EFFECT OF METHYLAXANTHINE ANALOGUES ON MOBILIZATION OF LIPID AND CARBOHYDRATE RESERVES

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Analogues of methylxanthines (Ethymizole, Promizone, Ethephyl) induce mobilization of lipid and carbohydrate reserves (the concentration of free fatty acids and glucose in the blood plasma is increased) in intact and hypophysectomized rats.

It was shown previously [6, 7, 9] that the neurotropic methylzanthine analogues Ethymizole (bis-methylamide of 1-ethylimidazole-4, 5-dicarboxylic acid) and propylnorantiphein (bis-methylamide of 1-propylimidazole-4, 5-dicarboxylic acid, laboratory name Promizole), in experiments on animals of different species, effectively stimulate the secretion of ACTH-glucocorticoids and prevents the inhibitory action of exogenous corticosteroids on the hypothalamic centers controlling ACTH secretion [6, 7, 9]. ACTH is known to be an active factor controlling lipolysis [13, 17], while glucocorticoids affect the decisive stages of carbohydrate metabolism [16]. Glucose and higher free (nonesterified) fatty acids (FFA) are the most important trasnportable form of fuel for the cells, completely meeting the requirements of the body for for sources of energy [18].

With these facts in mind, the present investigation was undertaken in order to study the effect of the methylxanthine analogues Ethymizole and Promizole and also the compound Ethephyl (bis-ethylamide of imidazole-4, 5-dicarboxylic acid) and, for comparison, caffeine and theophylline (in the form of euphylline) on mobilization of FFA and glucose.

By studying the character and mechanism of action of neurotropic crugs on the neuriendocrine regulation of metabolism, it is possible to broaden the pharmacological characteristics of these substances and to discover new indications for their use.

EXPERIMENTAL METHOD

Experiments were carried out on 339 adult male rats (170-190 g). The animals were given an ordinary diet (concentrates, root vegetables, and water ad lib.). Hypophysectomy was performed on the rats by the transauricular approach using a semistereotaxic apparatus [12]. The animals were used in the experiments two days after the operation. FFA were determined in the blood plasma as in [14], and glucose by the glucose oxidase method [11]. Specific secretion of ACTH under the influence of the drugs studied was assessed from the concentration of 11-hydroxycorticosteroids (11-HCS) in the blood plasma, which were determined fluorimetrically [5]. Ethymizole, Promizole, and Ethephyl were injected intraperitoneally in doses of 5-20 mg/kg, equivalent to 0.033-0.01 LD_{50} for small laboratory animals [2, 3], and caffeine and theophylline (in the form of euphylline) were given in doses of 20-35 mg/kg. In control experiments equal volumes of physiological saline were injected. The rats were decapitated 60-120 min after the injection of the drugs, and the blood was collected. The results were subjected to statistical analysis [1].

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TABLE 1. Effect of Methylxanthines and Their Analogues on Concentrations of 11-HCS, FFA, and Glucose in Blood Plasma (M ± m)

Substance injected	Dose (mg/kg)	11-HCS (in g %)		FFA (in meq/liter)		Glucose (in mg %)	
		after 1 h	after 2 h	after 1 h	after 2 h	after 1 h	after 2 h
Intact rats							
Physiological saline (control)	0,5 *ml*	8,6±0,6	7,5±0,5	323,3±8,1	313,3±7,9	142,0±3,0	137,6±3,6
Caffeine	20	9,2±0,4	7,4±0,6	317,7±11,8	368,8±25,9	147,3±4,3	138,0±1,5
"	35	8,8±0,5	7,3±0,7	306,0±12,8	313,0±5,4	134,9±2,1	138,5±3,1
Euphylline	20	18,7±0,4*	15,0±0,55*	667,5±23,9**	449,7±17,7*	153,2±1,9	156,5±4,4
Ethymizole	10	26,1±0,8**	16,2±0,6*	668,5±24,6**	433,0±29,5*	148,0±3,2	144,7±5,4
»	20	31,9±0,9**	18,0±1,0†	978,0±23,2**	512,0±20,8**	156,4±2,4	138,5±4,0
Promizole	5	26,7±1,3**	18,7±0,9†	659,6±24,3**	431,2±15,0	148,4±3,7	148,5±3,2
»	10	32,7±1,8**	26,6±1,2†	927,6±24,0**	762,2±25,7**	152,4±1,7	154,8±3,2
Ethephyl	20	13,3±0,6*	10,9±0,8	352,4±13,4	365,2±6,9	154,6±1,4	153,5±2,7
Hypophysectomized rats							
Physiological saline (control)	0,5 *ml*	4,7±0,37		299,2±8,6		146,5±4,7	
Euphylline	20	5,1±0,4		619,8±26,9**		161,8±3,1	
Ethymizole	20	4,6±0,4		625,0±20,3**		195,5±8,4*	
Promizole	10	5,5±0,5		636,1±32,0**		190.4±9,2*	

