Phenothiazine-Induced Hyperglycemia: Relation to CNS and Adrenal Effects[1] (35794)

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An increase in blood glucose levels after the administration of chlorpromazine has been observed in several animal species including man (1–5). Other phenothiazines have been shown to exhibit a hyperglycemic response when administered to rats (6). Neither a depression of the motor activity nor a reduced body temperature are decisive factors in the origin of the chlorpromazine-induced hyperglycemia (7).

Phenothiazine derivatives have been shown to release catecholamines from adrenal glands *in vitro* (8). The removal of the adrenal glands or the adrenal medulla from rats prevents the hyperglycemic action of chlorpromazine (6). These studies suggest that the primary mechanism of phenothiazine-induced hyperglycemia is through the release of epinephrine from the adrenal glands.

Four phenothiazines, with varying CNS activity (9), were compared for their ability to induce hyperglycemia, to produce CNS depression and to release catecholamines from isolated adrenal glands. These studies were done to test the hypothesis that catecholamine release is directly related to the hyperglycemia produced by various phenothiazine compounds. It was also of interest to determine whether the degree of CNS depression produced by these compounds was correlated with the degree of hyperglycemia associated with their use.

Materials and Methods. Animals. Male albino mice, weighing 20–24 g (Laboratory Supply Co., Indianapolis), were used in the blood glucose and CNS depression studies. Prior to experimentation the mice were housed in groups of 20 with free access to food and water. In some experiments, bilateral adrenalectomy was performed 5 days prior to drug treatment and the animals were maintained on a regular diet with 0.9% sodium chloride in the drinking water.

Determination of blood glucose. The animals were sacrificed by decapitation. Blood was collected in beakers wetted with oxalate, and then assayed for glucose by the glucose oxidase method.[3]

Evaluation of CNS depression. Depression of spontaneous motor activity was determined for groups of 5 mice using circular photocell activity cages (Woodward Research Corp., Herndon, Va.).

Perfusion of isolated adrenal glands. Isolated bovine adrenals were used to obtain a measure of the catecholamine releasing abilities of CPZ and its analogs, since it was not possible to use mouse adrenals for this purpose. Fresh bovine adrenal glands were obtained at a local abbatoir and placed on ice during transport. About 45–60 min postmortem and glands were cannulated through the adrenal vein and perfused in a retrograde manner with aerated Locke's solution (10) at 37°. Flow rates were maintained at 5 ml/min using a multichannel metering pump (Harvard Apparatus Co.). A perfusion period of 45 min was allowed to elapse prior to the addition of the drugs, dissolved in Locke's solution (10 ml). The number of drug perfusions per gland was limited to three to avoid depletion of catecholamine stores and a 15-min interval was allowed between drug stimulations. The 2-min perfusate fraction prior to drug stimulation and the

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[3] Glucostat, Worthington Biochemical Corp., Freehold, New Jersey.

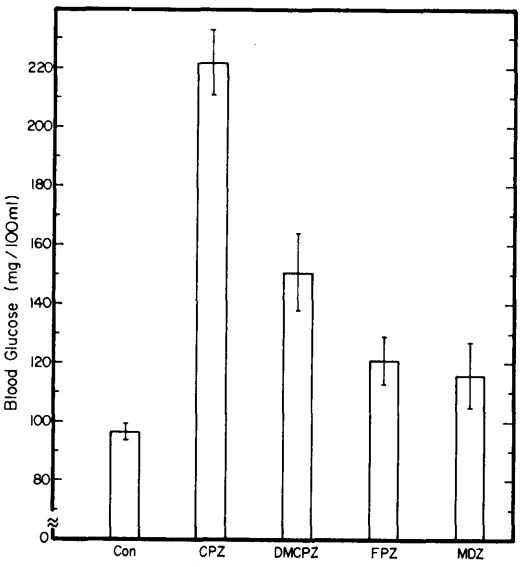

FIG. 1. Effect of various phenothiazines (3 mg/kg, ip) on blood glucose in mice: Each bar represents the mean blood glucose levels of 10 mice determined 1 hr after drug treatment. Con = saline control; CPZ = chlorpromazine; DMCPZ = mono-N-demethyl chlorpromazine; FPZ = fluphenazine; MDZ = methdilazine.

2-min perfusate fraction immediately following drug stimulation were collected. These perfusates from bovine adrenals were analyzed for total catecholamines by the colorimetric method of VonEuler and Hamberg (11). Drug-induced catecholamine release was determined by subtracting "resting catecholamine release" obtained in the 2-min period just prior to drug stimulation.

