GLIBENCLAMIDE Complexes (Complexes of Mg,Cr,Ni,Zn and Cd salts) Synthesis and characterization

Dr krishnasarma Pathy , Ch Siva subramanyam, N Laxman,
Shakti bioscience ltd vapi

ABSTRACT

Glibenclamide is the commonly used hypoglycemic agent in NIDDM. Metal complexes of glibenclamide have been synthesized by reaction with different metals such as magnesium, chromium, cobalt, nickel, zinc and cadmium in the form of their chlorides. These complexes were characterized by their physical characteristics, ^{1}H-NMR, IR and Atomic absorption studies.

Keywords: Antidiabetics, glibenclamide, transition metals, NMR, IR, Atomic absorption spectroscopy, magnesium, chromium, cobalt, nickel, zinc, cadmium.

INTRODUCTION

Glibenclamide (Danoil, Euglucon), 1-[4-[2-(chloro-2-methoxybenzamido)ethyl]-benzenesulphonyl]-3-cyclohexyl-urea, 5-chloro-N-[2-[4[[[(cyclohexyl(amino) carbonyl]-amino] sulfonyl] -phenyl]ethyl]-2-methoxy benzamide or 1-[[p-[-2-(5-chloro-o-anisami do)ethyl]phenyl]-sulphonyl-3-cyclohexylurea, a sulphonyl urea derivative (figure 1) having melting point 169-174^{0}C is a white or almost white crystalline odorless powder, practically without taste, insoluble in water, sparingly soluble in methylene chloride, slightly soluble in ethanol, methanol and insoluble in diethyl ether. It dissolves in dilute solutions of alkali hydroxides (Martindale, 1999; British Pharmacopoeia, 1998 and USP24 NF19, 2000).

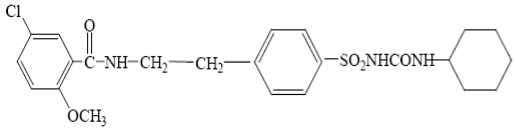

Fig. 1

Glibenclamide is a second-generation oral hypoglycemic agent which is more potent than those of first group (Lebovitz and Feinglos, 1983) and is used to assist in the control of mild to moderately severe type II diabetes mellitus (adult, maturity-onset) that does not require insulin, but that can be adequately controlled by diet alone. It is a drug of choice for initiating treatment in noninsulin-dependent diabetes when diet and weight control fails. It stimulates the secretion and enhances the utilization of insulin by appropriate tissues (Long, 1990). It is 90-100% absorbed in gastrointestinal tract (Rupp *et al*., 1972; Schmidt and Petrides, 1969 and Borchet *et al*., 1976). The highest concentrations of the drug were found in liver and kidney while lower concentrations were detected in other tissues. Glibenclamide shows no significant enterohepatic cycling in man (Christ *et al*., 1969).

A number of drug interactions on glibenclamide have been reported many of which are potentially hazardous (Seltzer, 1979). Glibenclamide binds to plasma proteins by non-ionic forces because of its large non-polar chemical group (Brown and Crooks, 1976), consequently, bound glibenclamide is less susceptible to displacement by other drugs. When given with captopril and enalapril enhance the hypoglycemic effect (Rett *et al*., 1988 and Arauz-Pacheco *et al*., 1990). Rifampicin reduces the glibenclamide serum level and hypoglycemic activity (Self *et al*., 1980 and 1989), while verapamil resulted in higher level of glibenclamide by interfering metabolism of glibenclamide to undo its insulin stimulating property (Semple *et al*., 1986; Bauomy and Hassan, 1987; Ahmad, 1995, Whitecroft *et al*., 1990 and Andersson and Rojdmark, 1981). During interaction between antibiotics, erythromycin (Avery, 1973), chloramphenicol succinate and cefaloridine with glibenclamide, these drugs increased the hypoglycemic effects (Atta *et al*., 1983). It is also found to influence the metabolism of xenobiotics (Stroev and Belkina, 1989). Bioavailability of glibenclamide is influenced by antacids (Zuccaro *et al*., 1989; Neuvonen and Kivisto, 1991 and Kivisto and Neuvonen, 1991). The hypoglycemic activity of glibenclamide remained same when given with cimetidine (Shah *et al*., 1984 and 1985), while in another study plasma glucose concentrations were higher when glibenclamide

Table 1: Physical characteristics of complexes of glibenclamide with essential and trace elements

