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Effect of some *N N*-disubstituted dithiocarbamates on catecholamines level in rat brain

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DISULFIRAM, an *in vitro* and *in vivo* dopamine-β-hydroxylase inhibitor,[1] lowers the level of noradrenaline (NA) in various tissues, including the brain.[2] A similar effect is exerted by diethyldithiocarbamate (EE),[3] a compound regarded as the active metabolite of disulfiram.[4] Compounds with this type of action may serve as a tool in the investigations on functional role of NA in the central nervous system.

The mechanism of action of EE and of many other known dopamine-β-hydroxylase inhibitors depends upon their ability to form complexes with copper,[5] a metal playing the central role in the mechanism of oxidative hydroxylation of dopamine (DA).[6] *N,N*-disubstituted dithiocarbamates, analogs of EE, also form complexes with copper. It therefore seemed interesting to investigate their effect on the brain catecholamine (CA) level. In this paper the effects of four EE analogs on DA and NA levels in the whole brain of rats are presented. EE was also investigated as a reference compound.

MATERIALS AND METHODS

Sodium salts of the following dithiocarbamates were tested: *N,N*-dimethyl (.2H_2O) (MM), *N,N*-di-*n*-butyl (.H_2O) (BB), *N,N*-dicyclohexyl (CC), *N*-ethyl-*N*-phenyl (PhE, *N*-ethyldithiocarbanilate), and EE (.3H_2O). Male Wistar rats were injected i.p. with saline solutions of the investigated compounds. CC, owing to its poor solubility, was administered as a suspension in saline with Tween 80. Compounds were given at doses corresponding to approx. $\frac{1}{2}$ and $\frac{1}{4}$ of their LD_{50} values for mice.

Animals were killed by transcervical dislocation at various times after treatment, the brains were immediately removed and homogenized in ice-cold 0·4 N $HClO_4$. CA were absorbed on alumina[7] and determined spectrofluorometrically.[8]

RESULTS AND DISCUSSION

All the compounds investigated lowered the brain NA level (Table 1). MM was the most, and PhE the least potent in this respect. In most cases the maximum effect at the higher dose was not much greater than that of the lower. The maximum effect was observed in most cases after 2 hr.

The effect of the compounds on DA level varied. MM and PhE, similarly to EE, tended to enhance the brain DA level. BB and CC significantly decreased the DA brain level; the action of the latter depended upon the dose applied.

The results presented here show that, as expected, the *N,N*-disubstituted dithiocarbamates lower the NA brain level in rats. The dimethyl derivative was more potent in this respect than EE, and further investigations on this compound may prove to be rewarding. The results obtained with EE were similar to those reported by others.[3, 9] The effect of BB and CC on the DA brain content was unexpected, and the elucidation of the mechanism of this action needs further investigation. The

TABLE 1. EFFECT OF *N,N*-DISUBSTITUTED DITHIOCARBAMATES ON THE BRAIN CATECHOLAMINES IN THE RAT

Compound	Dose	NA level as % of level of controls			DA level as % of level of controls		
	(mg/kg)	1 hr	2 hr	4 hr	1 hr	2 hr	4 hr
EE	250	61·6±5·0‡	30·6± 4·7‡	51·3±6·2‡	98·0± 4·0	133·5±10·2*	117·9±26·0
EE	500	60·1±3·6‡	35·5± 2·0‡	45·3±2·9‡	86·1±10·0	116·7±13·7	128·9± 9·3*
MM	250	42·8±3·3‡§	57·2±14·4	45·5±5·5‡	121·9± 8·0§	124·2±10·3	89·0±13·5
MM	500	42·8±2·8‡§	23·1± 1·7‡§	39·2±4·3‡	88·6±21·9	189·2±11·5‡§	104·9±30·5
BB	75	74·3±5·2†	54·4± 4·7‡	69·2±3·6‡	74·2± 8·2*	69·4± 4·3†	59·6± 5·0‡
BB	150	47·5±3·4‡	50·3± 6·5‡	45·3±6·8‡	74·2± 5·0*	70·9±15·3	63·1± 8·0‡
CC	40	75·4±7·5†	56·1± 2·5‡	78·7±8·1*	59·0± 5·2‡	69·7± 3·7‡	66·1±10·3*
CC	80	48·4±4·0‡	41·7± 2·0†	49·8±2·4‡	44·1± 8·6‡	40·3± 5·0‡	48·6± 6·4‡
PhE	200	91·1±5·6	76·7± 8·1	88·2±5·2	119·0±17·9	119·4±11·8	93·7±24·0
PhE	400	80·5±2·5*	39·5± 3·3‡	69·3±4·4†	121·5±11·2	115·5±10·2	130·1±10·1†

Male Wistar rats received the compounds at doses corresponding approx. to $\frac{1}{4}$ and $\frac{1}{2}$ of LD_{50} in mice. NA and DA were determined in the whole brain excluding the olfactory bulbs. The results were compared with the levels of control animals (no injections) sacrificed on the same day, and expressed as percentages of the control values ± SE. The data are means of 5–9 determinations. The control levels (in ng/g of fresh tissue) were: NA: 551±22 (67), DA: 804±42 (66).

Different from the control: * $P < 0.05$, † $P < 0.01$, ‡ $P < 0.001$.

§ MM values significantly different from EE values, $P < 0.05$.

pharmacologic investigations, which are still in progress, revealed some differences between the effects of DA-lowering and DA-enhancing dithiocarbamates in pharmacological tests.

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Effect of tranquillizer drugs on liver tyrosine-α-ketoglutarate transaminase activity

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THE ACTION of reserpine and phenothiazine group of tranquillizers in animals is accompanied by adrenocortical hyperactivity.[1–6] It has been further demonstrated that some transaminases in the liver, *viz.* alanine-α-ketoglutarate transaminase,[7–10] tyrosine-α-ketoglutarate transaminase,[11–15] and tryptophan-α-ketoglutarate transaminase[16] are elevated by the release of ACTH or administration of corticosteroids. Therefore, it was thought of interest to study the effect of tranquillizer drugs on the liver transaminase system. This paper deals with the *in vivo* effect of reserpine and some well-known phenothiazine tranquillizers on the liver tyrosine-α-ketoglutarate transaminase (TKT) activity.

MATERIALS AND METHODS

Male albino rats weighing 120–140 g have been used. Drugs (10 mg/kg) or saline (control) were injected intraperitoneally. Rats were sacrificed at different time intervals. TKT activity was assayed in the liver homogenate by the method of Chan and Cohen.[17]

RESULTS AND DISCUSSION

In Fig. 1 is presented the effect of a single dose of reserpine and three phenothiazine tranquillizers on liver TKT. It is observed therein that amongst the phenothiazines used, prochlorperazine and trifluopromazine caused a maximum increase of the enzyme activity at 2 hr, while chlorpromazine at 6 hr following the injection, and the level of the enzyme returned to normal within 24 hr. But the maximum rise of TKT by reserpine was observed at 12 hr and the enzyme level was still high at 24 hr. Further results, not reported here, indicate that the enzyme activity is back to normal values after about 4 days.

In the present experiment the effect was studied at an equal dose level of the drugs and the results