Drugs. Chlorpromazine hydrochloride (CPZ) and mono-*N*-demethylchlorpromazine hydrochloride (DMCPZ) were supplied by Smith, Kline and French Laboratoreis. Fluphenazine dihydrochloride (FPZ) was donated by E. R. Squibb & Co. and methdilazine hydrochloride (MDZ) by Mead Johnson Laboratories. Drug solutions were prepared fresh daily. Volumes of 10 ml/kg of the drug solutions were administered intraperitoneally to mice.

Results. Phenothiazine-induced elevation of blood glucose. Drugs were administered in a single dose of 3 mg/kg ip; and blood glucose levels were determined 1 hr after treatment (Fig. 1). Elevation of blood glucose produced by CPZ was significantly greater than that produced by DMCPZ, FPZ, or MDZ ($p<.01$). Among the 4 phenothiazines, FPZ and MDZ were the least effective in raising blood sugar.

When the same experiment was carried out in adrenalectomized mice using the same doses (Fig. 2), only DMCPZ caused a significant ($p<.01$) elevation of blood glucose when compared to adrenalectomized control animals (143 vs 104 mg/100 ml).

Depression of motor activity. The ability of each phenothiazine to depress spontaneous motor activity in mice when administered in a single dose of 3 mg/kg ip is shown in Fig. 3. All phenothiazines demonstrated significant ($p<.01$) CNS depressant activity using

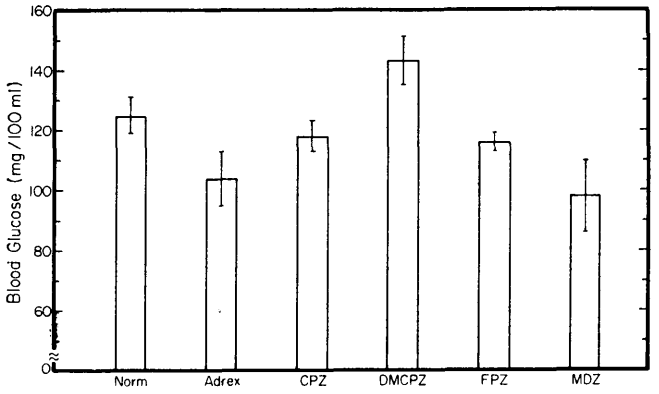

FIG. 2. Effect of various phenothiazines (3 mg/kg, ip) on blood glucose in adrenalectomized mice 1 hr after drug treatment: Each bar represents blood glucose levels in 10 mice. Norm = saline control; Adrex = adrenalectomized controls; other abbreviations are the same as for Fig. 1.

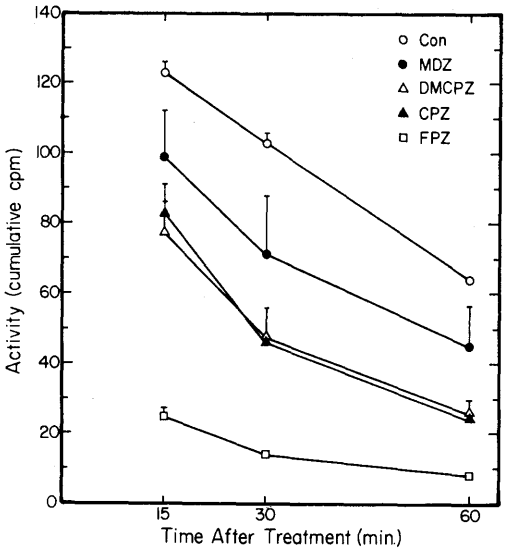

FIG. 3. Depression of spontaneous motor activity in mice by various phenothiazines (3 mg/kg, ip): Each point is the mean of the activity determined in 3 experiments each employing 5 mice in circular photocell activity cages. The cpm (counts per minute) are cumulative scores starting from the time the animals were introduced into the activity cages. Abbreviations used are the same as for Fig. 1.

Newman-Keuls method for testing difference between ordered pairs of treatments. FPZ was the best depressant tested, and was significantly more effective than CPZ ($p<.05$), DMCPZ ($p<.01$), or MDZ ($p<.01$). The least effective depressant was MDZ, while CPZ and DMCPZ showed intermediate effectiveness and were not significantly different from each other.