S. No.	Complex	Color	State	M.P °C	Yield (%)
1	Glibenclamide magnesium	White	Crystalline	158	50.21
2	Glibenclamide chromium	Green	Crystalline	157	49.52
3	Glibenclamide cobalt	Light blue	Crystalline	159	37.30
4	Glibenclamide nickel	White	Crystalline	156	44.32
5	Glibenclamide zinc	White	Crystalline	158	54.28
6	Glibenclamide cadmium	White	Crystalline	156	43.59

Table 2: IR absorption bands of glibenclamide metal complexes

Compound	Main IR absorptions in cm^{-1}
Glibenclamide (reference)	440 sm, 520 sm, 540 m, 570 m, 610 m, 640 sm, 680 m, 820m, 880 -900 db, m, 1010 - 1030 db, m, 1090 sm, 1120 sm, 1160 s, 1240 - 1270 db,m, 1300 sm, 1340 s, 1450 s, 1520 s, 1615 s, 1700 s, 2550-2600 db, 2900 s, 3300–3370 db,s.
Glibenclamide-Mg complex	440 sm, 520 sm, 540 s, 570 s, 610 s, 640 m, 680 s, 820 s, 880 - 900 db, s, 1010 - 1025 db ,s, 1090 m, 1120 m, 1150 s, 1240 -1270 db, sm, 1300 m, 1340 s, 1520 s, 1610 s, 1730 s, 2850 -2900 db, m, 3300 - 3350 db, m.
Glibenclamide-Cr complex	440 sm, 540 s, 570 s, 610 s, 640 sm, 680 m, 820 m, 880 sm, 900 m, 1010 - 1025 db, m, 1090 sm, 1120 sm, 1160 s, 1240m, 1270 sm, 1300 sm, 1340 m, 1520 m, 1620 s, 1730 sm, 2800-2900 db, m, 3100 sm, 3300–3340 db,m.
Glibenclamide-Co complex	440 sm, 510 sm, 540 s, 570 s, 610 s, 640 sm, 680 s, 820 s, 880 m, 900 s, 1010 - 1020 db ,s, 1090 -1120 db,m, 1150 s, 1240 m, 1270 m, 1300 sm, 1340 s, 1520 s, 1620 s, 1725 db, s, 2800 -2900 db, m, 3300 - 3360 db, sm.
Glibenclamide-Ni complex	440 sm, 510 sm, 540 s, 570 s, 610 s, 640 sm, 680 s, 810 s, 880 sm, 900 m, 1010 - 1025 db,m, 1090 m, 1120 m, 1150s, 1240 s, 1270 m, 1300 sm, 1340 s, 1520 s, 1620 s, 1720 s, 2800 -2910 db,m, 3300 - 3350 db, s.
Glibenclamide-Zn complex	440 sm, 510 sm, 540 s, 570 s, 610 s, 640 sm, 680 m, 820 s, 880 sm, 900 s, 1010 - 1015 db ,m, 1090 sm, 1120 m, 1150s, 1240 m, 1270 m, 1300 sm, 1340 s, 1520 s, 1620 s, 1750 s, 2850-2910 db, m, 3300 - 3340 db, m.
Glibenclamide-Cd complex	440 sm, 520 sm, 540 s, 570 s, 610 s, 640 sm, 680 m, 820 s, 880 sm, 900 s, 1010 - 1015 db ,m, 1090 sm, 1120 m, 1150 s, 1240 m, 1270 sm, 1300 sm, 1340 s, 1520 s, 1620 s, 1730 db, s, 2850-2910 db,m, 3300 - 3350 db, m.

was administered with cimetidine or ranitidine (Kubacka *et al*., 1987).

In present study glibenclamide was reacted with various essential and trace element salts of magnesium, calcium, chromium, manganese, iron, cobalt, nickel, copper, zinc and cadmium in order to ascertain their *in vitro* interaction, as these above or alongwith multi-vitamins preparations may possibly be co-administered with glibenclamide.

MATERIALS

Glibenclamide standard was a gift from M/s Ali Gohar Pharmaceuticals (Pvt) Ltd. Solvents and the metal salts used were of the analytical grade (E. Merck). Melting points were determined on a Buchi 531 m.p. apparatus and are uncorrected. pH values were determined on Gallenkamp pH stick meter. The ^{1}H-NMR spectra were recorded on a Bruker AMX 500 MHz spectrometer in $CDCl_3$ using TMS as an internal standard. IR studies were carried out by Shimadzu Model IR 470 infrared spectrophotometer. Atomic absorption studies were carried out by Pye-Unicam atomic absorption spectrometer.

