)} = 112.05$  mg/dl and the standard deviation SD<sub>(40-60)</sub> = 29.73 mg/dl; in group (60–...) years (N = 38) LDL cholesterol is  $M_{(60-...)} = 110.28$  mg/dl and the standard deviation SD<sub>(60-...)</sub> = 29.10 mg/dl. The mean values of LDL cholesterol measured at three age groups showed no significant differences (F = 0.07;  $p = 0.933 > \alpha = 0.05$ ) (Fig. 4b).

Average value of LDL cholesterol – females. For the group (20–40) years (N = 34) the mean LDL cholesterol is  $M_{(20-40)} = 111.97$  mg/dl and standard deviation  $SD_{(20-40)} = 26.53$  mg/dl; in group (40–60) years (N = 39) LDL cholesterol is  $M_{(40-60)} = 126.05$  mg/dl and the standard deviation  $SD_{(40-60)} = 25.34$  mg/dl; in the group (60–...) years (N = 30) LDL cholesterol is  $M_{(60-..)} = 131.80$  mg/dl and the standard deviation

 $SD_{(60-...)} = 27.64 \text{ mg/dl}$ . The mean values of LDL cholesterol measured at the three age groups differ significantly for at least two of the age groups considered (F = 4.88;  $p = 0.009 < \alpha = 0.05$ ) (Fig. 4b).

Triglycerides in adipose tissue and other tissues are the most important deposit of the body energy reserves. In adipose tissue are stored as glycerol, fatty acids and monoglycerides, which are converted in the liver into triglycerides that enters the constitution VLDL (80%) and LDL (15%) (Ref. 19).

Average value of triglycerides – males. For the group (20–40) years (N = 19) the average value of triglycerides is  $M_{(20-40)} = 117.78 \text{ mg/dl}$  and the standard deviation  $SD_{(20-40)} = 43.24 \text{ mg/dl}$ ; in the group of (40–60) years (N = 40) the average value of triglycerides is  $M_{(40-60)} = 141.40 \text{ mg/dl}$  and the standard deviation  $SD_{(40-60)} = 58.15 \text{ mg/dl}$ ; in the group (60–...) years (N = 38) the average value of triglycerides is  $M_{(60-...)} = 107.92 \text{ mg/dl}$  and the standard deviation  $SD_{(60-...)} = 44.78 \text{ mg/dl}$ . Mean values of triglycerides measured at three age groups differ significantly for at least two of the age groups considered (F = 4.441;  $p = 0.014 < \alpha = 0.05$ ) (Fig. 5).

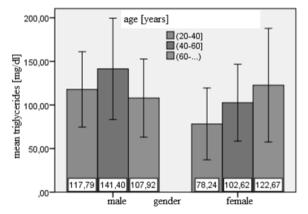


Fig. 5. Average value of the triglycerides in male and female by age groups

Average value of triglycerides – females. For the group (20–40) years (N = 34) the average value of triglycerides is  $M_{(20-40)} = 78.23$  mg/dl and standard deviation  $SD_{(20-40)} = 41.19$  mg/dl; in the group (40–60) years (N = 39) the average value of triglycerides is  $M_{(40-60)} = 102.61$  mg/dl and the standard deviation  $SD_{(40-60)} = 44.11$  mg/dl; in the group (60–...) years (N = 30) the average value of triglycerides is  $M_{(60-...)} = 122.66$  mg/dl and the standard deviation  $SD_{(60-...)} = 65.13$  mg/dl. Mean values of triglycerides measured at three age groups differ significantly for at least two of the age groups considered (F = 6.279;  $p = 0.003 < \alpha = 0.05$ ) (Fig. 5).

ALT is found primarily in the liver (the liver cells being located mainly in the cytosol) and in descending order of concentration in the kidneys, myocardium, skeletal muscle and pancreas. ALT is an indicator of cytolysis most frequently

explored in the opinion of the majority of the best placed to detect even minimal liver damage. ALT is more specific to liver disease than AST (Ref. 20).

Average value of AST – males. For the group (20–40) years (N=19) the AST average value is  $M_{(20-40)}$  =27.68 IU/l and standard deviation SD<sub>(20-40)</sub> = 8.13 IU/l; the group (40–60) years (N = 40) AST average value is  $M_{(40-60)}$  = 27.12 IU/l and standard deviation SD<sub>(40-60)</sub> = 7.99 IU/l; the group (60–...) years (N = 38) AST average value is  $M_{(60-...)}$  = 28.86 IU/l and standard deviation SD<sub>(60-...)</sub> = 7.36 IU/l. Between AST mean values measured at the three age groups there were no significant differences (F = 0.500; p = 0.608 >  $\alpha$  = 0.05) (Fig. 6*a*).

Average value of AST – females. For the group (20–40) years (N = 34) the AST average value is  $M_{(20-40)} = 25.08$  IU/l and standard deviation  $SD_{(20-40)} = 7.15$  IU/l; the group (40–60) years (N = 39) AST average value is  $M_{(40-60)} = 23.53$  IU/l and standard deviation  $SD_{(40-60)} = 7.60$  IU/l; the group (60–...) years (N = 30) AST average value is  $M_{(60-...)} = 24.50$  IU/l and standard deviation  $SD_{(60-...)} = 5.98$  IU/l. Between AST mean values measured at three age groups there were no significant differences (F = 0.452;  $p = 0.636 > \alpha = 0.05$ ) (Fig. 6*a*).

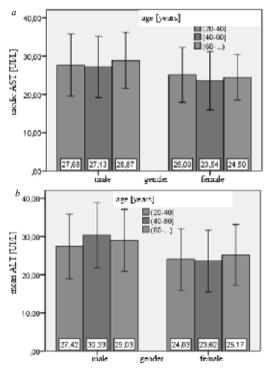


Fig. 6. Average value of AST (a) and ALT (b) in males and females by age groups

Average value of ALT – males. For the group (20–40) years (N = 19), the ALT average value is  $M_{(20-40)} = 27.42$  IU/l and standard deviation SD<sub>(20-40)</sub> = 8.42 IU/l; the group (40–60) years (N = 40) ALT average value is  $M_{(40-60)} = 30.32$  IU/l and standard deviation SD<sub>(40-60)</sub> = 8.55 IU/l; the group (60–...) years (N = 38) ALT average value is  $M_{(60-...)} = 29.02$  IU/l and standard deviation SD<sub>(60-...)</sub> = 8.13 IU/l. Between ALT average values measured at the three age groups there were no significant differences (F = 0.797;  $p = 0.454 > \alpha = 0.05$ ) (Fig. 6b).

Average value of ALT – females. For the group (20–40) years (N = 34), the ALT average value is  $M_{(20-40)} = 24.02$  IU/l and standard deviation  $SD_{(20-40)} = 8.04$  IU/l; the group (40–60) years (N = 39) ALT average value is  $M_{(40-60)} = 23.61$  IU/l and standard deviation  $SD_{(40-60)} = 8.08$  IU/l; the group (60–...) years (N = 30) ALT average value is  $M_{(60-...)} = 25.16$  IU/l and standard deviation  $SD_{(60-...)} = 7.97$  IU/l. Between ALT average values measured at three age groups there were no significant differences (F = 0.328;  $p = 0.721 > \alpha = 0.05$ ) (Fig. 6b).

#### CONCLUSIONS

Summing-up, in male patients, the average values of total lipids measured at three age groups showed no significant differences (F = 2.02;  $p = 0.138 > \alpha = 0.05$ ), while in women, the levels measured differed significantly for at least two of the age groups considered (F = 9.05;  $p < 0.001 < \alpha = 0.05$ ). The mean total choiesterol measured at the three age groups of men showed no significant differences  $(F = 0.192; p = 0.826 > \alpha = 0.05)$ . In women, the mean values of total cholesterol measured at the three age groups differ significantly for at least two of the age groups considered (F = 6.66;  $p = 0.002 < \alpha = 0.05$ ). In men, the mean values of HDL cholesterol measured at the three age groups showed no significant differences (F = 0.672;  $p = 0.513 > \alpha = 0.05$ ), while in females, the mean values of HDL cholesterol measured at three age groups differ significantly for at least two of the age groups considered (F = 4.55;  $p = 0.013 < \alpha = 0.05$ ). The mean values of LDL cholesterol measured at three age groups of men showed no significant differences  $(F = 0.07; p = 0.933 > \alpha = 0.05)$ . In women, the mean values measured at the three age groups differ significantly for at least two of the age groups considered (F =4.88;  $p = 0.009 < \alpha = 0.05$ ). Mean values of triglycerides measured at three age groups of differ significantly for at least two of the age groups considered both in men (F = 4.441;  $p = 0.014 < \alpha = 0.05$ ) and in women (F = 6.279; p = 0.003 < 0.003 $\alpha = 0.05$ ). Between AST mean values measured at the three age groups there were no significant differences both in men (F = 0.500;  $p = 0.608 > \alpha = 0.05$ ) and in women (F = 0.452;  $p = 0.636 > \alpha = 0.05$ ). Similarly, between ALT average values measured at three age groups no significant differences were identified, both in men  $(F = 0.797; p = 0.454 > \alpha = 0.05)$  and in women  $(F = 0.328; p = 0.721 > \alpha = 0.05)$ .

Due to the geographical area they live in, the inhabitants of Sulina, Danube Delta, are isolated and are constrained to eat more fish than other types of meat, Sulina being surrounded by water. People are more active than those from inland, they move around only with boats and they maximise the energy provided by total lipids, the final result being an overall good health.

From this statistical analysis, it results that fish and fish products really helps us maintain the normal level of total lipids and the liver metabolism, if, we include in the diet these products more frequently than normal. The results of this study confirm that hepatic evaluation and liver profile parameters of the screened patients show better values as compared to non-fish eating people. Consequently, including fish in one's diet can significantly improve the overall health state.

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# STUDY REGARDING THE QUALITY OF HAEMODIALYSIS WATER

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**Abstract**. The approached topic focuses on the importance of water treatment, water testing and the implications to the patient of water and dialysate quality. The quality of the dialysis fluids depends on two components: water and the concentrate (and the way they are prepared). Because the dialysate is composed mainly of water, it is very important for monitoring the chemical and microbiological purity of dialysis water. Dialysis water treatment implies water treatment system and a hydraulic circuit for the distribution of the purified water. Water system design and water distribution system should minimise opportunities for microbial growth and chemical contamination. The main objective of this paper was to evaluate an effectiveness of dialysis water treatment system. Investigations were carried out during the four year of exploitation since January 2008 to December 2011, in the dialysis centre Renamed Nefrodial Oradea. The water is produced on site by system: sand filter, carbon filter, filter for manganese and lead removal, softeners and reverse osmosis units. For the analysis of conductivity, free chlorine, chloramines, cooper, aluminium, lead, water samples were taken in accordance with the national norms. The dialysis water treatment from Renamed Nefrodial Clinic ensures the reject of the majority of contaminants: up to 95–98% of dissolved salts and up to 99% of bacteria, endotoxins and other chemical contaminants.

Keywords: dialysis water, water treatment, chemical contaminants, microbiological contaminants.

## AIMS AND BACKGROUND

The study focuses on monitoring the main parameters of treated water in dialysis clinic Renamed Nefrodial Oradea, during the validation and monitoring, with particular emphasis on: conductivity, free chlorine/chloramines, cooper, aluminium and lead, bacteria and endotoxins. The focus for reporting against these parameters was done for the following reasons: the patients on hemodialysis may show signs and symptoms caused by water contamination, which can lead to patient injury or death<sup>1</sup>. Some of the significant signs and symptoms generated by water contamination with parameters analysed in this study are listed in Table 1 (Refs 2 and 3).

<sup>\*</sup> For correspondence.

Chemical contaminants	Symptoms
1 chlorine/chloramines	anemia, hemolysis, blindness that inhibit the antioxidant ways, oxidise hemoglobin causing methemoglobinemia
2 aluminium	anemia, neurological disorders
3 cooper	anemia, hemolysis
4 lead	hypertension
Microbiological contaminants	
1 bacteria and endotoxins	hypotension, nausea and vomiting, death

Table 1. Risks in hemodialysis associated with water<sup>2,3</sup>

In dialysis are used significant amounts of water for the production of dialysis fluid. A dialysis patient who is treated 3 times a week with approximately 150 l of dialysis fluid each time is exposed to 23 400 l per year. This underlines the importance of water purification before its use in dialysis and the need for continuous and rigorous monitoring of the quality of water used for dialysis<sup>4</sup>. Water plays an important role in preparing the dialysate, so it is necessary to have the water whose quality from a chemical and microbiological point of view to ensure the protection of patient with haemodialysis<sup>5</sup>. Consequently, inside the clinic of haemodialysis, choosing a suitable system for the treatment of the dialysis water is crucial. However, it would be a big mistake to believe that making an optimum choice for the treatment system means that all issues relating to water quality have been resolved because of major importance should also be the maintenance and monitoring method of the system<sup>6</sup>.

The components of the treatment system are determined by the quality of the water supply and as a whole, its capacity to produce and maintain its proper quality. The failure to produce the adequate quality water may have serious consequences for the patients safety.

*Characteristics of water treatment at Renamed Nefrodial Oradea*. Water for haemodialysis requires additional treatment to remove contaminants that may be present in drinking water and is purified using operations that may include ion exchange, reverse osmosis, filtration or other suitable procedures. The typical water treatment system for dialysis will depend upon the quality of the incoming supply. The water treatment system adopted at Renamed Nefrodial Oradea had in view the following equipment: water supply storage tank; mechanical filter having the function of retaining the suspended solids; active carbon filter used to reduce colour, odour, chlorine and chloramine; filter having the role to removal iron and manganese ions; filter for adjusting acidity; softener to remove calcium and magnesium ions; reverse osmosis unit; hydraulic closed circuit without dead space.

Figure 1 presents a panoramic view of the water treatment station, the clinic SC Renamed Nefrodial SRL.

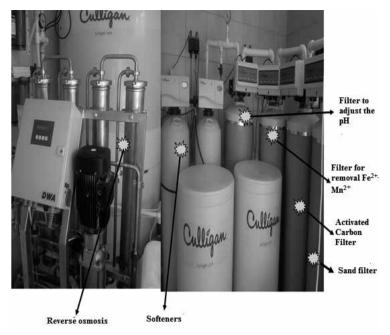


Fig. 1. Panoramic view of the water treatment station, the clinic SC Renamed Nefrodial SRL

*Water treatment monitoring and maintenance.* Water treatment quality control covers two aspects: one is the microbiological purity and the other is the chemical purity.

– Microbiological quality of water. The main microbiological contaminants of fluids used in dialysis are the bacteria that are naturally present in the water. These include gram-negative bacteria and mycobacteria, which can survive and multiply rapidly in water containing even small quantities of organic substances. The microbiological monitoring of water quality is mandatory, and the results should be used to determine the corrective strategies. The microbiological determinations provided in assessing the microbiological water quality of dialysis are the total viable count (TVC), endotoxins, respectively<sup>7</sup>.

- Chemical water quality. In the clinic was established as a water quality system to provide the monitoring of chemical parameters specified by the legislation in this domain and the frequency of their determination<sup>8</sup> (Table 2).

Contaminant	Monitoring		
Aluminium	monthly		
Copper	monthly		
Lead	monthly		

Table 2. Frequency of tracking the chemical quality of water<sup>8</sup>

# EXPERIMENTAL

In the dialysis clinics the focus is on establishing the critical points regarding the patients exposure to potential risks caused by the dialysis fluids quality. In this study, water quality analysis only covered the effluent of the treatment system in order to underline the importance of choosing the correct and appropriate method. The study was conducted to evaluate the microbiological and chemical quality of water which is a key factor in the exchange of fluids for patients with dialysis. The frequency of water and dialysate sampling are an essential part of water quality and dialysate monitoring. Sampling in order to determine these parameters was performed according to the national and international legal recommendations (Table 3). Such supervision is the only way to have a representative picture of water purity, the main component of dialysate.

