Impact of magnesium-aspartate hydrochloride on glucoregulatory system of two avian species

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Chronic administration of magnesium aspartate hydrochloride (Mg-Asp.HCl) induced hyperglycemia, hypoinsulinemia and hypercalcemia in common myna. But there were no changes in blood sugar and plasma insulin levels in the magnesium-fed pigeon. Plasma magnesium markedly increased in both the avian species, but plasma calcium level decreased significantly in pigeon, unlike common myna. Magnesium had no effect on pancreatic glutathione concentration in both pigeon and common myna. Concentration of magnesium and/or food habit may be the probable two factor(s) responsible for the different glycemic responses in pigeon and common myna.

THE interrelationship between magnesium and carbohydrate metabolism has regained considerable interest over the last few years. Insulin secretion requires magnesium. Magnesium deficiency results in impaired insulin secretion, while magnesium replacement restores insulin secretion. Experimental magnesium deficiency reduces the tissue sensitivity to insulin[1]. Paolisso and Ravussin[2] reported that intracellular magnesium acts as a cofactor for numerous enzymes involved in carbohydrate metabolism. Despite the reports in mammals, except for chicken[3], very little is known about the role of magnesium on the glucoregulatory mechanism of birds. In rats, depending upon the dose administered, magnesium-aspartate hydrochloride (Mg-Asp.HCl) exhibits Ca-antagonistic effect and inhibits the release of stress hormones[4,5]. In chickens, changes in plasma level of magnesium have been reported to cause alterations of the rate of insulin release[3].

The paper ascertains the role of Mg-Asp.HCl on gluco-regulatory mechanism with special reference to insulin release in two avian species belonging to different phylogenetic and nutritional status, and aims to establish, whether the response is related to the latter factor. Mg-Asp.HCl was used, since it is readily absorbed through the intestine[5].

Adult specimens of pigeon (*Columba livia*) and common myna (*Acridotheres tristis*) were acclimatized to laboratory condition for a week. The animals were starved 18 h prior to experimentation, though water was provided *ad libitum*. Mg-Asp.HCl was orally administered to pigeon (600 mg/bird/day) and common myna (300 mg/bird/day) in the morning at 10 a.m. for six consecutive days. On the 7th day, prior to autopsy, blood was withdrawn from the wing vein for biochemical assays. Birds were autopsied by cervical dislocation. The pancreatic tissues were dissected out and fixed in chrome-alum-Bouin's fixative[6] for routine microtomy and 5 μm thick paraffin sections were stained using Epple's[7] aldehyde fuchsin trichrome method. Using arsenomolybdate colour reagent, blood sugar was estimated by Nelson and Somogyi's method. Plasma insulin was measured by a direct radioimmunoassay with protocol and radiochemicals supplied by Board of Radiation and Isotope Technology, Mumbai, India. The radioactivity was measured in LKB-γ counter. Calcium was measured by using methylthymol blue method of Glindler and King and colour was measured in Perkin Elmer spectrophotometer (Model 550 S, USA). Magnesium was determined by the fluorometric method of Schachter and fluorescence was measured by Fluorescence Spectrophotometer (Hitachi, Model 650, Japan). Splenic lobe tissues of pancreas were collected and glutathione was estimated using spectrofluorometric method of Hissin and Hilf[8]. The statistical analysis was carried out by Student's *t*-test.

The islets of Langerhans are distributed more or less uniformly throughout the entire pancreas. Islets are composed of A, B and D cells. A cells are large with well marked outlines. The nuclei are round and oval with prominent nucleoli. The B cells consist of deep purple aldehyde fuchsin positive granules. The D cells are mostly spindle-shaped and are smaller than A and B cells. The D cell nuclei fill up the entire cell surface and possess light green positive cytoplasm. But no significant changes were observed in the islet cytology under light microscope after chronic treatment of oral Mg-Asp.HCl, both in pigeon and common myna.

A marked increase in blood sugar level and decrease in plasma insulin concentration were noticed in the magnesium-treated common myna. Blood sugar and plasma insulin levels remained unchanged in magnesium-fed pigeon. Plasma magnesium level significantly increased in both the experimental birds. Plasma calcium level significantly decreased in pigeon whereas in common myna its concentration showed a marked elevation. Pancreatic glutathione was not altered after treatment in both pigeon and common myna (Table 1).

An earlier report showed that disturbances in Ca^{2+}/Mg^{2+} ratio alter the B cell responsiveness[3]. It is known in mammals that the intracellular calcium is essential for insulin release, while magnesium inhibits Ca^{2+} uptake[5]. Thus the increase or decrease of plasma calcium level is expected to increase or decrease the insulin release. In the present investigation, chronic treatment of Mg-Asp.HCl produced hyperglycemia, hypoinsulinemia and hypercalcemia in common myna. The

Table 1. Changes in blood sugar, plasma insulin, plasma calcium, magnesium and pancreatic glutathione after chronic treatment (6 days) with Mg-Asp.HCl in pigeon (*Columba livia*) and common myna (*Acridotheres tristis*)

Species	Dose	Group	Blood sugar (mg %)	Insulin (μU/ml)	Calcium (mM/l)	Magnesium (mM/l)	Glutathione (mM/g)
Columba livia	600 mg/ bird/day	Control	185.14 ± 9.24 (7)*	9.06 ± 0.54 (7)	3.27 ± 0.13 (7)	0.67 ± 0.06 (7)	8.08 ± 1.03 (7)
		Treatment	192.85 ± 6.62 (8)	8.88 ± 0.52 (8)	2.60 ± 0.20 (9)	1.24 ± 0.09 (7)	7.94 ± 2.11 (7)
			NS	NS	$p < 0.005$	$p < 0.001$	NS
Acridotheres tristis	300 mg/ bird/day	Control	200.21 ± 11.44 (7)	27.18 ± 2.92 (7)	3.19 ± 0.14 (7)	0.36 ± 0.03 (7)	9.64 ± 1.42 (7)
		Treatment	250.31 ± 18.03 (7)	15.52 ± 0.84 (7)	4.27 ± 0.13 (7)	0.40 ± 0.0009 (7)	10.45 ± 0.94 (7)
			$p < 0.05$	$p < 0.005$	$p < 0.001$	$p < 0.001$	NS

*Mean ± SE; NS = Not significant.
Figures in parenthesis represent the number of birds.

earlier report had shown that both Ca^{2+} and Mg^{2+} ions compete for a common cationic carrier system located in the B cell plasma membrane[9]. Hence, in the common myna, magnesium probably inhibits glucose-induced calcium uptake and subsequent insulin release by the B cell. Similar findings are also reported in mammals[10]. Our finding is supported by the observation of Hazelwood[11] in chicken where absence of magnesium in the perfusate markedly increases insulin output. In contrast to common myna, pigeon exhibited no significant changes in blood sugar and plasma insulin levels after chronic treatment of magnesium. Plasma level also decreased significantly in pigeon as compared to common myna. Hence, our results indicate that pigeon deviates from common myna and chicken[11], in its glucoregulatory mechanism after chronic treatment with magnesium. Classen *et al.*[12] pointed out that concentration of Mg-Asp.HCl is very important in exhibiting its Ca-antagonistic action. Thus, it is quite possible that the magnesium concentration was not optimum to cause Ca-antagonistic action and in turn failed to alter insulin release in pigeon, unlike in common myna. The differential findings between the two avian species could be due to the different nutritional status of pigeon and common myna which support the earlier work of Bentley[13]. Furthermore, increased level of plasma magnesium has no effect on pancreatic glutathione content in the two avian species studied.

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