†$P < 0.01$ relative to appropriate control.

EXPERIMENTAL RESULTS AND DISCUSSION

The concentrations of 11-HCS and FFA in the blood plasma of the rats 1 and 2 h after injection of physiological saline were virtually unchanged. The most marked increase in the indices studied in the blood plasma were observed after administration of Promizole, especially 1 h after its injection (Table 1). Ethymizole also had an effective stimulant effect on the mobilization of 11-HCS and FFA, but only in doses twice as large as those of Promizole. It will be noted that LD_{50} of Ethymizole and Promizole for small laboratory animals is 224 and 250 mg/kg, respectively. LD_{50} for Ethephyl is close to that for Promizole (252 mg/kg).

Euphylline also had a marked stimulant effect on the mobilization of 11-HCS and, in particular, of FFA, although in comparable doses it was less active than Ethymizole. Under the influence of Ethephyl the effects were less marked, while caffeine had no effect on the indices studied.

Methylxanthines, especially theophylline, are known to have the property of inhibiting diphosphoesterase activity and preventing breakdown of cyclic 3.5-AMP [4]. This mechanism lies at the basis of many of the metabolic effects of theophylline, including its stimulant action of pipolysis. The writers have previously shown that Ethymizole potentiates the stimulant action of ACTH on secretion of glucocorticoids by the adrenal cortex [8, 9] and, like theophylline, it increases the permeability of the bladder and skin of frogs to water and sodium [4, 10].

The similarity between the effects of Ethymizole and theophylline provided a basis for the view that these methylxanthine analogues possess peripheral metabolic effects. To investigate this problem experiments were carried out on hypophysectomized rats. They showed (Table 1) that a definite increase in the concentration of FFA, similar in degree to the effects of euphylline, is observed under the influence of Ethymizole and Promizole and in the absence of secretion of the adenophypophysis. Compared with intact animals, the mobilization of FFA under the influence of euphylline showed little change, whereas under the influence of Ethymizole and Promizole it was reduced by about one-third. The fact will be noted that in hypophysectomized rats the substances studied induced a more marked increase in the blood glucose concentration that in intact animals.

The results showed that an important factor in the effect of the methylxanthine analogues Ethymizole and Promizole on FFA mobilization from the triglycerides of the fat depots is their peripheral theophylline-like action, which is potentiated by the secretion of ACTH and, possibly, other pituitary lipotropic hormones.

ACTH is known to induce hypoglycemia [15]. Presumably the absence of any significant effect of Ethymizole and Promizole on the blood plasma glucose concentration in the intact animals can be attributed to the increased ACTH secretion.

It is an interesting fact that injections of Promizole (5-10 mg/kg) and Ethymizole (10 mg/kg) twice a day completely prevent death of animals from a toxic dose of alloxan (280 mg/kg, intraperitoneally) whereas caffeine and theophylline were ineffective in this respect. Presumably in acute diabetes induced by alloxan, carbohydrate deprivation of the tissues is compensated by their utilization of FFA, which are mobilized in considerable amounts under the influence of the methylxanthine analogues studied.

Further investigations are necessary to study the mechanisms of this action.

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