Table 5. I requeite y of water testing for diarysis	<b>,</b>
Water analysis	Frequency
Bacteria	during the entire period of operation, monthly
Endotoxins	during the entire period of operation, monthly
Free chlorine/total chlorine/chloramines	during the entire period of operation, daily
Conductivity	during the entire period of operation, daily
Chemical contaminants: cooper, lead, aluminium	during the entire period of operation, monthly

Table 3. Frequency of water testing for dialysis9

The ports of water sampling were placed in crucial points of the production chain. Because in this study the focus is on the importance of choosing the water treatment system in a dialysis clinic reported to the water quality required for the dialysis fluid, the harvest ports are located in key points of the treatment plant, without having to refer to the quality of the fluid in the treatment wards.

The microbiological analysis provided two types of measurements: total viable count and endotoxins. For microbiological analysis, at harvest time, the sampling ports of the samples were disinfected. The water was collected in sterile glass containers after running for 10 min. The time for samples transportation and storage was as short as possible (less than 30 min)<sup>10</sup>. Determining the total number of germs was done at a temperature of 22°C, according to the national standard (the filtering system trough the membrane using as medium the nutrient agar culture in an aerobic environment)<sup>11,12</sup>. The endotoxins analysis was done with Limulus Amaebocyte Lysate (LAL) test with a sensitive assay and a detection limit of 0.25 EU/ml.

The basic requirements for an adequate control of the chemical purity of the produced water are summarised in this section. During the study the samples were taken for analysis of the following parameters: daily, tests to determine free chlorine, total chlorine and chloramine. The samples were taken from the port located after the active carbon filter, but after the water system has been in operation for at least 15–20 min. Removing them from the water subjected to the process of reverse osmosis insures a protection of the membranes. Analysis of free chlorine, total chlorine, respectively was done by colorimetric method with a kit Visocolor HE and chloramine was determined by the difference of the values of two parameters. The efficiency of the reverse osmosis process was monitored daily by measuring the conductivity of the influent and effluent the water RO system. This was calculated using the formula:

$$\mu (\%) = \frac{\text{conductivity influent RO}}{\text{conductivity effluent RO}} \times 100$$

Monthly the water samples were taken from the effluent water treatment system for the determination of aluminum, lead and copper. The metal determinations were performed by atomic absorption spectrometry method of atomisation in the oven (Pb), respectively in the flame (Al, Cu) with atomic absorption spectrophotometer AA-6300 Shimadzu<sup>13–15</sup>. The detection limits of the analytical method by AAS are the following: aluminium: 0.01 mg/l, cooper: 0.004 mg/l, lead: 0.0001 mg/l.

#### **RESULTS AND DISCUSSION**

The study was conducted over a period of four years (2008–2011) in the dialysis clinic Renamed Nefrodial Oradea.

*Efficiency of the reverse osmosis process.* The proper functioning of the reverse osmosis process was monitored by determining its efficiency/productivity. The monthly average values calculated during the study are shown in Fig. 2:

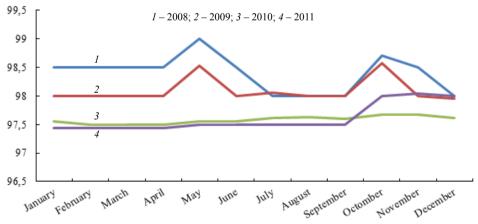


Fig. 2. Efficiency of the reverse osmosis process

The highest value of permeate conductivity was 4  $\mu$ S/cm and the minimum one was of 2  $\mu$ S/cm. As seen in the graphic representation, there is a slight decrease in the efficiency of the reverse osmosis process, together with increasing the duration of the membranes functioning, but even so, the decrease is not significant enough to influence the permeate quality.

*Free chlorine/total chlorine/chloramines*. One of the most critical issues regarding patients safety is the presence of chlorine and/or chloramine. They are added to municipal water systems to remove bacteria, but also effect the destruction of red blood cells. Inside the Renamed Nefrodial Clinic the removal of organic substances, of chlorine/chloramine from water was done through adsorption into columns with active carbon.

The values for free chlorine, total chlorine or chloramine were framed according to legal recommendation (total chlorine: < 0.5 mg/l and chloramine: 0.1 mg/l), without noticing malfunctions in operation of the active carbon filter, during the four years of monitoring.

*Aluminium – lead – copper*. RO will generally remove any molecular compounds smaller in size than water molecules. The metal ions studied are removed by the water treatment system. The results obtained are shown in Tables 4–6.

Month/year		Aluminiu	Maximum allowed		
	2008	2009	2010	2011	concentration (mg/l)
January	0.0017	< 0.0001	< 0.0001	< 0.0001	0.01
February	< 0.0001	< 0.0001	< 0.0001	< 0.0001	
March	< 0.0001	< 0.0001	< 0.0001	< 0.0001	
April	< 0.0001	< 0.0001	< 0.0001	< 0.0001	
May	< 0.0001	< 0.0001	< 0.0001	< 0.0001	
June	< 0.0001	< 0.0001	< 0.0001	< 0.0001	
July	< 0.0001	< 0.0001	< 0.0001	< 0.0001	
August	< 0.0001	< 0.0001	< 0.0001	< 0.0001	
September	< 0.0001	< 0.0001	< 0.0001	< 0.0001	
October	< 0.0001	< 0.0001	< 0.0001	< 0.0001	
November	< 0.0001	< 0.0001	< 0.0001	< 0.0001	
December	< 0.0001	< 0.0001	< 0.0001	< 0.0001	

**Table 4**. Monitoring of aluminium

Aluminum in the dialysate appears to be the major source of the metal in patients with chronic renal failure who develop aluminum toxicity. The aluminum content of the dialysate depends primarily on the content of the water with which it is prepared; as seen in Table 4, test results of water samples for the entire period of analysis have been fairly constant in all four years of functioning of the treatment station below the values recommended by specific national and international legislation. The lead being one of the elements responsible for the Balkan endemic nephropathy (our country being one of the countries with prevalence in the incidence of the Balkan endemic nephropathy, found especially in the south – west of the country), it determined once more its monitoring.

Month/year		Lead	Maximum allowed		
	2008	2009	2010	2011	concentration (mg/l)
January	0.0006	0.0005	0.0021	0.0005	0.005
February	0.0022	0.0025	0.0032	0.0022	
March	0.0032	0.0012	0.0020	0.0004	
April	0.0005	0.0005	0.0010	0.0005	
May	0.0006	0.0005	0.0030	0.0006	
June	0.0022	0.0012	0.0010	0.0028	
July	0.0032	0.0040	0.0032	0.0030	
August	0.0005	0.0003	0.0004	0.0005	
September	0.0006	0.0003	0.0002	0.0003	
October	0.0022	0.0020	0.0032	0.0025	
November	0.0032	0.0022	0.0020	0.0030	
December	0.0005	0.0003	0.0005	0.0002	

 Table 5. Monitoring of lead

Month/year		Cooper	Maximum allowed		
	2008	2009	2010	2011	concentration (mg/l)
January	0.001	0.001	0.001	0.002	0.1
February	0.0032	0.002	0.002	0.004	
March	0.002	0.002	0.002	0.0054	
April	0.002	0.001	0.002	0.02	
May	0.0031	0.003	0.0031	0.005	
June	0.001	0.001	0.001	0.0046	
July	0.001	0.0021	0.001	0.005	
August	0.001	0.0015	0.003	0.005	
September	0.0019	0.0010	0.002	0.005	
October	0.0024	0.003	0.004	0.0048	
November	0.001	0.0024	0.003	0.005	
December	0.001	0.003	0.0045	0.0052	

The efficiency of the reverse osmosis unit, an essential component in treatment of used water in hemodialysis, allowed reducing the concentration of aluminum, lead and copper up to more than 97% over the whole period of operation reducing these parameters below the maximum values permitted by legislation specific to this area .

*Microbiological analysis*. In the analysis performed it was found the absence of the mesophilic aerobic bacteria and endotoxins in water samples monitored during the first four years of study (the national recommendations establish limits: bacterial growth < 100 CFU/ml and endotoxins < 0.25 EU/ml).

From the experimental results obtained, the functioning of the treatment plant during, the choice of system components for water treatment, allowed us to obtain the water whose quality enables the use in dialysis centers so as to ensure the patient protection. No major differences were observed between the values obtained for the total number of germs and endotoxins in the years under review.

#### CONCLUSIONS

This article may assist in learning more about the importance of appropriate water treatment, water testing and the implications to the patient of water and dialysate quality. In the four years of operation, the treatment performance was feasible for most of the analysed pollutants. The dialysis water treatment from Renamed Nefrodial Clinic ensures the removal of the majority of contaminants: up to 95–98% of dissolved salts and up to 99% of bacteria, endotoxin and other chemical contaminants. Because during dialysis are consumed large amounts of water and energy it is indicated, as an important aspect of the management system of the water used in hemodialysis which must be taken into account in the future, to adopt an integrative system of reducing the amount of waste water and using renewable energy sources in hemodialysis clinics.

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# CONSIDERATIONS REGARDING THE BIOMARKER VALUE OF CIRCULANT CONCENTRATION OF PON<sub>1</sub> IN PATIENTS WITH HEART FAILURE THROUGH CHRONIC ISCHEMIC CARDIOMYOPATHY

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**Abstract**. The study has a double objective: to determine the clinical significance as a possible biomarker for risk of heart failure of one of the paraoxonase family members,  $PON_1$  and highlight a possible correlation between circulating levels of isozyme with the value of ejection fraction. As etiology of heart failure, there were selected for the study only the cases suffering from chronic ischemic heart disease without myocardial infarction history. This was due to the fact that myocardial hypoxia is widely recognised scientifically as the main source of production of oxidizing species, among different causes generating myocardial contractile deficit.

*Keywords*: congestive heart failure (CHF), reactive species of oxygen/nitrogen (ROS/RNS), paraoxonase, risk biomarker, ejection fraction.

# AIMS AND BACKGROUND

Paraoxonase (PON) was defined the property of being a plasmatic marker of the antioxidant capacity 70 years later since its discovery in 1949 by Abraham Mazur<sup>1–7</sup>. The three members of the family whose PON synthesis is regulated by the genes located on chromosome 7, develop lactonase type action, phosphodiesterase and/ or arylesterase, actions still insufficiently studied<sup>8–12</sup>. The similarity of processes occurring in the vascular wall in atherosclerosis with those taking place in the ischemic heart, in which reactive oxygen species (ROS) and/ or nitrogen (RNS) play an important role in the initiation and development of the processes of inflammation and structural remodelling of the myocardium has constantly oriented studies and research to the CHF. Their purpose is to identify serum parameters (biomarkers) or paraclinical markers enabling the assessment of risk and therapeutic benefit<sup>12</sup>.

Given the well-known fact that the environment with all its components: physical, chemical and psychosocial to directly influences on human health, we

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can deduce that the internal environment homeostasis is the expression of the living conditions or the environment. From the group of 208 patients, 139 (67%) were rural and 69 (33%) were from urban areas. It was observed that patients living in urban areas have events more severe disease (angina, exertional dyspnea, tachypnea, fatigue) than those living in rural areas as pollution and stress of urban leave a mark on patient quality of life and default on their health<sup>13–15</sup>.

### EXPERIMENTAL

The study group included 208 patients suffering from CHF, etiopathogenic induced myocardial ischemia. The group consisted of 58 women and 150 men (Figs 1*a,b*, respectively). Knowing the medical literature and the fact the serum PON<sub>1</sub> activity is reduced by obesity, smoking, we introduced the requirement to exclude all patients who smoked in the year previous to the study<sup>16</sup>. In terms of body weight, normal weight patients were admitted to having no more than +/– 10% of ideal weight. Furthermore, we did not includ the patients who, in the last 6 months, had followed statin therapy, in order not to reduce the influence of exercise on serum PON<sub>1</sub> concentration, the patients were hospitalised.

(1) Knowing that ageing causes a reduction in serum paraoxonase activity, we have adopted as a minimum condition the average age of the lot to maximise the influence exerted on the minimum values of isozyme. The average age of the cases was 54, 55, being of 53, age of 86 in women and 55 in men (Figs 1a,b, respectively).

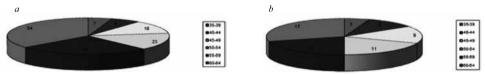


Fig. 1. Distribution of the study cases in relation to gender and age group of patients: distribution number of men -a, and distribution number of women -b

(2) Classes of contractile functional cardiac decompensation were those of NYHA classification, the batch including pending cases at least in Class II of myocardial contractile deficit (Table 1).

Functional		Distribution in the batch according to sex									
class		men			women						
	No	%	of	No	%	of	No	%			
	abs.	entire	only of	abs.	entire	only of	abs.				
		batch	men		batch	women					
		(n = 208)			( <i>n</i> = 208)						
II	63	30.30	35.33	17	8.17	29.3	80	38.46			
III	71	34.13	47.33	29	13.90	50	100	48.08			
IV	16	7.70	10.60	12	5.77	20.69	28	13.4			
Total	150	72.21	100.00	58	27.84	100	208	100			

 Table 1. Distribution of patients in the batch according to sex and functional class of the heart pumping function deficit, assessed based on NYHA classification criteria

(3) Considering as normal value/preserved ejection fraction (EF) the amount recommended (> 50% of the left ventricular telediastolic blood volume), based on distribution of the cases according to NYHA classification criteria, the group was divided into three subgroups: preserved systolic flow (subgroup I) and with low one (Table 2).

				, 0			5			
Func-	Total 1	number of	Distrib	oution with	in the lot dep	ending o	on the FE o	letermined		
tional	patient	s in the lot			val	ue	le			
call	No	%	FE > 50 (sublot I)				FE< 50% (sublot II)			
	abs.		No	9/	6 of	No	%	of		
			abs.	sublot	entire lot	abs.	sublot	entire lot		
II	80	38.46	63	69.23	30.29	17	14.53	8.17		
III	100	48.08	21	23.08	10.10	79	67.52	37.98		
IV	28	13.46	7	7.69	3.36	21	17.95	10.10		
Total	208	100	91	100	43.75	117	100	56.25		

Table 2. Distribution of cases in the lot, according to the determined values of the ejection fraction

In this study, we tested serum  $PON_1$  esterase activity using the substrate ethyl phenyl acetate. Determining the circulating blood serum of  $PON_1$  was done in the blood sampled in the morning, after fasting, twice within 14-day period which was the length of hospitalisation. For each set of measurements, when admitting the patient in hospital, respectively on the 14th day, subsequently the medium concentration was calculated, thereby obtaining two parameters, conventionally referred to as: the mean serum concentration of  $PON_1$  when the admission or discharge took place.

Restricting first measurement was done in the first 24 h of hospitalisation and we justify it by the chance that could have been developed by the treatment instituted, to adjust the amounts determined, by its action on the morphological and functional part, yet reversible, the myocardium contractile deficit. Subsequently, using the values of the two mentioned parameters we calculated the third one, the mean serum  $PON_1$  per lot, regardless of the time of sampling, the parameter that we will use in interpreting the study results.

The processing of the significance of serum values  $PON_1$  determined referred to the proper normal average values corresponding to the age of the study group, specified to be within the range of variation of 75–100 mg/ml (Refs 1, 17–19). To assess the risk, the value below the lower limit of normal, I graded it:

(a) > 75 mg/l: risk absent;

(b) values ranging 65 – 74 mg/ml: minimum risk;

(c) circulant levels with values of 50 – 64 mg/ml: high risk;

(d) seric concentrations < 50 mg/ml: very high risk<sup>1,19,20</sup>.

As objectives of the study, we aim at assessing:

- if the variation in serum PON<sub>1</sub> may be considered a biomarker of heart failure, regardless of functional class of contractile deficit (NYHA classification) and regardless of the patients gender;

– possible influence/correlation between serum  $PON_1$  and value of existing systolic volume in patients in group.