Reaction between glibenclamide and essential and trace elements

Methanolic solutions of metal chlorides (1mM) were individually added to methanolic solutions of glibenclamide

Table 3: Proton NMR of glibenclamide metal complexes

Compound	δ and Multiplicity
Glibenclamide (reference)	1.10-1.43 CH_2, 1.71-1.98 CH_2, 3.00-3.14 NH, 3.76 O-CH, 6.42 aromatic, 6.84-6.87 aromatic, 7.30-7.53 aromatic, 7.82-7.93 aromatic, 7.97-7.99 aromatic, 8.14-8.15 aromatic.
Glibenclamide-Mg complex	1.20-1.29 CH_2, 1.60-1.66 CH_2, 1.80-2.89 CH_2, 3.01-3.02 NH, 3.69 NHC-O-Mg, 3.77 O-CH_3, 6.40 aromatic, 7.34 aromatic, 7.40-7.59 aromatic, 7.81-7.84 aromatic, 8.14-8.15 aromatic.
Glibenclamide-Cr complex	1.15-1.57 CH_2, 1.65-1.80 CH_2, 3.00-3.04 NH, 3.70 NHC-O-Cr, 3.77 O-CH_3, 6.44 aromatic, 6.85-6.89 aromatic, 7.25-7.35 aromatic, 7.38-7.39 aromatic, 7.82-7.84 aromatic, 8.14-8.15 aromatic.
Glibenclamide-Co complex	1.12-1.41 CH_2, 1.65-1.69 CH_2, 1.80-1.85 CH_2, 3.00-3.05 NH, 3.71 NHC-O-Co, 3.75 O-CH_3, 6.43 aromatic, 6.85-6.88 aromatic, 7.25 aromatic, 7.36-7.43 aromatic, 7.82-7.85 aromatic, 8.14-8.15 aromatic.
Glibenclamide-Ni complex	1.19-1.34 CH_2, 1.57-1.69 CH_2, 1.80-1.84 CH_2, 3.00-3.04 NH, 3.73 NHC-O-Ni, 3.78 O-CH_3, 6.46 aromatic, 6.85-6.88 aromatic, 7.25 aromatic, 7.35-7.40 aromatic, 7.82-7.85 aromatic, 8.14-8.15 aromatic.
Glibenclamide-Zn complex	1.12-1.34 CH_2, 1.57 CH_2, 1.66-1.69 CH_2, 3.00-3.02 NH, 3.67 NHC-O-Zn, 3.79 O-CH_3, 6.43 aromatic, 6.85-6.89 aromatic, 7.25-7.59 aromatic, 7.81-7.85 aromatic, 8.14-8.15 aromatic.
Glibenclamide-Cd complex	1.11-1.55 CH_2, 1.64-1.68 CH_2, 1.79-1.83 CH_2, 3.02-3.04 NH, 3.66 NHC-O-Cd, 3.78 O-CH_3, 6.41 aromatic, 6.85-6.88 aromatic, 7.25 aromatic, 7.35-7.39 aromatic, 7.82-7.96 aromatic, 8.14-8.15 aromatic.

(1 mM; 0.424 gm) and refluxed for 2-3 hours. The colored solutions were then filtered and left for crystallization at room temperature for 24 hours. Crystals of different colors for different metal complexes were obtained which were filtered, washed, dried and their melting points determined. The resulting complexes so formed were characterized by their physical characteristics, IR, H^1-NMR and AA studies.

RESULTS AND DISCUSSION

Glibenclamide was treated with different metal salts of essential and trace elements in equimolar ratio to afford colored crystalline complexes. It was observed that glibenclamide did not form complexes with calcium, manganese, iron (Fe^{++} or Fe^{+++}) and copper salts, which was proved by no change in melting points or IR absorption spectrum. Hence, no further analytical studies for these were carried out. The physical characteristics of complexes are given table 1. The complexes formed with magnesium, chromium, cobalt, nickel, zinc and cadmium were diamagnetic and nonionic in nitrobenzene.

Infrared absorption studies

The infrared spectrum of glibenclamide and its metal complexes were recorded as KBr disc method on a Shimadzu Model IR 470 infrared spectrophotometer. The major absorption bands for the infrared frequencies and the corresponding assignments are listed in table 2.

