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* Linoleic acid, 99·5% pure, Nutritional Biochemicals Corporation, Cleveland.
† Brij 35, Atlas Chemical Industries, Inc., Wilmington, Delaware, U.S.A.

Inhibition of dopamine-β-oxidase by diethyldithiocarbamate

SIR,—Hydroxylation of dopamine to noradrenaline by dopamine-β-oxidase has been postulated by Goldstein & Contrera (1961) as the rate-limiting step in the biosynthesis of noradrenaline. If this hypothesis is correct, then the tissue levels of noradrenaline should decrease when this enzyme is inhibited. However, Nikodijevic, Creveling & Udenfriend (1963) using benzyloxyamine and benzylhydrazine analogues were unable to obtain significant decreases in the noradrenaline contents of guinea-pig tissues. More recently, Goldstein, Anagoste, Lauber & McKereghan (1964) found that diethyldithiocarbamate, a metabolite of disulphiram, was a potent inhibitor of dopamine-β-oxidase. In the present work we have shown that diethyldithiocarbamate markedly reduced the noradrenaline levels and simultaneously increased those of dopamine in the small intestine of both the rat and rabbit.

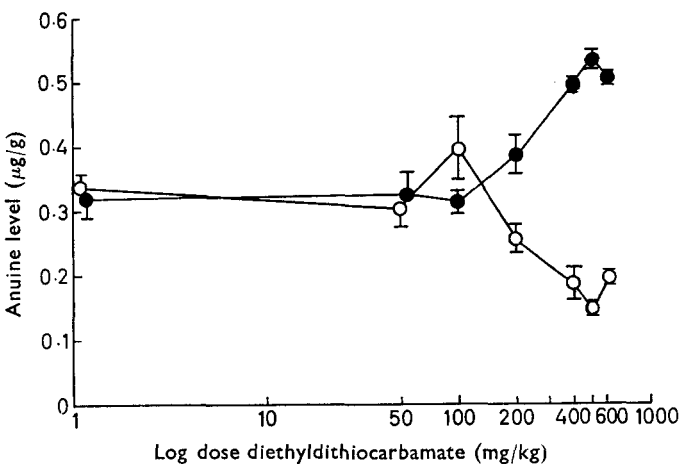

FIG. 1. The effect of different doses of the sodium salt of diethyldithiocarbamate (mg/kg), subcutaneously) on the noradrenaline (○——○) and dopamine (●——●) contents (μg/g ± s.e.) of rat ileum. Each point is the mean of 5 determinations.

The *in vivo* experiments were made by injecting groups of five rats subcutaneously with varying doses of the sodium salt of diethyldithiocarbamate and killing them at hourly intervals over 6 hr. Pieces of ileum were removed, washed, dried, weighed and stored at −4° until assayed for catecholamines. In other experiments, segments of cleaned ileum from untreated rats and rabbits were incubated at 33·5° in McEwen's Ringer solution containing the sodium salt of diethyldithiocarbamate (2×10^{-5}) and aerated with 95% oxygen and 5% carbon dioxide. Segments were removed at different times and treated as described above. The catecholamines (noradrenaline, adrenaline and dopamine) were extracted from tissues by the method used by Shore & Olin (1958). The

content of noradrenaline and adrenaline was estimated as noradrenaline, whereas the dopamine in the final acid extract was estimated by the method of Carlsson & Waldeck (1958, 1959).

The noradrenaline and dopamine content of ileum (μg/g $\pm$ s.e.) of uninjected rats was 0·381 $\pm$ 0·01 (24 animals) and 0·388 $\pm$ 0·011 (30 animals) and of rabbits was 0·357 $\pm$ 0·022 (9 animals), 0·237 $\pm$ 0·029 (7 animals) respectively. The minimal dose of diethyldithiocarbamate required to produce a significant fall in noradrenaline levels in rat ileum was 200 mg/kg, the maximal decrease being at 500 mg/kg when the level was only 43·7% of the control value (see Fig. 1). The corresponding maximal increase in dopamine level also occurred at 500 mg/kg, the value then being 161·3% of the control value.

Incubation for 6 hr resulted in a maximal decrease in the noradrenaline level to 36·7% of the control values in the rat ileum and 45·6% in the rabbit ileum. The corresponding maximal increases in the dopamine levels were 175·6 and 219·1% respectively.

Thus, diethyldithiocarbamate produces significant decreases in noradrenaline levels. It is unlikely that this is due to the release of noradrenaline; firstly, because Goldstein & others (1964) and Musacchio, Kopin & Snyder (1964) showed that the binding and uptake of tritiated noradrenaline was unaffected by disulphiram; secondly, the loss of noradrenaline in the present experiments was accompanied by an increase in dopamine levels; and, thirdly, the mean rate of loss of noradrenaline was estimated as 0·0221 $\pm$ 0·0009 μg/g/hr which was much less than that found with noradrenaline-releasing compounds (Kärki & Paasonen, 1959). The fact that diethyldithiocarbamate produced a significant reduction in endogenous noradrenaline supports the hypothesis that the conversion of dopamine to noradrenaline may be rate-limiting under physiological conditions.

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