# **RESULTS AND DISCUSSION**

#### REGARDING THE SERIC CONCENTRATIONS OF PON<sub>1</sub>

Comparison of serum  $PON_1$  average concentrations of hospitalisation or discharge patients and the average of the entire lot is summarised in Fig. 2.

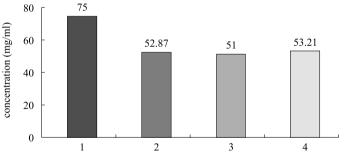


Fig. 2. Comparison of mean serum concentrations of PON at different times of determination on the part of the batch casuistry

1 – normal serum levels of PON (mg/ml); 2 – mean serum concentration PON entire lot (mg/ml); 3 – mean serum concentration PON admission (mg/ml); 4 – mean serum concentration PON discharge (mg/ml)

Reporting increasing of serum PON<sub>1</sub> calculated for all cases, included in the study with normal circulating levels, has shown consistently a reduction of these values:

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(a) for patients in subgroup I: 67%, the mean concentration of serum PON1, in the case of the average concentration of serum  $PON_1$  calculated based on the results of assays performed on admission; 66.6 % the mean concentration of serum  $PON_1$ , in the case of the average concentration of serum  $PON_1$ , based on determinations made at discharge.

(b) for patients included in subgroup II: 64.1%, the mean concentration of serum  $PON_1$ , in the case of average concentration of serum  $PON_1$ , based on determinations made on admission; 63.7% the mean concentration of serum  $PON_1$ , in the case of average concentration of serum  $PON_1$ , based on determinations made on discharge.

#### REGARDING INCIDENCE OF SERIC CONCENTRATION VARIATIONS OF PON<sub>1</sub>

Determination of serum average paraoxonase incidence in the study group, calculated as average of determinations at admission and discharge, allowed us the next 'stratification' according to the areas chosen (Table 3).

Evaluated value of seric	Number of patients							
activity	in the target	no.	% of the	casuistry				
	lot	abs.	with low values	entire lot				
< 50 mg/ml	208 = 100%	139	70.2	66.8				
50–64 mg/ml		38	19.6	18.2				
65–74 mg/ml		17	8.7	8.26				
Total of cases with values < 75 mg/ml		194	100	93.2				
> 75 mg/ml		14	_	6.8				

Table 3. Distribution of average seric concentrations of PON<sub>1</sub> in the lot, regardless of the patients sex

Depending on the sex ratio of the patients in the lot, the incidence of serum PON<sub>1</sub> below the normal average was stratified according to data from Table 4.

In relation to systolic flow value, preserved or reduced, the dispersion of 194 cases of serum PON<sub>1</sub> reduction of the lot is subdivided into 88 cases (42.3%) belonging to the target subgroup I, and the remaining cases = 51% - 106 distributed -target subgroup II (Table 4).

Evaluated value	Number of patients (208/lot)									
	entire t	arget lot		subgroup $(n = 91)$		S	subgroup $(n = 117)$			
	No abs.	%	No abs.	%	<sup>0</sup> ⁄0 <sup>**</sup>	No abs.	%	%**		
< 50 mg/ml	139	66.8	69	33.17	75.8	70	33.6	59.8		
50–64 mg/ml	38	18.2	12	5.77	13.18	26	15.5	22.1		
65–74 mg/ml	17	8.26	7	3.365	7.69	10	4.8	8.54		
Total of cases with < 75 mg/ml	194	93	88	42.3	96.7	106	51.0	91.3		
Normal	14	6.8	3	1.43	3.3	11	5.27	9.4		
Number of patients in lot/subgroup	208	100	91	43.7	100	117	56.2	100		

**Table 4**. Distribution of mean serum  $PON_1$  in the entire group, namely on the constituent subgroups, regardless of sex of the patients

\* percentage of the entire lot; \*\*percentage of the patients in the respective subgroup.

Centralisation of average incidence with serum  $PON_1$  reduced in the study group, compared to the value of ejection fraction and sex (Tables 5*a*,*b*).

Table 5a. Incidence of mean seric concentration values of PON1, in the casuistry consisting of pa-
tients with heart failure, according to gender and systolic flow/distribution of cases with low serum
PON <sub>1</sub> activity in subgroup I, by gender

Assessed value		Number of patients (208/lot)								
	enti	re lot	subgroup I $(n = 91)$		depending on the gender of patients					ients
	No	%	No	%**		males			female	es
	abs.		abs.		No	%**	%*	No	%**	%*
					abs.			abs.		
< 50 mg/ml	139	66.8	69	75.8	40	44.0	19.2	29	31.7	13.9
50–64 mg/ml	38	18.2	12	13.1	9	9.9	4.32	3	3.3	1.44
65–74 mg/ml	17	8.26	7	7.8	5	5.49	2.40	2	2.2	0.9
Total cases with < 75 mg/ml	194	93	88	96.7	54	59.3	25.9	34	37.2	16.3
Normal	14	6.8	3	3.2	2	2.2	1.04	1	1.1	0.5
Number of patients in lot/ subgroup	208	100	91	100	56	61.5	26.9	35	38.4	16.82

\* percentage of the entire lot (n = 208); \*\* percentage of the patients in I subgroup.

	I											
Assessed value	Number of patients (208/lot)											
	entire lot		subgroup II		depending on the gender of patients							
		C I				males			females			
	No abs.	%	No abs.	0⁄0**	No abs.	%	%**	No abs.	%	%**		
< 50 mg/ml	139	66.8	70	59.2	62	29.8	53	8	3.84	6.75		
50–64 mg/ml	38	18.2	26	23.2	18	8.65	15.4	8	3.84	6.75		
65–74 mg/ml	17	8.26	10	8	7	3.36	5.9	3	1.4	2.57		
Total cases with <75mg/ml	194	93	106	90.5	87	83.1	74.3	19	9.1	16.2		
Normal	14	6.8	11	9.4	7	3.36	5.9	4	3.36	3.41		
Number of patients in lot	208	100	117	100	94	45.2	80.3	23	11.2	19.6		

**Table 5***b*. Incidence of mean seric concentration values of PON1, in the casuistry consisting of patients with heart failure, according to gender and systolic flow/numerical distribution in subgroup II of the cases according to the mean serum concentration of PON,

\* percentage of the entire lot (n = 208); \*\* percentage of the patients in II subgroup.

In appreciation of the lot scatter determinations we have not resorted to calculating the dispersion, but to the standard deviation from the values of the average concentration of serum PON<sub>1</sub>, which is 52,  $87 \pm 19.19$  mg/ml. The value of this indicator in patients with heart failure, patients regardless of gender, had an incidence of normality almost of 7%, the difference – 93% representing the number of cases with small values of seric concentration of the isoenzyme. The share with which circulating level of PON<sub>1</sub> was reduced was  $22.13 \pm 0.3$  to mg/ml, representing 29.5 percent of the lower limit of the normal value (Fig. 3).

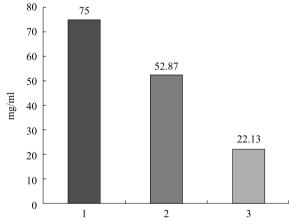


Fig. 3. Highlighting the share of value reduction of serum PON<sub>1</sub> activity in the study group compared to normal

1 – the lower limit of the average normal serum concentration (mg/ml); 2 – mean serum concentration value of PON-1 averaged over the entire lot (mg/ml); 3 – share value reduction (mg/ml)

To interpret the meaning of these latter determined values, we admit that as inotropic function of the heart diminishes, there is an overload of  $PON_1$  functional tissue that interferes reactively-adaptively, being activated by oxidative stress increasing quasipermanently.

In relation to the fields of reduction of serum PON<sub>1</sub> chosen, significant is the incidence on the enzyme activity placed under 50 mg/ml: 139 of 194 patients with plasmatic enzyme activity below normal. We believe that this incidence gives the isozyme a great specificity as a marker of risk in patients with CHF occurred through ischemic cardiomyopathy. In support of this hypothesis come also the data resulting from comparing the incidence of serum PON<sub>1</sub> averages in the studied group in relation to functional class (NYHA classification), regardless of the patients gender. The 'behaviour' of PON<sub>1</sub> serum confirms the existence of a correlation of the level of serum enzyme activity reduction with the functional deficit class, both as incidence and as values. This can be considered as an argument that allows to frame the circulant determination of the isozyme between specific risk markers in chronic heart failure due to ischemic heart disease.

Interpreting the distribution of mean serum reduced  $PON_1$  in the gender lot relative to the patients gender, there is a predominance of frequence among men (69.7% versus 23.6% for women). This implies that out of four patients afflicted by CHF, with reduced serum activity of paraoxonase, with a very high probability, three patients are men and only one will be a woman (Fig. 3).

Taking into account the 194 cases of CHF with reduction of serum  $PON_1$ , in relation to the value of existing ejection fraction existing in patients constituting the lot, the distribution is the one in Fig. 4.

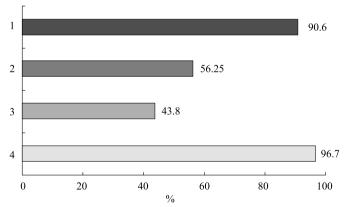


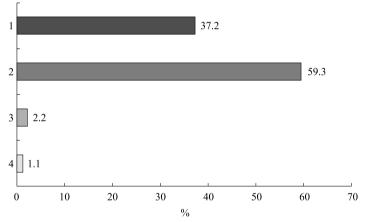
Fig. 4. Incidence of average reduced seric  $PON_1$  values in the batch depending on the ejection fraction of patients

1 – number of cases with heart insufficiency and preserved systolic flow associating the reduction of mean seric concentration of PON<sub>1</sub>/no of cases in subgroup I = 88/91 = 96.7%; 2 – number of cases with heart insufficiency and preserved systolic flow in subgroup I/no of cases in the entire lot = 91/208 = 43.8%; 3 – number of cases with heart insufficiency with systolic flow reduction in II subgroup/no of cases in the entire lot = 117/208 = 56.25%; 4 – number of cases with heart insufficiency with systolic flow reduction as well as of mean seric concentration of PON<sub>1</sub>/no of cases in II subgroup = 106/117 = 90.6%

Out of the 194 cases constituting the entire batch, that had reduced serum for PON<sub>1</sub> (93%) it is mentioned that 17 cases (8.26% of all cases), fall in the minimal risk category, while the remaining cases – 177, the equivalent of 85.1% – have average seric concentrations of PON<sub>1</sub> below 65 mg/ml, i.e. real risk revealing values (Table 3).

Research of the average values of serum  $PON_1$  incidence, placed below the normal, relative to the value of ejection fraction existing in patients batch, regardless of gender showed a growth of over 9%, in the cases with decreased systolic volume, compared to that in which it was preserved (42.3% in subgroup I, 51% in the subgroup of those with heart failure and reduced systolic flow) (Table 4). Particular is the fact that reducing serum  $PON_1$  in patients with reduced systolic flow was present in all cases, both in determinations at hospital admission and in those of discharge. The intensity/degree of reduction of serum  $PON_1$  is clearly hightlighted in patients with decreased systolic flow, predominating values below 65 mg/ml (96 cases out of 117, the equivalent to a rate of 82 out of a total of 106 cases with reduced enzyme values). The statistical data allow us to claim that reducing serum  $PON_1$  consists in laboratory test that is specific to heart failure, as a general disease, with a large specificity for reduced systolic flow.

The research of average values of  $PON_1$  incidence below the normal, within the subgroup formed by 91 patients with preserved ejection fraction shows that reducing the enzyme activity is placed percentage of 96.5 (Fig. 5).



**Fig. 5**. Distribution in relation to the gender of patients suffering from heart failure with preserved systolic flow of cases based on the value of the average concentration of serum PON<sub>1</sub> 1 - number of male cases with low average concentration of serum PON-1; 2 - number of female cases with low average concentration of serum PON-1; 3 - number cases of male constituent lot; 4 - number cases of female constituent lot

Hypothetically, we postulate that in cases of heart failure with preserved systolic flow reduction of seric concentrations of paraoxonase-1 occurs in 19 cases out of twenty, which gives the determinations of this kind of disease the specificity of a biomarker. It is also significant that in this subset, 3 out of 4 patients show extremely important reduction in serum enzyme activity < 50 mg/ml: 75.8%.

In subgroup I, the incidence of reduction of mean seric  $PON_1$  concentration by patients' gender shows that six patients (59.3%) out of ten male patients suffering from ischemic heart failure occurred by ischemic cardiomyopathy, with preserved systolic volume presents values of average seric  $PON_1$  concentration relevant of evolutionary existence of a risk. On the other hand, in the situation of women suffering from the same disease maximum four in ten develop low circulating levels for  $PON_1$ . Based on this statistical argument, we state that there is a better expression of risk in heart failure with preserved systolic flow in men, if one takes into account the amount of serum  $PON_1$  average. In this regard, it is worth noting that the ratio of the sexes significance of serum reduced  $PON_1$  is almost twice (Table 5*a*).

In relation to the patients gender constituting subgroup II, the incidence of serum paraoxonase reduction is shown in Fig. 6.

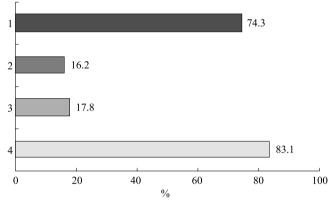


Fig. 6. Incidence of serum paraoxanase reduction

1 – no of male cases with seric reduction of PON<sub>1</sub>, in subgroup II/no. of cases with reduction of PON-1 in subgroup II = 87/106 = 83.1%; 2 – no of female cases with seric reduction of PON1, in subgroup II/no of cases with reduction of PON-1 in subgroup II = 19/106 = 17.8%; 3 – no of female cases with seric reduction of PON/no of cases in subgroup II = 19/117 = 16.2%; 4 – no of male cases with seric reduction of PON/no of cases in subgroup II = 87/117 = 74.3%

In percentages, the subgroup of patients with heart failure and decreased systolic flow, the number of those with seric  $PON_1$  reduction was 4,5 times more common for men (men/women = 74.3/16.2). This allows us to support that the change in seric concentration in patients with heart failure of ischemic cardiomyopathy cause and with reduced systolic flow is dependent on gender.

#### CONCLUSIONS

The findings regarding the 'behaviour' of the  $PON_1$  marker in the group consisting of patients with heart failure occurred on the background of chronic ischemic heart disease with and without preserving the value EF allow to support the following:

In patients with heart failure, regardless of their gender or ejection fraction value, the mean seric  $PON_1$  concentration based on the dosages at hospital admission and discharge, as a semi-sum of dosages previously mentioned was evaluated at levels consistently falling below lower limit of normal circulating, which constitutes the argument of serum enzyme dosage recommended as useful diagnostic biomarker for heart failure.

In support of such a proposal come also the results on the distribution of serum  $PON_1$  in the batch, regardless of the patients gender who showed an extremely high incidence of cases with reduced amount of serum enzyme activity; this is 93%.

Based on statistical argument, we state that  $PON_1$  mean serum value expresses the true risk of heart failure with preserved systolic flow in men compared to women suffering from the same type of condition.

If patients with heart failure and reduced cardiac serum PON<sub>1</sub> average concentration value determined both at hospital admission and at discharge is less compared to similar values of patients constituting subgroup I, in which the systolic flow was preserved.

Variance analysis of values of serum  $PON_1$  average concentration during the 14 days of hospitalisation, on subbatches, allows to conclude that both in patients with heart failure and preserved systolic flow and for patients suffering from the same disease, having reduced systolic flow, there is not an improvement in the average concentration of  $PON_1$  values, determined at discharge, compared with that given by dosing when being hospitalised.