Glibenclamide showed a prominent IR absorption band in the region of 3300-3370 cm^{-1} due to urea NH stretching, a very sharp peak observed at 2900 cm^{-1} due to –CH stretching, at 1710 cm^{-1} the absorption was due to C=O, a strong absorption band at 1520 cm^{-1} due to C=C, at 1340 cm^{-1} due to C-O and at 1160 cm^{-1} the peak of SO_2 was observed. The absorption peak of chlorine occurred at 540 cm^{-1}. The carbonyl band was observed at 1710 cm^{-1} in reference standard, but in transition metal complexes this stretching frequency increased towards 1720 – 1750 cm^{-1} region. For magnesium complex the band was observed at 1730 cm^{-1} in the form of sharp peak whereas in case of chromium complex it occurred at 1730 cm^{-1} as a weak band. In case of cobalt the carbonyl peak appeared at 1725 cm^{-1} as a sharp-doublet. Nickel complex showed a sharp band in the region of 1720 cm^{-1}, whereas in zinc complex it was observed at 1750 cm^{-1} as a sharp band. In case of cadmium complex the carbonyl peak appeared at 1730 cm^{-1} in the form of a sharp-doublet band.

Other substitutions

The absorption peaks between 600-800 cm^{-1} were due to the deformation of adjacent hydrogen, in metal complexes this was also present with slight changes, which were negligible. In glibenclamide, the urea NH stretching appears at 3300-3370 cm^{-1} as a sharp doublet in case of magnesium and cadmium complexes it appeared in the region of 3300-3350 cm^{-1} in the form of a medium doublet, while in case of nickel it was a sharp doublet and in the same range. Chromium complex showed medium doublet in the range of 3300-3340 cm^{-1} whereas in cobalt complex it appeared in the range 3300-3360 cm^{-1} as small doublet. In case of zinc complex the urea NH stretching existed in the range of 3300-3340 cm^{-1} as a medium doublet.

NMR studies

NMR data of all the complexes are summarized in table 3 and their proposed structures are given in figure 2. The ^{1}H–NMR of glibenclamide metal complex displayed nitrogen – metal signals in the range of δ 3.66 – 3.73 due to the deshielding of N-bearing protons. The ^{1}H– NMR of glibenclamide-Mg(II) complex displayed nitrogen–Mg signal at δ 3.69, glibenclamide-Cr(II) complex at δ 3.70, glibenclamide-Co(II) complex at δ 3.71, glibenclamide-Ni(II) complex at δ 3.73, glibenclamide-Zn(II) complex at δ 3.67, glibenclamide-Cd(II) complex at δ 3.66 due to deshielding of N-. bearing protons. In all these cases both the nitrogens of the amide took part in bonding to metals (fig. 2).

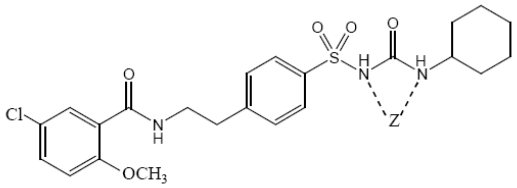

Fig. 2: Glibenclamide metal complex; Z = metal

We recently reported the synthesis and characterization of gliclazide metal complexes with magnesium, calcium, chromium, manganese, iron, nickel, copper, zinc and cadmium salts (Arayne *et al* 2005). The bonding site of these metals was also amide nitrogen of gliclazide and comparable ^{1}H-NMR spectral properties were observed. In those case ^{1}H– NMR of gliclazide-metal complex displayed signals in the range of δ 3.29 – 3.72 due to the deshielding of N-bearing protons.

AA studies

This analysis was carried out by direct method which gave total metal content. A number of reference standard solutions of each metal were also prepared having various concentration ranges. Absorbance of these solutions were measured at the specific wavelength of each metal using background correction technique (Smith *et al*., 1982). A graph was plotted between absorbance and concentration of each metal solution, which showed a straight line in each case. The concentrations of unknown solutions were calculated from the absorbance of unknown solutions by using the standard values. Results of analysis are given in table 4.

Table 4: Estimation of metals by AA spectroscopy

Compound	Metal calculated	Metal found
Glibenclamide-Mg complex	4.69	4.70±0.001
Glibenclamide-Cr complex	9.53	9.93±0.012
Glibenclamide-Co complex	10.67	10.39±0.019
Glibenclamide-Ni complex	10.63	11.05
Glibenclamide-Zn complex	11.70	12.15±0.003
Glibenclamide-Cd complex	18.55	18.64±0.010

CONCLUSION

The differences in melting point of all of these complexes as compared to glibenclamide suggested that a new product was formed. The shifts of peaks in IR region as well as new signals around at δ 3 due to the deshielding of N-bearing protons in ^{1}H-NMR further confirmed the drug metal complexation. The final proof of metal incorporation in glibenclamide was obtained by the estimation of the metals from these complexes by atomic absorption spectroscopy.

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