EFFECT OF Mn^{++} ON BLOOD SUGAR LEVEL IN RATS

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Summary : Mn^{++} produced hypoglycemia in normal rats. The effect was dose-dependent. However, Mn^{++} did not produce hypoglycemia in alloxan–treated rats.

There was a rise in liver glycogen following Mn^{++} treatment which was correlated with fall in blood sugar at 1½ and 3 hr. There was also increase in the glucose uptake by rat diaphragm upto 47% between 2 and 3 hr.

The results suggest that functioning, intact pancreatic beta–cells are necessary for the action of Mn^{++} and that Mn^{++} may have a peripheral effect on glucose entry into the cells.

Key words : manganese hypoglycemia diabetes

INTRODUCTION

Everson and Shrader (3) have reported the impairment of glucose tolerance and utilisation in guinea pigs deficient in Mn^{++}. This was reversed by feeding a diet containing adequate Mn^{++}. In their earlier studies (4), the effect of feeding a diet low in Mn^{++} i.e. less than 3 p.p.m. to guinea pigs, was associated with absence or decrease in the size of the pancreas in neonatal animals. Decreased blood and hepatic manganese has been reported in hyperglycemic pancreatectomised dogs and diabetic human subjects. In South Africa, Rubinstein and associates (7) have observed that extracts of Lucerne in alfalfa or Medicago sativa containing high concentration of Mn^{++} i.e. 45.5 *mg/kg* produced dramatic hypoglycemic response in one patient. However, in another study (1) in normal control subjects and juvenile and obese adult onset diabetes even with large doses of Mn^{++} a hypoglycemic effect was never observed. These diverse reports in the literature prompted us to investigate the hypoglycemic effect of Mn^{++} in the experimental animals and if possible its mechanism of action.

MATERIAL AND METHODS

Unless otherwise specified, Haffkine strain rats of either sex weighing 100-120 *g* were used throughout the study. Six rats were included in each group.

Bloods sugar levels :

Blood was collected by cardiac puncture Blood sugar was estimated by the method of Nelson and Somogyi (6).

To induce diabetes, alloxan was administered in a dose of 100 *mg kg* (ip) along with ascorbic acid (10 *mg/rat*) orally for two consecutive days. Blood sugar estimations were performed 72 hr after the second dose of alloxan.

Mn^{++} was used as $MacI_2$ dissolved in normal saline. It was administered in the doses of 0.25, 0.5 and 1.0 *mg/kg* (ip).

Liver glycogen content :

Glycogen content of the liver was studied in control rats and rats given Mn^{++} treatment by the method of Montgomery (5).

Glucose uptake by rat diaphragm :

Haffkine strain rats weighing 60-80 g were used. These rats were fasted for 24 hr before use. Glucose uptake by the rat diaphragm was studied according to Bernstein and Post (2). Paired hemidiaphragms were preincubated for 90 and 180 min at 37°C in Krebs-Ringer solution containing 200 *mg*% glucose with and without Mn^{++}. Glucose was analysed from the perfusate by Nelson's procedure. Glucose uptake was calculated as the difference between the initial and final glucose content of the incubation medium.

RESULTS

Mn^{++} produced hypoglycemia in normal rats. The effect was dose dependent (Table I). However, Mn^{++} did not produce hypoglycemia in alloxan-treated rats (Table II).

There was a significant ($P<0.001$) rise in liver glycogen from 3 to 6 *mg*% following Mn^{++} treatment at $1\frac{1}{2}$ hr and 5.0 *mg*% at 3 hr. There was also an increase in the glucose uptake by rat diaphragm following Mn^{++} pretreatment (Table III).

TABLE I : Effect of Mn^{++} on blood sugar levels.

Dose mg/kg (ip)	*Mean blood sugar level (mg% ± S.E.M.)*		
	Fasting	*After Mn^{++}*	
		1½ hr	*3 hr*
0.25	76 ± 1.6	55 ± 2.5**	44 ± 2.7*
0.5	80 ± 2.0	56 ± 2.5**	41.5 ± 2.5*
1.0	80 ± 2.0	46 ± 3.0*	27 ± 2.0*

There were 6 rats per group.

Value differs sipnificantly from the corresponding control.

* $P < 0.001$
** $P < 0.01$

TABLE II : Effect of Mn^{++} on blood sugar level in alloxan–diabetic rats.

	Blood sugar, mg% (Mean ± S.E.M.)
Normal rats	113 ± 2.4
Diabetic rats	245 ± 6.8*
Diabetic rats + $MnCl_2$ 1 *mg/kg* (ip)	240 ± 4.3

There were 6 rats per group.

* Alloxan effect was statistically significant ($P < 0.01$).

TABLE III : Glucose uptake by diaphragm obtained from control and Mn^{++} treated rats.

% glucose uptake	*Time (minutes)*									
	0	5	10	20	40	60	90	120	150	180
Untreated rats	0	2	4	10	12	12	12.5	12	12.6	12.6
Treated rats ($MnCl_2$, 0.5 *mg/kg* (ip), ½ hr before)	0	2	5	16	20	22	25	47	47	38

NOTE: Values of the glucose uptake at 90, 120, 150 and 180 min are considerably higher though the statistical significance is not evaluated; the values are means of three independent experiments.

DISCUSSION

Mn^{++} produced a well defined hypoglycemia in normal rats in the doses uses. However, it did not have any hypoglycemic effect in the alloxanized diabetic rats. This indicates that functioning, intact pancreatic beta-cells are necessary for the action of Mn^{++}. In this respect, the action of Mn^{++} seems to be similar to that of sulphonylureas. The results also indicate that (i) the rise in the liver glycogen following Mn^{++} may be due to insulin release which in turn stimulates glycogen synthetase (conversion of glucose to glycogen) (ii) that Mn^{++} promotes cellular transport of glucose in rat isolated diaphragm.

Thus, Mn^{++} produced significant hypoglycemia, but failed to do so in alloxan-treated rats, suggesting that functioning beta-cells are essential for its action. It is, however, not clear why Mn^{++} did not have any effect in alloxan-diabetic rats, although Mn^{++} was found to promote cellular trasport of glucose in rat isolated diaphragm.

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