We interpret the difference of variation in the reduction level of serum  $PON_1$  activity in the two subgroups as expressing the increased degree of myocardial ischemia as a result of overlapping between ischemis to the coronary artery disease and the one occurred by the reduction of the systolic flow and diastolic shortening, as a result of tachycardia. The consequence of their action is an increased production of ROS/RNS by emphasising the poor irrigation of the organ, concomitent with the overloading of its labor. In this way, it is imposed the intensification of ventricular myocardial remodelling, mechanisms which we consider that imply the modulation of paraoxonase.

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# EVALUATION OF DISABLED CHILDREN IN TURKEY WITH THE AIM OF THEIR NEEDS TO GREEN SPACE

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**Abstract**. Even though there is noticeable size of disabled populations in Turkey, there are no comprehensive studies about socioeconomic characteristics, problems, and expectations of those communities. According to the Education Ministry, there are 1 million and 100 thousand disabled children between ages of 4–1; 45 thousand with vision impairment. The main objective of this study is to evaluate the condition of this group of children and evaluation their circumstance. All children, including disabled children, have the right to play, have fun and take part in recreational activities that is important for their development. This article tried to put light on characteristics of disabled children, their needs, and conditions they have in Turkey. With regard to all statistic data and analyses, this paper makes attempt to give a clear image of this amount of population for designers, urban planners, instructors, and other experts who want to accomplish an action for their sake in Turkey. Finally, it emphasises the urgent need of peculiar green areas for those communities, so that they will be able to enjoy and entertain in the free green places as much as other ordinary persons have.

Keywords: disabled children, landscape design, special needs, Turkey.

# AIMS AND BACKGROUND

Collecting statistics about disabled population has been given great importance with the purpose monitoring and evaluation of related policies. Children need a place to play and they also need space with informality, and freedom to move around and make noise. They should express themselves through experiment and investigate the world around. Disabled children need this freedom even more than others. In surrounding which stimulates their imagination and challenge them to face and overcome risks, they will be given opportunities to build their self-confidence and independence<sup>1</sup>. All children, including disabled children, have the right to play in green space, have fun and take part in recreational activities. This is important as play has a very important role in a child development. Research indicates that children in natural settings play and learn with more vigor, engagement, imagination,

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and cooperation than children playing on artificial surfaces. Play in green space can help develop speech, sensory skills, imagination, independence and social skills<sup>2</sup>.

The UN Convention on the Right of the Child states that disabled children have the right to be included in their local community and to do the kind of things that non-disabled children do. They have the right to take part in play and leisure activities, the equal right to access cultural, artistic and leisure opportunities and the right to support to help them do this<sup>1</sup>. Social barriers such as fear, embarrassment or discriminatory attitudes also need to be tackled so that an accessible natural setting is also an inclusive one in which disabled children and their families fell welcome. The purpose of this study is to evaluate conditions of disabled children in Turkey and definition needs of these children, specially needs to going any kind of green area, for example parks, nature region, so on.

According to the UN statistics (2006), more than 500 million people in the whole world have different disabilities. In 1992, Eurostat showed that 11% of the Europe population had different disabilities<sup>3</sup>. It has been reported that 12/29% of the population in Turkey have mental or physical disabilities<sup>4</sup> and out of this percentage, 9 million people are at the age of 1 - 12 who have special needs. According to the Ministry of Education, there are 1 million and 100 thousand disabled children agreed between 4 and 18 and among them 45 thousand ones have vision impairment, 130 thousand ones have hearing impairment, 500 thousand of them have mental and emotional disabilities, and 300 thousand of them suffer from physical disabilities. Enrollment rate in Disabled Children School is around 2%. The number of disabled children who benefit from primary education including special education is 28 thousand (Ref. 5).

This research was exploratory in nature. It attempted to gather data for which there is little or no prior knowledge. There have not been study relating to the benefits of Parks and Recreation programs for children with disabilities and there are not sufficient Parks and Recreation programs currently available for children with disabilities in the Turkey. This paper will review all various literatures on nature for children suffering from disabilities. The purpose of this research is definition of benefits of natural setting for children with disabilities in a clear and compact way with help of recent significant researches on gardens. The finding revealed that there is necessary need to improve and construct well equipped park and recreational setting for children with disabilities.

#### HEALTH OUTCOMES OF THE GREEN SPACE

Why is nature restorative? There are many theories regarding the effects nature has on human beings. In Clare Cooper Marcus book 'Healing Gardens', Rodger Ulrich writes in an article 'Effects of Gardens on Health Outcome: Theory and Research', that people learn to associate restoration with nature settings and as-

sociate stress with urban settings. As an example, people may find vacations in rural settings relaxing because as a society we were 'taught' these landscapes were calming and restorative. Another theory is that the urban or built environment is over stimulating, causing increased stress in the body. Nature is less complex making it more restorative. Evolutionary theories contend that many cultures have the same positive responses to nature. Humans may be genetically inclined to respond to certain landscapes more positively because these environments were favorable for their survival<sup>6</sup>. Nancy Gerlach-Spriggs contends that our 'response to nature is not Gardens have the ability to heal in many ways'. The simple exposure to nature causes heart rate to decrease. Muscle tension and blood pressure also decrease. Rodger Ulrich explains that 'it is justifiable to propose that gardens in healthcare situations are important stress-mitigating resources for patients and staff to the extent that they foster: a sense of control and access to privacy, social support, physical movement and exercise, and access to nature and other positive distractions'<sup>6</sup>. Ulrich explains in another article entitled 'View through a Window May Influence Recovery from Surgery' that patients who were able to view nature had a shorter hospital stay, lower analgesic use and fewer complaints during recovery<sup>7</sup>. Additionally, there are several other advantages found in healing gardens'.

*Social support*. Users are encouraged to meet other people, sharing stories or experience and receiving spiritual support or physical aid that can help to re-build their self-confidence and enhance healing<sup>8</sup>.

*Sense of control*. A sense of control is an important factor affecting a person ability to cope with stressful events or situations, including stress associated with illness and hospitalization<sup>8,9</sup>. Assisting patients to retrieve their capacities will improve the rate of recovery.

*Exercise*. Exercise can decrease stress, blood pressure and depression<sup>10</sup> and strengthen body and mind, especially the functions of heart, lung and nervous system<sup>9</sup>.

*Positive distractions*. Positive distractions are environmental criteria that can attract people attention, thus disturbing negative thoughts or emotions and improving the issues of blood pressure and stress<sup>11,</sup> for instance, music, art, flowers and animals.

*Reduction of mental fatigue.* Experiencing a natural environment has the efficacy of decreasing mental fatigue. People feel relax and recover from attentional fatigue by engaging in natural surroundings and being away from stressors<sup>12</sup>.

#### IMPORTANCE OF PLAY FOR DISABLED CHILDREN

As given in literature, it is necessary that sportive and physical activity fields should be taken out of indoor facilities as much as possible for the adaptation of the individuals with disabilities and their parents<sup>13</sup>. During the first years of life, the

roles of children are characterised by play. They actively explore and interact with people and objects in their world. Mouthing, banging, touching, grasping, holding, rolling over, and clapping are just a few of the actions which furnish information to the child. These interactions provide the child with an understanding of control and causality. Children repeat actions to establish relationships between materials and objects. They then modify their interactions to examine new relationships. During the early years language emerges and play unfolds, from exploration to using objects to pretend and mimic activities in the world; play and playfulness. Although play is difficult to define, it is easier to recognise. Children are playful if they are intrinsically motivated, internally controlled, free to suspend reality, and able to set and maintain a play frame<sup>14</sup>. Play is a critical occupation of childhood. In fact, it is the main occupation of early childhood<sup>15</sup>. Play, often described as the 'work of children', is far more than a job to be completed. Play starts and stops when the player wants it to. Its self-initiated, self-directed quality offers a flexibility not found in work. Unlike working, a player can do what he wants to do, including changing play at any time, restructuring it, choosing a new play partner, or restarting the game<sup>16-18</sup>.

The environment education in Turkey and international models clearly display that environment education in Turkey is not yet on the desired level<sup>19</sup>. They develop language, symbolic thinking, social skills, and motor skills<sup>20</sup>. In addition there is an essence to just playing, with no ulterior motive of gaining and/or refining skills, that is equally important to typical development<sup>21</sup>. Play is the basic nature of young, typical children. Parents of typically developing children are seen as the facilitators of play as they respond to and promote the interactions of their child. They function as play partners, and companions, following the play initiations of the child<sup>22</sup>. Parents of children with disabilities may unwittingly promote play deficits, and feelings of incompetence experienced by their children. Research suggests that mothers of children with disabilities play less with, and are more controlling of their children than are mothers of typically developing children<sup>23,24</sup>. Interactions between mothers and their disabled infants indicate that infants provide fewer cues and initiate interaction less frequently then do their non-disabled peers. Mothers of these infants often have a high level of activity and dominate initiations<sup>23</sup>. This pattern of interaction has been noted to diminish optimum development<sup>24</sup>. Interestingly, this pattern changes over time, as parents of toddlers with disabilities have been noted to withdraw from their child, playing less with them as the child grows older<sup>25</sup>.

# ESSENTIAL DESIGN ELEMENTS AND ENVIRONMENTAL QUALITIES

Design, architecture and engineering professions are directly responsible for the development of our physical environment. Landscape architects make decisions effecting the environment and public health and safety in many aspects of their practices<sup>26</sup>. An emerging area of research and design focus within landscape architecture has sought to address the relationship of designed natural environments to health and healing. As landscape architecture increasingly addresses the interface between designed natural environments and health, confusion has developed regarding various terms applied to this concept. 'Healing gardens' is a term frequently applied to gardens designed to promote recovery from illness. 'Healing', within the context of healthcare, is a broad term, not necessarily referring to the cure from a given illness. Rather, healing is seen as an improvement in overall well-being that incorporates the spiritual as well as the physical. Numerous healthcare institutions within and outside the United States have begun to incorporate therapeutic landscape design. As Clare Cooper-Marcus and Marni Barnes have noted in their book 'Healing gardens focus on providing stress relief, alleviation of physical symptoms, and improvement in the overall sense of wellness for both patients and healthcare staff'.

# CONCLUSIONS

The development of children starts at birth and continues into adulthood. Children need to develop in five crucial areas for proper growth: social/emotional, intellectual, sensory, perceptual-motor, and physical development<sup>27</sup>. All children, regardless of their abilities, pass through the same developmental stages in the same sequence; only the timing and rate vary. The crucial concept to understand is that the similarities between children with disabilities and able-bodied children are far greater than the differences. At the same time, differences in the rate of development may occur in one, or more, developmental areas according to the child disability; social emotional disabilities, perceptual-intellectual disabilities, and physical disabilities<sup>28,29</sup>. These disabilities should not, however, lead to their separation from other children or the underestimation of their needs in public places. The disabled children have greater need for recreation or play services because their limited circumstances to some extent prevent their exploring for opportunities for themselves. Additionally, 'many children with disabilities spend a disproportionate part of their time in an environment, such as a school or institution, where play tends to be structured, organised and restricted'. In order to provide meaningful play opportunities for all children, especially those with disabilities, it is necessary to understand the needs of children and the implications of a disability on the child. Children need a place to play and they also need space with informality, and freedom to move around and make noises<sup>1</sup>. Society has often viewed people with disabilities as less attractive, helpless and needy. These societal attitudes and perceptions have caused people with disabilities to be less likely to partake in recreation programs<sup>30,31</sup>. 'Lack of participation does not make them 'less of a person' (a common perception of utility measures) as the cause may well be environmental or societal'<sup>32</sup>. This feeling of inferiority of children with disabilities is one factor that may limit participation in recreation programs. There is an inconsistency of the inclusion of children with disabilities in recreation programs due to a host of other barriers<sup>33</sup>.

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# PARTICULARITIES OF TUBERCULOSIS IN PREDIALYSIS PATIENTS FROM SOUTHEASTERN ROMANIA

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**Abstract**. In immunocompromised patients, a high incidence of pulmonary and extrapulmonary tuberculosis (TB) has been reported. This category also includes patients with chronic kidney disease (CKD), which can develop paucisymptomatic clinical forms, atypical onset and unusual clinical forms, frequently causing late diagnosis, and multiple complications. The study represents a prospective trial conducted between January 2013 and December 2015, and included initially a number of 364 patients with moderate CKD, monitored both in the Nephrology Department from the Emergency Clinical County Hospital from Constanta, Romania, and in Pneumophtysiology Hospital from Constanta, Romania. From our study group, 43 patients (11.8%) were diagnosed with tuberculosis. Extra-pulmonary TB (53.6%) predominated over pulmonary TB. Nine patients (20.9%) were suspected to have pulmonary TB by clinical and radiological signs but negative cultures. The therapy used in our patients was the same that guidelines recommend for those with normal kidney function. After therapy, 37 patients (86%) were declared cured from TB, 3 patients (6.9%) died during treatment and 3 patients were lost for follow-up. Any patient with CKD with an abnormal chest x-ray consistent, or previous history of extrapulmonary TB should be monitored. Appropriate therapy should be conducted only with both pulmonologist and nephrologist consultation.

Keywords: tuberculosis, chronic kidney disease, treatment.

# AIMS AND BACKGROUND

Tuberculosis (TB) is a known cause of morbidity and mortality worldwide, but especially in low to middle-income countries. Different studies report that extrapulmonary tuberculosis infection accounts for 5.8–44.4% of all TB cases and affects the lymph nodes, pleura, urinary tract, bones, joints, eyes, and skin. In patients with immunocompromised status, like HIV patients and patients with

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chronic diseases, including those with moderate-to-severe chronic kidney disease (CKD), extrapulmonary TB was reported more frequently than in healthy people<sup>1</sup>.

The relationship between tuberculosis (TB) and chronic kidney disease were studied in the last decades, but the interactions and associations between these two diseases have been poorly understood until now. Kidney transplanted and dialysis patients seem to be at a higher risk, but the environmental, socioeconomic, demographic and comorbid factors are cited, too<sup>1,2</sup>.

Considering the increasing prevalence of CKD in tuberculous endemic areas, the combination of these chronic diseases could have significant public health implications, especially in middle-income regions, like south-eastern of Romania. The global TB incidence in Constanta County is 80 cases to 100 000 inhabitants, above the national values (70%000).

It seems that CKD patients have 30 times higher prevalence of TB than in general population. Also, it has been estimated that patients undergoing dialysis have 10 to 12 fold higher risk of developing TB compared to general population. The incidence of TB in renal transplant recipients has been ranged from 1 to 4% in Northern Europe, 0.5 - 1% in North America and nearly 5 to 10 % in India<sup>3,4</sup>.

*Pathogenesis of increased incidence of TB in CKD patients*. The host response against intracellular agents, including *Mycobacterium tuberculosis* (MTB), is producing a higher production of interferon (IFN)-c. Elevated IFN-c is determined by increased Th1-cell response, with the important implication of interleukin (IL)-12.

The effects of uremic serum, specific for predialysis patients, are characterised by:

- acquired immunodeficiency state, leading to excessive infections with related morbidity and mortality,

- granulocyte dysfunctions regarding chemotaxis, adherence, and phagocytosis,

- defective antigen-presenting cells functions,

- persistent inflammatory state of monocytes5.

*Clinical features.* CKD patients can develop oligo-symptomatic clinical forms, difficult to diagnose early, as well as atypical clinical forms and many complications, sometimes life-threatening. The onset is usually insidious, patients accusing loss of weight, accompanied by anorexia and sub-fever, all these symptoms being sometimes covered by the uremic syndrome.

In a study published by Pien et al., the authors found fever occurring in  $\sim 72\%$  of the cases (range 29–100%), malaise in  $\sim 69\%$  (range 29–100%), and weight loss in a mean of 54% (range 10–100%). Nevertheless, asthenia, sweats and night subfever, as well as chronic, persistent cough and hemoptysis, considered as classic symptoms of TB in the general population, are less frequently reported in dialysis patients (mean 22% of cases; range 5–71%) (Ref. 6).