Catecholamine release from bovine adrenals. As shown in Fig. 4, DMCPZ is the most effective of the 4 phenothiazines in releasing catecholamines from isolated bovine adrenals. Using Duncan's multiple range test, the release of catecholamines by DMCPZ was significantly greater ($p<.05$) than the release induced by the other agents. Alternatively, the release initiated by CPZ was significantly less ($p<.05$) than that induced by the other phenothiazines. The amounts of catecholamines released by FPZ and MDZ were intermediate and were not statistically different from each other.

Discussion. CPZ was the most effective of the four drugs studied in elevating blood glucose, whereas FPZ was the most effective in depressing spontaneous motor activity. FPZ had little effect on blood sugar. Therefore, of the 4 phenothiazines studied, FPZ produces the greatest CNS depression with relatively little hyperglycemia. These data suggest that it may be possible to eliminate the hyperglycemic action from the phenothiazine moiety without sacrificing CNS activity, by synthesis of the proper analog.

The presence of the adrenals was necessary for the hyperglycemic response to all the phenothiazines studies except DMCPZ. It appears that much of the elevation of blood glucose produced by DMCPZ is independent of release of adrenal catecholamines.

The relative effectiveness of the 4 phenothiazines in raising blood sugar is not correlated with their relative effectiveness in releasing catecholamines from isolated bovine adrenals. For example, CPZ was the most effective of the compounds studied in elevating blood glucose in the intact mouse, yet catecholamine release by CPZ from isolated adrenals occurred only at concentrations of 3×10^{-4} *M* and above. The other phenothiazines released catecholamines at a concentration of 6.6×10^{-5} *M*. Although species differences may account for this lack of correlation, it seems likely that other factors are involved. For example, the extent of the hyperglycemic response may be related not only to adrenal epinephrine release but also to the degree of enhancement of the hyperglycemic effect of epinephrine. Ghafghazi *et al.* (12) showed that epinephrine-induced hyperglycemia was markedly potentiated by CPZ as well as phenoxybenzamine, and suggested that the potentiation resulted from the α-adrenergic blockade produced by these compounds. Therefore, the lack of correlation between hyperglycemic action and epinephrine releasing ability among the phenothiazines studied may be due to a variation in other actions ultimately affecting blood sugar and not due solely to variation in adrenal catecholamine release.

Summary. The ability of 4 phenothiazines to induce hyperglycemia, to decrease motor activity in mice, and to release catecholamines from isolated bovine adrenals, were not

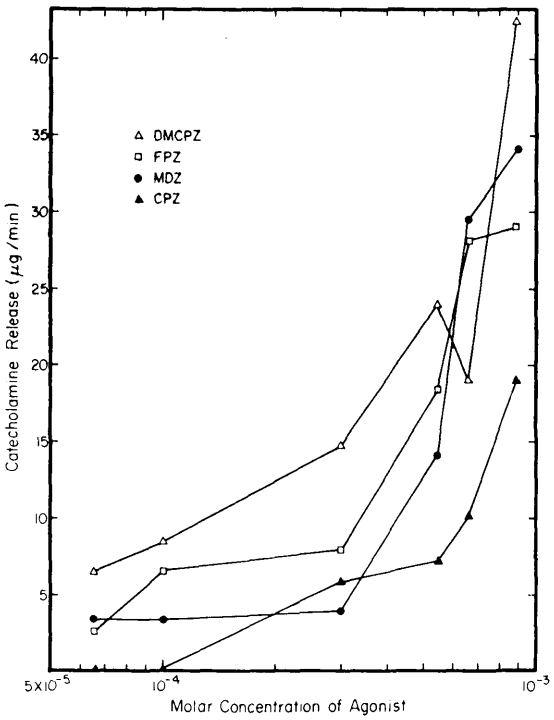

FIG. 4. Dose–response curves for catecholamine release from isolated bovine adrenals by various phenothiazines: Each point is the mean of 8 to 15 determinations of catecholamine release during the 2-min period following infusion of the phenothiazine compound. These values were corrected for resting secretion determined just prior to drug infusion. Abbreviations used are the same as for Fig. 1.

well correlated. Fluphenazine produced the greatest CNS depression with a minimal hyperglycemia. Chlorpromazine produced the greatest hyperglycemia but was not the best releasing agent in isolated adrenals. It appears that the hyperglycemic action of the phenothiazines may be minimized without sacrificing CNS activity by selecting the appropriate derivative. Variation in extra-adrenal effects on blood sugar may explain the lack of correlation between production of hyperglycemia and adrenal catecholamine release by the phenothiazines studied.

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