Diagnosis. The diagnosis of TB is based on the finding of an acid-fast bacillipositive smear, positive culture of *M. tuberculosis*, positive molecular tests, and typical histopathologic findings. Efforts should be made to obtain appropriate materials for culture, which should include sensitivity testing. The prevalence of latent TB infection (LTBI) in end-stage renal disease (ESRD) patients is elevated, and infected patients are at high risk of developing active disease. The tuberculin skin test (TST), considered the classic diagnostic tool for LTBI has poor sensitivity (because of a high prevalence of anergy in CKD patients) and specificity [with false-positive tests in those vaccinated with bacilli Calmette-Guérin (BCG)]. After a long time of skin testing use for the diagnosis of latent TB infection, clinicians are using now blood-based, immunological tests diagnostics, like interferon  $\gamma$ release assays (IGRAs), associated with a higher sensitivity and specificity in the diagnosis of latent TB infection. Therefore, current strategy in predialysis and dialysis patients should use these tests instead of TST for LTBI screening7. However, IGRAs and (PPD) (Mantoux) skin tests, which are positive in 40-100% cases, diagnose LTBI and are indirect elements for sustaining active TB disease.

Methods used for diagnosis of active TB are:

- positive PCR GeneXpert for Myc. tuberculosis (MTB);

- BK positive in sputum examination;

- Imagistic findings (chest X-ray, CT scan) usually associated with clinical context. CT scan is more sensible to discover the characteristic TB lesions usually disposed of the upper lobes. The pulmonologist should appreciate the activity of the disease and could sustain the diagnosis and start the treatment before bacteriological confirmation which usually needs two months, sometimes a lengthy process to delay the treatment.

*TB therapy in CKD*. Nevertheless, TB therapy in CKD patients is difficult, because of the multiple drug interactions and modified pharmacokinetics of drugs in renal dysfunction.

Factors that influence treatment of a CKD patient with tuberculosis are:

- drug pharmacokinetics, excretion by kidneys, as well as clearance by dialysis filters (in hemodialysis patients) and peritoneum (in peritoneal dialysis patients), with consequences on the serum concentrations of drugs and their toxic effects.

- multiple co-morbidities, with possible drug interactions.

- acquired immunosuppressive status induced either by uremic serum, in endstage renal disease (ESRD), or by drugs (in renal transplant recipients).

Treatment duration should, however, follow guidelines, 6–8 months for most cases of a fully sensitive TB, except of disease involving the central nervous system (CNS), when treatment should be for 1 year (Ref. 8).

Isoniazid is metabolised by the liver into less active compounds which are finally excreted by the kidneys. Pharmacokinetic studies monitoring isoniazid levels in CKD did not confirm the need for dosage reduction. However, there was no evidence that reducing isoniazid decreases the side effects, as well as the risk of drug resistance.

Recommended doses of Isoniazid in CKD are:

Stage 1-3 of CKD Isoniazid 300 mg;

Stage 4–5 of CKD Isoniazid 300 mg.

Renal Transplant Recipients 15 mg/kg BW, maximum 900 mg X 3 times/ week (Ref. 9)

Rifampicin is metabolised mainly by the liver, but its inactive metabolite, formyl-rifampicin, has urinary excretion, and its major metabolite, 25-desacetyl rifampicin is excreted in the bile. So finally, only about 10% of rifampicin are found unchanged in the urine. The side effects caused by rifampicin seem to have almost the same frequency in patients with CKD or on dialysis, in spite of the fact that this tuberculostatic drug has been cited as a rare cause of acute kidney injury. So, it is agreed between pulmonologists and nephrologists that the dose of rifampicin remains unchanged in CKD patients and that drug levels need no monitoring during therapy<sup>10</sup>.

In conclusion, isoniazid (H), rifampicin (R) and pyrazinamide (Z) can be used in usual doses even in patients with altered kidney function. Randomised controlled clinical trials have shown that pyrazinamide is therapeutically more effective if administered three times per week than with daily administration. In the meantime, pyridoxine supplementation should be given with isoniazid to prevent the occurrence of peripheral neuropathy, knowing that uremic serum of CKD patients already determines severe polyneuropathies in this special population group.

The recommendation doses of the first line drugs are shown in Table 1 (adapted after Ref. 11).

Drug	Stage 1–3 CKD	Stage 4–5 CKD*	Renal transplant re- cipients
Isoniazid	300 mg daily	300 mg daily or 15 mg/kg max 900 mg 3×/week	300 mg daily
Rifampicine	>50 kg: 600 mg daily <50 kg:1.5 g daily	<50 kg: 450 mg daily >50 kg: 600 mg daily 25–30 mg/kg 3×/week	<50 kg: 450 mg daily >50 kg: 600 mg daily <50 kg: 1.5 g daily
Pyrazinamide**	>50 kg: 2 g daily		>50 kg: 2g daily
Ethambutol	15 mg/kg daily	15–25 mg/kg 3×/week (max 2.5g)	15 mg/kg daily
Ofloxacin/ Moxifloxacin	400 mg/day	no dose adjustment required	no dose adjustment required

Table 1. Recommended doses of first-line drugs in chronic kidney disease (CKD)

\*Also applies to dialysis; \*\*check uric acid and monitor for gout.

The grades of renal impairment in CKD were recorded after Milburn and contributors<sup>11</sup>:

Stage 1 CKD:  $eGFR > 90 \text{ ml/min}/1.73 \text{ m}^2$ , but morphological/urinary abnormalities > 3 months; Stage 2 CKD:  $eGFR = 60-89 \text{ ml/min}/1.73 \text{ m}^2$ ; Stage 3 CKD:  $eGFR = 30-59 \text{ ml/min}/1.73 \text{ m}^2$ ; Stage 4 CKD:  $eGFR = 15-29 \text{ ml/min}/1.73 \text{ m}^2$ ; Stage 5 CKD: eGFR < 15 ml/min with or without dialysis (HD or PD).

The aim of the present study was to monitor the clinical, laboratory profile, management and outcome of pulmonary or extra-pulmonary TB in patients with CKD.

#### EXPERIMENTAL

The prospective study was conducted in a single centre between January 2013 and December 2015, and included a number of 364 patients, with moderate chronic kidney disease (eGFR 30–60 ml/min/1.73 m<sup>2</sup>), according to MDRD formula. They were monitored both in the Nephrology department at the Emergency Clinical County Hospital, Constanta, and in Pneumophtysiology Clinical Hospital, Constanta, Romania.

From our study group of 364 patients, there were 43 patients (11.8%; M/F = 24/19), a mean age of  $46 \pm 18$  years, either with positive diagnosis or with high clinical and/or imagistic suspicion of pulmonary or extrapulmonary TB.

Positive diagnosis was sustained on molecular detection of MTB in urine/ sputum /broncho-alveolar lavage (BAL), using molecular tests as Gene Xpert (GeneXperRIFTB) and /or LPA (Geno Type Line Probe Assays) and identification in culture on Löwenstein-Jensen solid medium and rapid culture as Bactec- MGIT 960 using liquid medium.

All CKD patients with a diagnosis of TB started anti-tuberculosis treatment (ATT) using isoniazid, rifampicin, pyrazinamide  $\pm$  ethambutol and/or ofloxacin/ moxifloxacin, usually 9–12 months, in doses adjusted according to the evolution of renal function.

The demographic characteristics of our study group, with moderate CKD and diagnosed with tuberculosis (TB +) are presented in Table 2.

Characteristics	CKD patients/TB + $(n = 43)$	
Age (years)	$46 \pm 18$	
Gender M/F	24/19	
eGFR (ml/min/1.73 m <sup>2</sup> )	$49.7 \pm 19.3$	
Urban/rural	31/13	
Arterial hypertension	32/43	
DM type 2	12/43	
Gout/hyperuricemia	7/43	
Obesity	5/43	

 Table 2. Demographic characteristics of CKD patients with tuberculosis

The Modification of diet in renal disease (MDRD) Study equation and the Chronic kidney disease epidemiology collaboration (CKD-EPI) equation are the most widely used for estimating GFR in adult patients (age > 18 years). The IDMS-traceable MDRD Study equation (for creatinine methods calibrated to an IDMS reference method) is:

eGFR (ml/min/1.73 m<sup>2</sup>) =  $175 \times (S_{cr})^{-1.154} \times (age)^{-0.203} \times (0.742 \text{ if female}) \times (1.212 \text{ if African American}).$ 

The equation does not require weight or height as variables, the results being reported normalised to the medium accepted average of  $1.73 \text{ m}^2$  body surface area. The CKD-EPI equation has been validated extensively in Caucasian and African American populations between the ages of 18 and 70 years old, with impaired kidney function (eGFR< 60 ml/min/1.73 m<sup>2</sup>) and, until now, has shown good performance for patients with chronic kidney disease<sup>12</sup>.

#### RESULTS

From our 43 CKD patients, TB positive, in 11 patients (25.5%) the diagnosis of pulmonary tuberculosis was sustained by positive cultures either in sputum or BAL, and in the rest it was empirical based on clinical picture, pleural fluid analysis, and radiological tests. Tuberculin skin test was negative in 25 patients (58%).

Nine patients (20.9%) were suspected to have pulmonary TB by clinical and radiological signs but negative cultures, and 23 patients (53.6%) proved to have extra-pulmonary TB. Extrapulmonary TB (53.6%) predominated over pulmonary TB. The localisation of extra-pulmonary tuberculosis, were: urinary -9 cases; pleural -5 cases, pericardial -2 cases; peritoneal -2 cases; miliary TB -2 cases; spinal -1 case; central nervous system -1 case; lymph node -1 case (Table 3).

Localisation	No of patients	%
Pulmonary	22	51.2
– miliary TB	2	
– proven TB	11	
– suspected	9	
– TB		
Urinary	9	20.9
Pleural	5	11.6
Pericardial	2	4.6
Peritoneal	2	4.6
Spinal	1	2.3
Lymph nodes	1	2.3
Central nervous system	1	2.3
Total	43	100

Table 3. Localisation of tuberculosis in CKD patients

Fever/subfebrilities, malaise, weight loss were the most common symptoms in our CKD patients, observed at the admittance in the Pneumology department. In more than 25% of the patient elevation of non-specific inflammatory tests was noticed (ESR, CRP, unexplained leukocytosis).

TB positive patients were treated with tuberculostatic drugs for 6 to 9 months and, finally, 37 patients (86%) were considered cured with no relapse, 3 patients (6.9%) died during treatment, and 3 patients were lost to follow-up.

The therapy used in our patients was: 1 - initiation phase: 2 - months isoniazid (H 5 mg/body weight), rifampicin (R 10 mg/body weight) and pyrazinamide (Z 30 mg/body weight) 7 days/week (7/7) and ethambutol (E 25 mg/body weight) 3 days out a week (Monday, Wednesday, Friday); <math>2 - consolidation phase: 6 months H10mg/body weight, R10 mg/body weight 3/7.

From the total group of 43 TB treated patients, 29 (67%) accused several side-effects potentially related to anti tuberculous therapy (ATT).

The most common ATT-related adverse event were:

- allergic drug reaction (16.3%),

- gastro-intestinal (14%), followed by
- drug induced hepatitis (11.6%) and
- cutaneous-mucous bleedings (7%).

Other notable adverse reactions included: isoniazid-induced psychosis (4.6%) and blindness (2.3%), ethambutol-related administration, and *worsening of* renal function, which was observed in 9.3% and only one patient started haemodialysis (Table 4).

Side effect	No patients	%
Allergic reactions	7	16.3
Gastro-intestinal	6	14.0
Drug-induced hepatitis	5	11.6
Aggravation of CKD	4	9.3
Bleedings	3	7.0
Psychosis	2	4.6
Blindness	1	2.3
Stroke	1	2.3

Table 4. Adverse reactions ATT-related in CKD patients

From the 6 patients with gastrointestinal side effects, 2 patients were diagnosed with haemorrhagic gastritis, that stopped under conservatory therapy (proton-pump inhibitors and haemostatics i.v.).

The study group had a 6.9% mortality rate, 3 patients died because of: severe respiratory failure, associated with miliary tuberculosis (1 case), stroke associated with miliary TB (1 case), tuberculous meningitis (1 case) (Fig. 1).

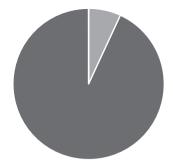


Fig. 1. Mortality in CKD patients with tuberculosis

There was an exceptional case, of an 83 years old male patient, in which the evolution under therapy showed even renal function improvement after appropriate treatment for urinary tuberculosis.

#### DISCUSSION

In this study, it was shown that patients with chronic kidney disease (CKD) have a high incidence (11.8%) of tuberculosis (TB). The frequency was similar to other studies from low to middle-income countries, like India<sup>13</sup> or China<sup>3</sup>. It is evident that our patients diagnosed with chronic kidney disease considered at risk for tuberculosis should have a history of previous TB or TB contact, an appropriate clinical and paraclinical approach (chest X-ray, abdominal ultrasound, CT scan). Furthermore, patients with previous adequate TB treatment should regularly be monitored; even they switch to renal replacement therapy and reached to a private dialysis centre, and referred periodically to a pulmonologist<sup>7,14</sup>.

As we already mentioned, TB positive patients were treated with tuberculostatic drugs for an interval of time of 6–9 months, and 47% were considered cured, with no relapses and only 3 patients, with severe co-morbidities died during the therapy. The treatment used in our CKD patients was: in the initiation phase – 2 months Isoniazid (H 300 mg), rifampicin (R 600 mg) and pyrazinamide (Z 1500–2000 mg) 7 days/week (7/7) and Ethambutol (E 800) 3 days/7; and in the consolidation phase: 6 months H600 R600 mg (3/7).

From the total group of 43 TB treated patients, 29 (67%) accused many sideeffects potentially related to antituberculous therapy (ATT).

In a study that monitored all cases of tuberculosis and CKD in patients from Manchester Royal Infirmery, in the interval of time 1986–1999, 24 cases were identified by diagnostic coding, microbiology records and a TB database, 8 in predialysis and 16 requiring regular dialysis. TB occurring in the dialysis group was extra-pulmonary in every case. Adverse effects of treatment occurred in 2 of 8 (25%) in the predialysis group, and 56% in the group treated by dialysis. The

most frequent side effects of the tuberculostatic drugs in CKD were: neurological, hepatic and gastrointestinal. Because of severe neurological and psychiatric effects, CKD patients can be considered high-risk group, needing careful monitoring during therapy<sup>15</sup>.

A rare case of stroke, associated with miliary tuberculosis, that lead to the death of our patient, was noticed, a similar case being reported in our region in a TB patient with normal kidney function<sup>16</sup>.

#### CONCLUSIONS

We observed an 11.8% incidence of tuberculosis in our CKD pre-dialysis patients from Constanta county. The extrapulmonary form of tuberculosis (53.6%) predominated over pulmonary form. From 43 TB patients, 37 were considered cured under anti-tuberculosis therapy. The most common related adverse events in CKD patients were: allergic drug reaction, gastro-intestinal, drug induced hepatitis and bleedings. Worsening of renal function was observed in 9.3% and only one patient started hemodialysis. The study group had a 6.9% mortality rate because of the extensive forms of TB.

Patients with moderate to severe chronic kidney disease, associating unexplained increase of nonspecific inflammatory tests, abnormal chest X-ray and antecedents of pulmonary or extrapulmonary tuberculosis, should be monitored regularly and considered for referral to a pulmonologist. The chemoprophylaxis for LTBI should be made only after discussion with the patient and a team formed by the pulmonologist and nephrologist.

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Sustainable landscape planning

# MODELLING OF THE ASSESSMENT OF THE ENVIRONMENTAL IMPACT FOR THE ECOLOGICAL RESTORATION OF POLDERS IN THE DANUBE DELTA

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**Abstract**. The dynamics of the systems belonging to the natural capital is the result of the interaction between their own structural and functional parameters with external factors, among which one can identify those generated by projects for ecological reconstruction. Starting from this conclusion, one can compare the parameters before and after applying the polders ecological reconstruction projects; the noticed differences shall be the types and forms of physical expression evaluated in impact and value. Taking into account a series of parameters that characterize the types and forms of impact, aggregation of different impacts is considered necessary, so that the net impact and the opportunity of ecological reconstruction projects can be determined.

Keywords: structure, functioning, status transition, net impact.

# AIMS AND BACKGROUND

The aim of this paper is to improve the role of cognitive factors and to promote the adaptive ecosystem management of socio-ecological complexes by strengthening the support system of environment decision making.

Generating alternative methodologies for environmental impact assessment (part of the ecological impact) has an impact on the following elements of the system and adaptive ecosystem management support<sup>1</sup>:

• infrastructure for social and economic analysis (packages of methods and techniques for the economic evaluation of the natural capital);

• infrastructure for environmental impact assessment (package of methods for analysis and environmental impact assessment);

• infrastructure for assessing the sustainability (packages of processes and indicators to assess and quantify the level of sustainability).

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In a broader sense, the environmental impact assessment modelling is an approach meant to contribute to addressing eco-crisis, among which the loss/damage of services generated by wetlands that is their self-support capacity (Fig. 1).

On the other hand, methodologically, we consider that although there is agreement on environmental impact assessment stages, their content is not sufficiently systematised in terms of the interdependencies of the factors and the elements which are analysed/evaluated<sup>2</sup>.

Respectively, the impact assessment (identifying the types and forms of impact, and possibly measuring their physical expression), without performing the evaluation phase itself (in monetary expression), can not allow aggregation of the impacts and determining their 'net impact'<sup>3</sup>. The importance of going through the second phase is even higher as the resources allocated to programs and projects for conservation of the natural environment rose significantly, thus emphasising the need for formulation and adoption of environmental decisions in such a manner that they should be economically efficient (not only socially and ecologically)<sup>4</sup>.

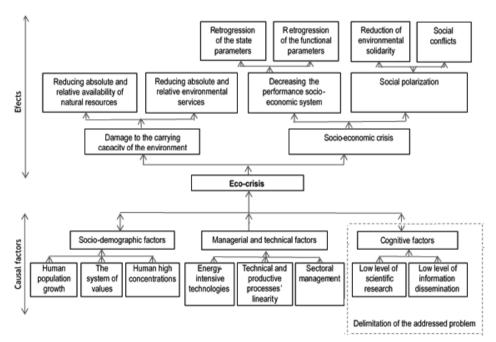


Fig. 1. Dematerialisation of the development in the context of eco-crises problems (source: own representation, 2016)

#### EXPERIMENTAL

Initially, Babina island (as Cernovca island), 2237 ha (1985) was an area that operated under natural conditions, lower land with its own hydrographic network, connected to the river Danube (Chilia) by Horbal Backwater.

Between 1985–1989, with the enforcement of the 'program for improvement and full exploitation of the natural resources in the Delta' (Decree No 92/1983), the structural and functional parameters of the area mentioned experienced significant changes (Table1).

The transformation of the Babina island in the Agricultural Area (goal fulfilled as late as 1989) involved mainly hydro dam and drainage work, followed by terracing and levelling, the execution of deep plowing of 28–30 cm (for the destruction of the reed rhizome) work that radically changed the sub soil, vegetation, and fauna.

Reconnecting the Babina Agricultural Area with the flooded Danube (April 1994), with the purpose of restoring the ecosystems typical for the alluvial areas, although it did not mean a complete return to the regime of operation before 1985 was designed to satisfy these requirements<sup>5</sup>:

• water levels in the area must be kept as long as possible during the bioactive period;

• water levels in the area must be given by the water level of the branch of the Danube;

• a permanent movement of water must be ensured as far as possible (given that most of the river system of the agricultural area had been abolished).

Consequently, the habitat structure, as well as the vegetation and fauna recorded a rapid return to the characteristics of the specific alluvial areas, accompanied, of course, by a regain of the ecological functions<sup>5</sup>.

#### **RESULTS AND DISCUSSION**

The issue in question highlights the significant impacts of the transition of the status of Babina island from agricultural are to that of the natural/half-natural area, following the implementation of the project of re-vegetation by making openings in the dam, based on an optimum solution in the given situation.

Parameters of the reference area	Status of the reference area				
	natural	anthropo-	natural/		
	(multi-func-	genic	half-natural		
	tional)		(mono-func-		
		tional)	tional)		
Structural parameter	ers (of habitat	s)			
Pond	Х,-	X, ↓	$X\uparrow$		
Meadows	Х,-	X, ↓	$X\uparrow$		
Plots for traditional agriculture	Х,-	0	Х,-		
Groves	Х,-	0	Х,-		
Functional parameters (	ecosystem ser	rvices)			
Habitat for plants and animals typical of al- luvial zone	X	0	-		
Habitat and breeding area for fish	Х	0	-		
Habitat and breeding area for birds and water- fowl: mud	Х	?	Х		
Reservoir for biodiversity and genetic resources	Х	0	Х		
Bio-corridor and exchange genetic	Х	?	Х		
Biological production	Х	Х	Х		
Bio-geo-chemical circuit	Х	0	Х		
Sediment retention and fixation of toxic sub- stances	Х	0	Х		
Bio-filter for the Black Sea	Х	0	Х		
Connectivity	100%	?	< 100%		
Energy flows	natural	anthropogen- ic + natural	natural		

Table 1. Change of structural and functional parameters within the Babina island

X – existing category of habitat/ecosystem service; 0 – absent class of habitat/ecosystem service; -= constant;  $\uparrow =$  rising;  $\downarrow =$  falling.

Taking responsibility for the option in such a case requires effort and environmental impact assessment (EIA) of the various technical solutions with the aim of determining the net impact and comparing it with the investment effort. Of course, such an approach is meant to reinforce the decisional support (along with the economic and social impact), so that the decision of re-vegetation and the solution adopted should be relevant in terms of efficiency.

To this end, the present research has been oriented towards these two directions to promote the systems approach (based on matrices of interdependencies between the explanatory factors of going through each stage of EIA) and ensuring finality of EIA, in terms of the decisional act:

- matrix expression of stages of EIA;
- qualitative and monetary assessment of EIA.

As a process, the environmental impact assessment is performed in a sequence of steps over which, in the existing literature<sup>6–9</sup> there is a fairly broad consensus:

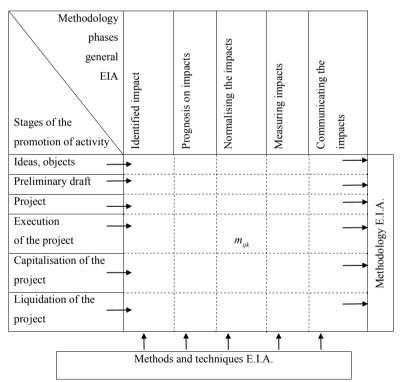
- identification of impacts,
- forecasting impacts,
- normalisation of impacts,
- measuring impacts,
- communication of impacts.

The innovations we propose in this regard, for the shaping of the evaluation of environmental impacts are derived from a matrix presentation of some of the steps mentioned, which can bring more relevant results and their interpretation.

We appreciate that the generation of alternative evaluation methodological structures can be achieved depending on (Fig. 2):

• steps to promote economic activities and social/ecological reconstruction of polders,

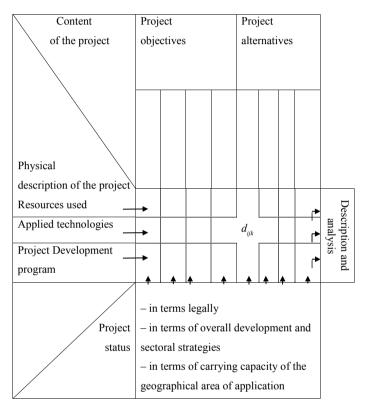
- stages of the general methodology of environmental impact assessment,
- a package of methods and techniques of environmental impact assessment.



**Fig. 2.** Methodological alternatives for matrix structures to assess the environmental impact<sup>5</sup>  $m_{ijk}$  – component of EIA methodology: 'j' is the EIA method/technique utilised in the 'j' stage of the EIA methodology and the 'k' stage of project promotion.

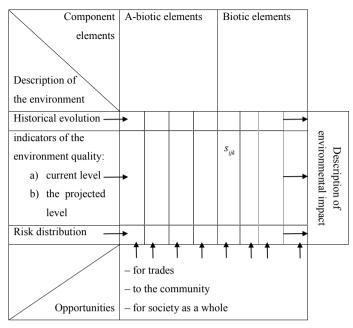
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In terms of identifying environmental impacts, and the interdependencies between the natural environment and the project to be realized, organizing data and information, ensuring relevance, can rely on the following matrix (Figs 3 and 4). Project description and analysis (Fig. 3) are meant to emphasise the complementary nature or the joint responsibility of its objectives and the potential pressure on natural resources, both in appearance and structure, influencing the structure and the extent of the measures for remedying the impacts.



**Fig. 3**. Matrix project description and analysis of an activity and its alternatives<sup>5</sup>  $d_{ijk}$  – component of the project description/analysis: *i* is the resource/technology necessary to achieve the goal 'j' in the context of the 'k' restriction (restriction can be administrative, socio-economical or related to the conservation of the ecosystem services).

We believe that a special place in the description of the project should be given to the level of integration in the local and regional development strategies, so that some synergistic impacts/ effects on the natural environment can be anticipated. In turn, the description of the natural environment to which the project will be connected (Fig. 4) is based on a series of indicators that allow a better understanding of the dynamics and ecological vulnerability.



**Fig. 4**. Matrix to describe and analyse the environmental impact<sup>5</sup>  $s_{ijk}$  – component of the description of the ecosystemic service/natural impact environment: '*i*' is the opportunity for capitalisation of the ecosystemic service/the natural environment element '*j*', in the context of the '*k*' characteristic of the ecosystemic service/the natural environment element '*j*'.

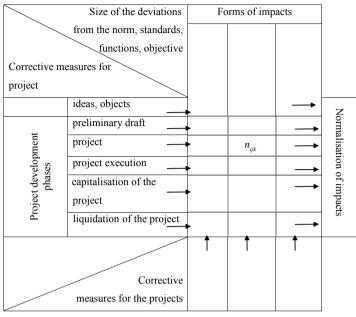
If the identification of impacts, i.e. comparison of the tree structures of the project and of the natural environment, is a classic approach, we appreciate that we cannot speak in the same terms when we go through the stage of normalisation of impacts. In this context, the proposed matrix (Fig. 5) endorse the concerted action and not unidirectional, namely the identification of corrective measures not only for the project but also for the natural environment, to ensure the largest possible compatibility between the two elements.

The functional profile of the polder is defined on the basis of the cardinal and ordinal utility of ecosystem services.

Cardinal usefulness will be assessed depending on the score obtained by the ecosystem services identified by the local users, the regional, and the national ones.

Ordinal usefulness will be evaluated taking as a starting point the technique based on the choice modelling, which is part of the hypothetical market technique.

For expressing the ordinal usefulness one uses important coefficients of ecosystem services that define the functional profile of the polders, established by the help of the DELPHI technique, meaning the 'comparison of pairs'. The level of importance coefficient which are used (Table 2) is determined as an average, describing the place and the role of the different categories of users based on their majority rule: local users 51%; regional users 34%; national users 15%.



#### Fig. 5. Normalisation of impacts matrix<sup>5</sup>

 $n_{ijk}$  – possibility to normalise the impact: '*i*' is the corrective measure of the project/alternative, compatible with the '*j*' corrective measure of the natural impact environment for the reduction of the '*k*' deviation from the norm/standards.

Table 2. Functional profile of Babina island	(hypothetical data)
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Eco-systemic service	Coefficients of importance* (ordinal usefulness)					
(cardinal usefulness)	local	regional	national	pondered average		
Habitat for plants and animals				0.179		
Habitat and reproductive area for fish				0.166		
Habitat and reproductive area for birds				0.103		
Tank for biodiversity and genetic resources						
Production of fish				0.073		
Production of reed				0.066		
Production of fodder				0.062		
Agricultural production				0.054		
Resources for tourism				0.049		
Sediment retention and fixation of toxi substances	с			0.046		
Black Sea bio-filter				0.036		
Flood protection				0.035		
Information resources for research				0.027		
Scenery resources				0.019		

\* weighted average of the coefficients determined at local/regional/national level.

Obviously, there are differences (sometimes significant) regarding the perception of ecosystem services, but we believe that the decision makers/national and regional users must develop programs/projects to improve ecosystem services perception by local users. Considering the ordinal usefulness, one can identify the representative eco-systemic service from the view point of its reflection in the real market.

In the present case we believe that we can opt for the fish biomass based on the following considerations:

• there is a growing demand for the natural good 'fish';

• the fish market operates within competitive conditions, starting with fishermen associations, distributors and retailers.

The whole fish offer is characterised based on the following elements:

- average annual production increase in the fish biomass;
- level and trend of the total catch of fish;
- fish biomass structure and that of and the fish capture.

The level of exploitable fish biomass taken into account (commercial efficiency of the Babina island/potential bid of fish) is the one resulted from the systematic investigation conducted by the Danube Delta National Institute/DDNI, based on the method of De Lury. Measurements taken after the natural re-birth found the fisheries potential of 71 t/year, meaning 34 kg/ha/year. Considering how the fishing activity is organised, the methods and means used for this purpose, we appreciate the level of catches of fish (demand) of 57 t, meaning 27 kg/ha/year (approximately 80% of the commercial efficiency of the polder). The price used to determine the market value of the catch of each species of fish reflects the structure of the species and the conditions of delivery 'venue arrangement'). Its level, as a result of the official decision stated by the County Council No 90/2009 (amended) relating to the justification of the payable sum for fishing grounds is 6 RON/kg. Calculating the consumer surplus has as support the relationship between the consumer and the market value, both dimensioning the consent to pay.

CD = the maximum bearable demand, in terms of the sustainable polder;

CP = the actual demand expressed/actual catch;

PP = market price; VP = market value; SC = consumer surplus.

Determining the value of the eco-systemic service (VS): VS = VP + SC.

No	Indicators	Symbol	M. U.	Level of the indicator
1	maximum bearable demand	CD	kg/ha/year	34
2	effectively expressed demand	HP	kg/ha/year	27
3	market price	PP	RON/kg	6
4	market value (row $2 \times row 3$ )	VP	RON	162
5	consumer surplus	SC	RON	310
6	value of the eco-systemic service	VS	RON	472

Table 3. Value of the eco-systemic service

The values are determined (Table 4) by interpolation, considering the usefulness of ecosystem services (their ordinal utility).

Eco-systemic service (cardinal usefulness)	Value
Habitat for plants and animals	1157
Habitat and breeding area for fish	1073
Habitat and breeding area for birds	666
Tank for biodiversity and ensuring genetic resources	550
Production of fish	472
Production of reed	427
Production of fodder	402
Agricultural production	349
Resources for tourism	317
Sediment retention and fixation of toxic substances	297
Black Sea bio-filter	233
Flood protection	226
Information resources for research	174
Scenery resources	123
Total	6466

 Table 4. Value of ecosystem services RON/ha/year

The state transition of the reference area/ Polder Babina (Table 1) reveals that during its evolution as agricultural area, its functional parameters, with few exceptions (parameters 2.3, 2.5) have zero values, thus the value of the restauration project impact is equal to + 6466 RON (approximately 1440 EUR/ha/year). We emphasise, however, that the amount mentioned was computed considering the amount of using the eco-systemic service 'fish biomass'; a possible determination of the values of non-utility would lead us to a greater positive impact.

# CONCLUSIONS

Systematising the stages of evaluating the Environmental Impact Assessment (EIA) implies an effort towards the efficiency of the matrices involved, to offer more transparency, relevance and credibility of the results. All of these are absolutely necessary given the growth of the social environmental decision. Establishing the functional profiling of the reference area – Babina island – requires an effort in consulting the local/ primary/ secondary/tertiary/ regional and national users, to measure the cardinal and ordinal usefulness of services of the eco-system. For the EIA, we have to combine the factual data with the hypotheses formulated by the expert groups, after gathering the information and the facts.

The net environmental impact, established as the difference between the value of the eco-systemic services before and after the Babina island restoration, represents a minimum level, comparable to the one established for other wet areas.

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Sustainable landscape planning

# DETERMINATION OF THE VITAL ECOLOGICAL NETWORKS: THE CASE OF EUROPEAN SIDE OF TURKEY

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**Abstract**. The European side of Turkey is under environmental pressure due to the intensive use land. In this study, the identification of existing ecological value and ecological networks planning possibilities were explored. Within the scope of this research, a general overview of the status of the ecologic corridors forming the ecologic networks on the European side of Turkey was exhibited with 1/100 000 scale. The corridor areas, which are determined by prioritising the vegetative density and area usage decisions, are important elements that provide livelihood of the living in the area. There is a very dense human-oriented pressure on the peninsula due to reasons such as agriculture, industry and rapid population increase. It is necessary to support the ecologic corridors, one of the most important arguments to decrease the damage to the ecologic diversity on the peninsula, and preserve them. This study evaluates the Turkish side of the Thrace Peninsula. However, the same study needs to be evaluated in the sides of Greece and Bulgaria. It is necessary to conduct the Balkans ecologic network system planning in a holistic way. The ecologic network planning studies of the European side of Turkey should be conducted cross-border along with Turkey, Greece, Bulgaria and other Balkan countries. The cross-border needs to be planned altogether to preserve and develop the ecologic network.

Keywords: ecologic networks, European side of Turkey, landscape planning, Balkan countries.

## AIMS AND BACKGROUND

Planning of ecological networks has vital importance for sustainabile approach in nature conservation. Ecological networks have many functions such as flood control, contamination filtration, erosion prevention and biodiversity conservation. As an important element in landscape planning and design, ecological networks can be used to reduce the negative impact of landscape fragmentation. According to their different structures and functions, ecological networks can be classified into linear, strip, and stream corridors<sup>1,2</sup>.

<sup>\*</sup> For correspondence.

A number of definitions of corridors and connectivity have been proposed over time. In earlier references, corridors were defined as routes that enhanced speedy and unselective spread of biota between regions<sup>3</sup>. Some times corridors are referred to as habitat corridors, wild life corridors, or ecological structures. They can be part of or the same as ecological or habitat networks, which encompass core areas, corridors, and connecting nodes; or they can be synonymous with greenways, greenbelts, or open space<sup>4</sup>.

Greenway has been an interdisciplinary process involving wildlife biologists, landscape architects, foresters, horticulturists, planners, lawyers, and area residents. The development and preservation of the greenway requires the cooperation of citizens and governmental agencies. The process addresses large-scale planning issues, detailed site design activities, and continued research investigations<sup>5</sup>. Taken as a driver of landscape degradation, soil sealing exacerbates the susceptibility of land to climate changes, erosion, forest fires and water shortage<sup>6</sup>. Green networks can also deliver a wide range of other benefits: improving health and well-being, enhancing bio-diversity, helping to mitigate against climate change, providing business and educational opportunities, encouraging tourism and promoting sustainable use of scarce land resources<sup>7</sup>.

Land use changes can affect ecosystem services values<sup>8</sup>. Land cover includes natural and planted vegetation and human construction, which covers the surface of the Earth. Growing urbanisation and industrialisation have caused increase of the construction and disapperence of natural area. As a result of this intensive lands use change causes many environmental and ecological degeneration<sup>8,9</sup>. This degeneration increasingly continues and creates negative effects at European side of Turkey<sup>10–15</sup> which does not have ecologic planned for land use. Ecologic networks are landscape elements that make up for the negative effects of habitat fragmentation in agricultural or natural landscapes.

Ecological networks concept is increasingly popular. For this is reason, it is important development of plans for ecological corridors and greenways have been rapid for the last decade. European side of Turkey is located in the western most corner of Turkey along the borders with Bulgaria and Greece, which are EU members. Along with hosting the highway passages between Turkey and Europe. Therefore planning of ecological corridors is even more important because it exceeds the cross-border in the region. Thrace region is under environmental pressure due to the intensive use land<sup>11,16</sup>.In this study, the identification of existing ecological value and ecological networks planning possibilities were explored.

#### EXPERIMENTAL

*Study area*: European Turkey is geographically part of Southeast Europe, all in the eastern part of the historical region of Thrace. The area includes all the territories

of the Turkish provinces of Edirne, Tekirdag and Kirklareli, as well as those territories on the European Continent of the provinces of Canakkale and Istanbul<sup>17</sup>. East Thrace has an area of 23 764 km<sup>2</sup> (3% of the country) and a population of about 10 million people; the population density is around 430 people/km<sup>2</sup>. The two are separated by the Dardanelles, the Bosphorus and the Sea of Marmara. The southernmost part of Eastern Thrace is called the Gallipoli peninsula. European side of Turkey is bordered on the west north by Bulgaria for 269 km and by Greece for 212 km, on the north with the Black Sea to the north-east and the Aegean Sea to the south-west<sup>17</sup>.

1/5000 scale municipal plan, 1/25 000 and 1/100 000 scale maps (land use/ lands cover maps, topographic maps, soil maps), implementation projects of under protection areas by Turkey in European side of Turkeyand coordinate measuring device (GPS) were used as study materials.

*Research method.* It has been handled in the context of ecologically crucial core areas and offering ecological corridors representing the spatial relations between them. Components of ecological network are core areas, buffer zone and ecological corridors. Ecological corridors established all connection of isolated core areas. Definition of ecological corridors is very important, for this reason this study aimed at determination of the vital ecological networks in European side of Turkey.

In the land classification CORINE database has been used<sup>18,19</sup>. First step of the study is evaluation of land use and land cover of study area. National Land Cover 2006 project applicated by Ministry of Forest and Water Affairs all over the Turkey. The project based on the CORINE (Co-ordination of Information on the Environment) standards. These standards are developed by European Environment Agency. The Thrace Region CORINE land cover and land use is obtained from this project. Thus land/cover use maps of the area were determined. Additionally plant density is generated by NDVI analysis of study area. NDVI (Normalised difference vegetation index) is commonly used in plant density and its distribution. NDVI is determinated all area between bare soil and vegetation cover areas. Thus NDVI is classifying all area less density to more density.

Also literature is used to determine to ecological sensitive areas.

In order to handle ecological networks as a whole, existing sensitive and pressure receiving core areas and ecological corridors that can bind these together have been prepared in upper scale (scale: 1/100 000) (Ref. 20). So as to determine these areas, areas that have been put under protection by state in Thrace have been handled alongside with many factors<sup>21</sup>.

The research procedure consisted of 4 steps. In the first step, natural structure of the research area and nearby was analysed and potential protection areas was determined.

In the second step, potential environmental negative impacts, the relations and contradictions between the other uses were identified. Areas provide connections between ecological networks and other land uses (like suburban, urban, etc.), were evaluated from the view point of planning and desing principles. Third step was the evaluation phase and it included the analysis of collected data. In the last phase of the study proposals that by utilising all the data, the progress of habitat fragmentation and environmental effects were analysed and fragmented spaces and their distributions were mapped.

# **RESULTS AND DISCUSSION**

Natural habitats and ecological systems provide living space for one or more living beings, and they are the source of biodiversity on Earth<sup>22</sup>.

Important protected areas in the Thrace region<sup>23</sup>. There are three protected zones in the Tekirdag province. These are 'Gungormez cave' near the road to Kastro in Saray district, plane trees on the coastal line of Ucmakdere bound to Sarkoy district, and the last one is in Kumbag village bound to the centre of the province named as Dut (Limani) harbour where the Sutluce monastery is located. On the Black sea coast in the Thrace region, on the border line of Turkey-Bulgaria, approximately 300-600 m away from the Black sea, near to Igneada, there is about 900 ha area of longos forests - an important bird area, and one of the most important longos forest entities in Europe. The Igneada longos has been given the status of Nature Site in 1991. There are 2 nature protection areas in this area: Kasatura Bay, Saka Lake Longos. Kiyikoy Natural Site: An important part of the Pabucdere and Kazandere creeks that flow into the Black sea is also taken under protection as a 1st grade Natural Site. Edirne province has a great potential due to its important protected areas. The Gala and Pamuklu lakes are declared to be Natural Parks in 2005. Also Black Sea SCENE will strengthen the contribution to necessary data and rehabilitation of the Black sea ecosystem<sup>24–27</sup>.

*Ecological network analysis in the study.* Determining ecologically important fragmented landscapes and constructing spatial relations between these landscapes<sup>23</sup> have been aimed; analyses for determining core areas and corridors have been carried out; in 9 chosen areas, core areas and ecological corridors are shown in Fig. 1. Coordinates and current situation analysis plates of these sample areas chosen in Thrace are given in Fig. 2.



Fig. 1. Proposal of ecological networks and ecological corridors in the study area



Fig. 2. Ecological network analysis in the study area

Core areas and corridors appointed as offer in Thrace Region and shown in Figs 3*A*,*B* have got a heterogeneous structure. This mosaic structure created by a combination of diverse vegetation is at the same time a factor that supports diversity of species in the research area. In the research, are as that stand between core areas and corridors and intense field use should be used as a buffer zone.Northern part of Thrace Region and Black Sea shore line constitutes the most natural area. Various area usages are increasing the pressure on these areas day by day<sup>23</sup>. In the work of constructing ecologic network, ecologic corridor is projected to be built and bound along Ergene River that passes amongst Istranca Forest, Demirkoy Igneada Longoz Forest National Park and Cilingoz Nature Park. By this way, a green network structure can be planned by binding different areas with two corridors. Areas surrounding Saroz gulf and especially Gala Lake National Park as well as Gallipoli Peninsula Historical National Park and green area texture on Koru Mountains have been seen to be some areas that can be evaluated within South Thrace ecological network system.

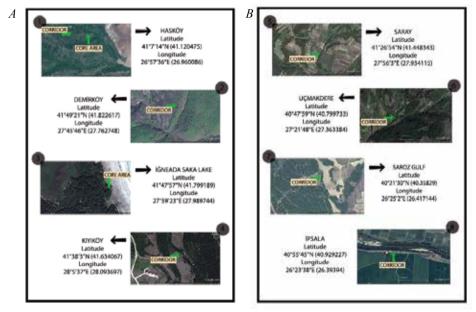


Fig. 3. Analysis of some ecological corridors and core areas in the study area

#### CONCLUSIONS

The analysis is based on findings obtained as a result of our survey. As a result of our analysis, we determine strong and weak zones of ecological networks in the north coastal part and South coastal part of Euperaen Turkey. According to findings

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it appears that the most powerful features of ecological networks in the north Trakya are rich natural resources, longos, convenient climate<sup>28</sup>. Planning the ecological networks is not an approach that depends on a specific scheme within the framework of strict rules; it is a process formed by objectives, expectations and possibilities. In other words, basic aim is to protect natural habitats and wild life, and also so many approaches discussed all over the world so as to accomplish the aims. However, no matter which approach is used for planning ecological networks, reaching detailed. sensitive and up-to-date information is fundamental<sup>29</sup>. Turkey has signed many international agreements in order to preserve its natural vegetation and fauna wealth. In order to preserve these areas, ecological networks and as components of these network systems, core areas and ecological corridors need to be determined. In this context, very limited number of researches has been performed so far. Hepcan has enabled this issue to be comprehendible and applicable in Turkey by determining core areas and ecological corridors in the scope of his dissertation, subject of which is 'Identification and Planning of Ecological Networks as a SustainabileApproach in Nature Conservation; The Case of Cesme-Urla Peninsula'. Within this scope, for the researches concerning ecological networks in Thrace, technical facilities, data supply and process should urgently be provided<sup>30</sup>. Within the scope of this research, a general overview of the status of the ecologic corridors forming the ecologic networks on the Thrace peninsula was exhibited with 1/100 000 scale. The corridor areas, which are determined by prioritising the vegetative density and area usage decisions, are important elements that provide livelihood of the living in the area. There is a very dense human-oriented pressure on the peninsula due to reasons such as agriculture, industry and rapid population increase. It is necessary to support the ecologic corridors, one of the most important arguments to decrease the damage to the ecologic diversity on the peninsula, and preserve them. This study evaluates the Turkish side of the Thrace Peninsula. However, the same study needs to be evaluated in the sides of Greece and Bulgaria. It is necessary to conduct the Thrace green network system planning in a holistic way. The ecologic corridor planning studies of the Thrace Region should be conducted cross-border along with Turkey, Greece and Bulgaria. The cross-border needs to be planned altogether to preserve and develop the ecologic network.

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Sustainable landscape planning

# IMPORTANCE OF THE BIOCLIMATIC COMFORT ON LANDSCAPE DESIGN: CASE STUDY ON CANAKKALE ESKI KORDON

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**Abstract**. Bioclimatic comfort is the climate conditions in which individuals feel them healthy and dynamic. In generating these environments different climatic parameters play a role and their roles are important in landscape design. As the climatic components that constitude comfort could be the main parameters – heat, relative humidity, wind alongside with radiation and noise – it could also be other forces. In this study bioclimatic comfort state of the shore area of Eski Kordon, a busy area which is located in Canakkale, was determined in terms of climatic data. In the study, data were obtained in the actual place by using measurement tools such as Termohygrometer, Microwave Radiometer, Digital Sound Level Meter, Luxmeter and Anemometer. As a result of these measurements, approximate heat, relative humidity and wind rate, light alongside with radiation and noise rate have been interrelated with structural and vegetative equipment. Bioclimatic comfort standard of the areas where there were a flowing vehicle and pedestrian traffic was also investigated by analysing relevant data. As the result of this study it was determined that the rates of vehicle and pedestrian roads do not meet with the required comfort standards, whereas some of the green areas provide these standards at some particular spots.

Keywords: bioclimatic comfort, Canakkale, Eski Kordon, landscape design.

## AIMS AND BACKGROUND

As the result of growing urbanisation and technological advancements both in our country and the world, there has been deterioration in urban ecosystems and in ecological stability. One of the most important outcomes of this destruction is climate changes<sup>1</sup>.

Climate is one of the most important factor for the livings on earth showing either positive or negative effects in people lives<sup>2,3</sup>. This factor, which also affects individual lives intensely, is important to be taken into account in urban designs<sup>4</sup>

<sup>\*</sup> For correspondence.

(Fig. 1). Bioclimatic comfort has been defined as the individual compatibility with their surrounding while using minimum of energy<sup>5</sup>.

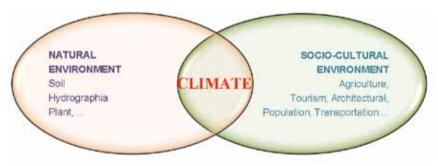


Fig. 1. Climate in the natural and socio-cultural environment<sup>6</sup>

For establishing the thermal comfort by modifying the elements of climate such as wind direction temperature, water surfaces are designed or planting and grading are done in the area<sup>7</sup>. Four factors that influence bioclimatic comfort are stated below, in order of importance (Fig. 2). These are:

- temperature (dry-bulb thermometer);
- atmosphere humidity;
- air movement;
- radiation<sup>8–11</sup>.

In addition to these main factors, forecast, diseases and pests that emerge in relation to the weather incidents, alongside with air and noise pollution, oxygen in the atmosphere and amount of light affect individual comfort<sup>9,12</sup>. It is important for all these parameters to be evaluated in the studies of urban design, in order to provide comfort in terms of bioclimatic perspective. In this study, the Eski Kordon area at the centre of Canakkale has been studied in terms of climactic comfort. The aim of this study is to determine variations in the rates of the approximate heat, relative humility and wind, light alongside with radiation and noise in the areas where structural and vegetative equipment were used.

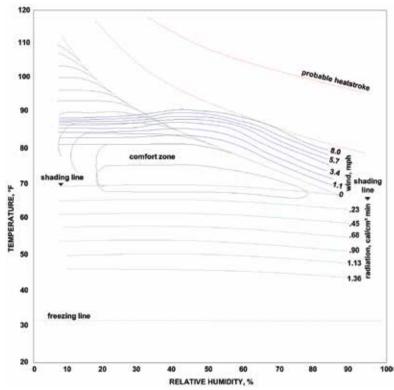


Fig. 2. Bioclimatic graph13

#### EXPERIMENTAL

The main material of this study comprises bioclimatic comfort parameters of the Eski Kordon area located at the centre of Canakkale. In order to conduct landscape design studies, an assessment took place at the Eski Kordon, as the appointed research area, for determining areas of structural and vegetative equipment which had different climate rates during the day.

In the research, whilst literature scan of the subject was done, photos were taken and maps of the related area were used. In the study landscape research method that relies on study, data collection, analysis, synthesis and assessment were conducted by studying the actual place. This study took place in the Eski Kordon on the date 23rd August 2015, in 5 different spots where the sun light starts to change at hours 07:00 am - 12:00 pm and 17:00 pm (Fig. 3).

During the measurement which took in the actual place; approximate heat, relative humility and wind rate, light alongside with the radiation and noise rate were determined by using measurement tools – a Trotec labelled Termohygrometer,

Microwave Radiometer, Digital Sound Level Meter, Luxmeter and Anemometer-1.5 m high from the ground. These data which have been obtained from the Eski Kordon were assessed in terms of bioclimatic comfort.



Fig. 3. Eski Kordon measurement points

## RESULTS

Canakkale strait as one of the most important waterway in the world runs along the city shore. Appointed study area, which comprises the Eski Kordon area is located at the centre, between Cimenlik Castle and Canakkale Sergeant Officer Club. The Eski Kordon identification card that contains the information related to the studied field is given in Fig. 4.

As it may be noted in Fig. 1, in green areas where there were trees with broad leaves, there was no sudden temperature increase from the early morning. However, it was observed that relative humidity rate which under the normal circumstances expected to be raised with the temperature increase, has fallen. Particularly during the measurement that took place at 17:00 pm, wind force in the firm ground increased however, in the green area, trees acting as a curtain decreased the wind force.

In firm grounds, light force alongside with radiation reached its peak in measurement especially during afternoon hours whereas these rates were kept in a reasonable level in green areas. In other areas noise rate increased due to observable traffic and busy pedestrians in the day and evening whereas, in the same hours it was measured as the normal rate in green areas.

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DAC	and Sign	Enough	IIY St					
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8	Kiosk		P	E avec a second				
URAL	Plastic object		and Use and Activities	Diversity	Not enough			
RUCT	Playground equipment	Not Enough	Land	Sustainability	Land mark, intensive use for pedestrian and vehi			
20	Bridge			The second second	and the second se	Imagene		
= -	Hard	2	Mainly Vegetation	Plane Tree		Pletenus spp		
Ground	Ground			Desert Fan Palm Beech		Washingtonia filifera Fegus sp.		
Bro				Ash		Prevenus spp.		
I V	Soft Ground	20%		Em		Ulmus app.		

Fig. 4. Eski Kordon identification card (Ref. 14)

The average temperature relative humidity, wind, photo period, lightening and the noise values that have been taken at the end of conducted measurements are given in Table 1.

			17:00	68.9	63.9	61.4	79.1	67.8
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	/e 2)		17:00	0.07	0.45	0.12	0.02	0.02
	Microwave (mW/cm <sup>2</sup> )		12:00	0.16	0.06	0.04	0.04	0.03
	Ψ.		07:00	0.15	0.04	0.03	0.02	0.02
	sity		17:00	999 x100	952 x10	999 x100	999 x100	999 x100
	Lux)	time (Lux)	12:00	198 999 999 x10 x100 x100	1120 x10	999 x100	999 x100	999 x100
	Ligh		07:00	198 x10	540 x10	411 x10	410 x10	1416 x10
	Ч	ti	17:00	6.0	3.0	4.5	6.5	8.0
	Air speed (m/s)	(3/11)	12:00	3.40	3.80 4.80	6.02	4.55	4.80
			07:00	3.56 3.40 6.0	3.80	3.94	3.86	3.42
	Relative humidity (% rH)		17:00	72.0 50.3 51	57	57	57.5	55
umeters			12:00	50.3	59.0	60.0	53.0	49.4
			07:00		66.1	72.5	71.1	75.9
fort par	ture		17:00	30	29	30.2	29	29
tic com	empera (°C)		12:00	31.3	28.2	28.0	30.0	32.3
sioclima	Air t		07:00	25.3 31.3	26.6 28.2	25.3 28.0	24.1 30.0	23.6 32.3
Table 1. Bioclimatic comfort parameters	Mea- Air temperatu surement (°C)	areas		-	7	ю	4	S

## CONCLUSIONS AND RECOMMENDATIONS

Urban texture becoming dense with each passing day coupled with the busy traffic which it brings and with the increase of firm grounds cause bioclimatic comfort to decrease. Equipment such as cement, asphalt, bricks, etc. which cover the city surface generate important increase in the bioclimatic comfort parameters. This equipment cannot absorb but reflect heat, light, radiation and noise and increase these rates. Besides they release the energy which they have absorbed in the morning, in long thermal wave radiations and heat waves during the night time. Green open areas and plants which they consist of that stuck in the urban texture contribute to the regulation of the climate. These areas not only increase visual landscape quality but also make important contributions to urban ecosystems by reducing thermal radiation via absorbing light, wind force and heat as well as by acting as a green curtain to reduce noise.

Although natural stone equipment have been used in the firm ground of the Eski Kordon area, the fact that these grounds were wide and excluded from green bands, they get cooler in the evening and suddenly hot in the day that it affects the other parameters of the bioclimatic comfort negatively.

Sudden and extreme changes have been observed in bioclimatic comfort parameters which were taken particularly in the morning and evening on firm grounds. On the other hand, it was noted that in the green areas these rates changed at a slower pace or kept stable.

Some of the measures that need to be taken in order to ensure visual quality as well as bioclimatic comfort in urban landscape designs that would take place in the Eski Kordon area in Canakkale are listed below:

• Instead of using artificial equipment in firm grounds, natural equipment should be preferred;

• Sustainable green areas should be generated in the general working area;

• Taking into account the wind potential that Canakkale has and for the reason that the study area is by the seashore and open to wind, green wind curtains should be generated;

• In green areas, preferred plant texture should be in medium texture with broad leaves in order to reduce light and radiation;

• Due to location of the study area green bands should be generated in order to prevent noise, particularly on the part where there is a vehicle road;

• Especially in selecting equipment that is used for firm ground, residence unit and fitting components dark colours should be avoided.

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Information



B.EN.A. – Balkan Environmental Association

# CONCLUSIONS

# 6th International Conference on '*Protection of Natural Resources* and Environmental Management: The Main Tools for Sustainability' – PRONASEM 2016

11-13 November, 2016, Romanian Academy, Bucharest, Romania

The 6th International Conference on '*Protection of Natural Resources and Environmental Management: The Main Tools for Sustainability*' PRONASEM 2016 was jointly organised by the Balkan Environmental Association (B.EN.A), the Romanian Academy and University Polytechnica of Bucharest (UPB), with supporting co-organisation of the European Organisation for Nuclear Research (CERN), and the National Authority for Scientific Research and Innovation (ANCSI) of Romania, in collaboration with the European Environmental Association (EEA) and the Black Sea Commission, Istanbul, Turkey. The conference was held at the Romanian Academy Library during 11–12 November, 2016, in Bucharest, Romania.

In the frame of the Official Opening, the talk addressed by Dr. Mariana Golumbeanu, on behalf of PRONASEM Organising Committee, emphasised the role of the late Prof. Dr. Fokion Vosniakos in promoting cooperation and collaboration within the B.EN.A. scientific family, thus the PRONASEM Conference and the Book of Abstracts were dedicated to his memory! Further speeches were addressed by Acad. Bogdan Simionescu, vice-president of the Romanian Academy, Prof. Emmanuel Tsesmelis, CERN, Dr. Caner Zanbak, vice-president of B.EN.A., and Emeritus Prof. Aurelia Meghea.

This high level scientific event brought together representatives of academia, research, graduate students, scientific organisations and economic operators concerned with environmental protection and sustainable development, providing for a framework for promoting new collaboration projects that will further enhance

the Balkan countries participation in European Union programmes and integration in the European Research Area.

The Conference hosted 156 registered attendees from a variety of disciplines, making 63 oral and 108 poster presentations. The attendants were from Albania, Bosnia and Herzegovina, Bulgaria, CERN-Switzerland, Croatia, Czech Republic, FYR Macedonia, Greece, Italy, Moldova, Montenegro, Serbia, Turkey and the United Kingdom, with a special attendance of a European Parliament member from Portugal. Additionally, a workshop entitled '*Sustainable Development and Challenge of e-innovation for a Green Society*', organised by the 'Simone Cesaretti' Foundation, was held as the last session of PRONASEM, with seven presentations on IT and green society interrelations.

The keynote speakers in the Plenary Section presented a SWOT analysis on Romanian natural resources, followed by a comprehensive presentation of CERN Institute – A Gateway to Science, Technology and Innovation, along with a brief discussion on the Responsible Care<sup>©</sup> initiative implementation of the chemical industry on a case in Turkey, while the final lecture illustrated relevant aspects of sustainable development in Republic of Moldova based on local fossil fuel resources.

#### ORAL SESSIONS

*Session 1*. Primary Natural Resources, Fossil Fuels and Renewable Energy: The following topics were covered in this session:

 application of geomatics to promote sustainable resources management and protection, especially in cross-border areas, modelling of hydrocarbon pollution risks sourced from the marine transportation activities in the Danube Delta Biosphere Reserve to assess the regional current conditions and evaluate potential threats;

 – evaluation of a novel and fast technique of hybrid linear ion-trap-orbitrap mass spectrometry as applied to monitor the seasonal distribution of pesticide pollutant load in natural waters in Greece;

– optimisation and economic analysis of solar-assisted heating systems based on solar radiation values within the Balkan region and evaluation of rare earth elements presence in the coal ash from the power plants in Serbia;

- CERN main activities were presented with emphasis on environmental awareness of scientific research laboratories.

Session 2. Secondary Natural Resources: Industrial Wastes and Recycling: Presentations focused on raising awareness and technological/administrative and social aspects of waste management with special emphasis on collection, recovery and recycling issues as applied to beneficiation of industrial and household waste within the context of Circular Economy. The specific topics discussed were on achieving sustainable development:

- setting environmental protection priorities with due emphasis on economic and social aspects;

- application of monitoring instruments and technologies, field and laboratory measurements, statistical data evaluation approaches;

- technical and legislation proposals for future tasks for the administrations and the scientific communities;

- industrial aspects of processing various secondary products while improving pollution prevention/elimination, elaborating on best practices, technological solutions/alternatives for different industries proposing application of more environmentally friendly methods and products;

- potential of biogas as a promising biofuel in reducing the greenhouse gasses while saving the fossil fuel resources.

*Session 3.* Biodiversity and Protected Areas (inland and marine): This session covered presentations from Portugal, Romania, Norway, Greece, Turkey, Bulgaria, United Kingdom, Italy and Czech Republic, emphasising on the following issues for protection and conservation of biodiversity and protected areas:

- the needs for powerful management tools, policy engagement and strengthened international dialogues, cooperation and dissemination/transfer of scientific knowledge and technological developments and review and support of regional and global policies;

- the role of FP7 projects (e.g. MareFrame, Perseus, SeasERA, REEFs), and also national programs (funded through the National Authority for Scientific Research and Innovation ANCSI) is crucial in the development of new tools and indicators supporting the coastal zone sustainability and Decision Support Framework for management of living and non-living resources;

- importance of improved awareness in regional communities and promotion of Excellence Concept to support sustainable economic growth.

The oral presentations dealt with modelling aspects of environmental issues; protected areas and sustainability of coastal environments; fish resources, microbial biodiversity and improvement of green tourism, while stimulating the participant interest to the various topics and leading to fruitful discussions.

*Session 4*. Environmental Monitoring and Impacts; Risk Assessment – Life Cycles Analysis: Specific presentations in this session included evaluation and discussions of site-specific impacts:

- on the role of phosphorous in eutrophication processes in Shkodra Lake, Albania;

- the radiological environmental impact of high-power accelerators at CERN and the mitigation measures employed;

 importance of water quality and sanitation along with air quality studies their management aspects as part of the sustainable development efforts through use of best available monitoring positioning including biomonitoring air quality using lichens;

– novel application of photovoltaic systems installed on ships on the trade routes in the Northwest Black Sea basin, their life cycle/cost-benefit analysis.

Session 5. Agro Environment – Agro Ecology – Food Quality and Safety: This Session included presentations on an innovative soil sampling method for increased sampling speed and volume of soil sample that can lead to improvement and importance of optimization of balanced fertilization for sustainable agriculture; identification of genetic resistance of wheat cultivars and its importance in agro-ecological applications; and a comprehensive review of anthropogenic channel morphology changes in development of agricultural lands.

A workshop entitled 'Sustainable Development and Challenge of E-innovation for a Green Society', organised by the 'Simone Cesaretti' Foundation, was held as the last session of PRONASEM, with seven presentations on IT and green society interrelations. The speakers emphasised that sustainable development has been in the center stage in all national, international and trans-national environmental policies during the last two decades. Sustainable development aims to improve quality of life for the communities along with economic development and taking into consideration of protection of the environment. EU is a pioneering community supporting environmental legislation, initiatives and projects on the issues, yet it is defined that the main tool to implement sustainable development is the use of *'E-innovation and ICTs'*, for the roadmap to a green society and a green European continent by 2050.

Poster sections held during the conference had seven main topics and included presentation of 110 posters of which majority of the authors were graduate students from 10 countries as listed below:

1. Primary Natural Resources (air, water/fresh-marine, soil), fossil fuels and minerals, renewable energy (solar, wind, geothermal energy, biofuels, etc.) -10 posters.

2. Secondary Natural Resources: industrial wastes and recycling - 8 posters.

3. Biodiversity and Protected Areas (inland and marine) – 27 posters.

4. Environmental Monitoring and Environmental Impact. Risk Assessment-Life Cycle Analysis – 19 posters.

5. Agro Environment – Agro Ecology – Food Quality and Safety – 16 posters.

6. Green Smart Cities – Green Planning. Tourism/Ecotourism – 13 posters.

7. Public Health and Environmental Medicine – 17 posters.

A total of 20 awards were presented to the authors of the posters which were evaluated by referees.

The PRONASEM Conference was concluded with a Special General Body Meeting of B.EN.A. members declaring a Conclusionary Statement summarising the proposed approaches for reorganisation of B.EN.A. in the coming term of the Executive Board.

13th November; 2016



# ERRATA

On page 958 of book 3, vol. **17** (2016), the number of the project in **Acknowl-edgements** should be GAJU 094/2016/Z instead of GAJU 063/2013/Z ('ENERGY CROPS GROWING – IMPACT ON GREENHOUSE GASES EMISSIONS' by J. BERNAS, J. MOUDRY Jr., Z. JELINKOVA, M. KOPECKY, P. KONVALINA, J. MOUDRY).

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The title page should include the title, authors and their affiliations, complete address of the author to whom correspondence should be sent and an Abstract.

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