CHANGES OF HUMAN PLASMA DOPAMINE-β-HYDROXYLASE ACTIVITY AFTER INTRAVENOUS ADMINISTRATION OF THEOPHYLLINE

D. AUNIS, P. MANDEL & M.T. MIRAS-PORTUGAL
Centre de Neurochimie du CNRS, Faculté de Médecine, 11 rue Humann, 67085 Strasbourg Cedex, France

G. COQUILLAT, F. ROHMER & J.M. WARTER
Clinique Neurologique, Hospices Civils, 67005 Strasbourg Cedex, France

The intravenous administration of theophylline to ten healthy human subjects produced either an increase of circulating plasma dopamine-β-hydroxylase or no change. The rise of plasma enzyme activity may reflect the increased peripheral catecholamine release induced by theophylline.

Introduction Dopamine-β-hydroxylase (3,4-dihydroxyphenylethylamine, ascorbate oxygen: oxireductase (beta-hydroxylating) E.C.1.14.17.1) is localized in the chromaffin granules of the adrenal medulla (Kirshner, 1957) and in the synaptic vesicles of the nerve endings (Potter & Axelrod, 1963). It has been suggested that exocytosis is the mechanism by which noradrenaline secretion occurs from the adrenal medulla (Viveros, Arqueros & Kirshner, 1968) and the sympathetic nerves (Geffen, Livett & Rush, 1969; Weinshilboum, Thoa, Johnson, Kopin & Axelrod, 1971). Dopamine-β-hydroxylase (DBH) activity has been demonstrated in the blood of man and other species (Weinshilboum & Axelrod, 1971) and it has been proposed that plasma DBH activity may serve as an index of the activity of the sympathetic nervous system (for review, see Geffen, 1974).

Methylxanthines have been reported to cause release of catecholamines from the adrenal medulla (Peach, 1972) and sympathetic nerves (Wooten, Thoa, Kopin & Axelrod, 1973). The present investigation was done with the aim of supporting the hypothesis that DBH activity in the serum may serve as a reliable index of catecholamine release in man as judged by variations of the enzyme activity following the intravenous injection of theophylline.

Methods Theophylline (200 mg) was administered intravenously to 10 healthy friends or employees of the Neurological Clinic of the Hospices Civils (Strasbourg). At the indicated time intervals, a sample of blood was drawn from an antecubital vein for the measurement of DBH activity. The blood was collected in heparinized tubes, placed on ice and centrifuged. The plasma was frozen at -30°C until assayed. Plasma DBH activity was estimated according to the technique of Nagatsu & Udenfriend (1972), except that Cu^{2+} was added to the incubation mixture instead of N-ethylmaleimide to reverse endogenous inhibitors. For each individual sample, DBH activity was determined without copper (non-stimulated DBH activity) and at the optimal copper concentration which gave maximal stimulation (copper-stimulated DBH activity). All DBH assays were performed in duplicate on separate 50μl aliquots of plasma and the results of the two measurements were found to fall within ±0.5 units of the mean of the two analyses. The DBH activity was expressed as μmol of octopamine formed per min and per litre of plasma (International Units, i.u.).

Results The data presented in Figure 1 illustrate the changes in circulatory DBH activity following theophylline administration to various individuals. Whereas no change could be observed in five subjects, theophylline induced an increase of DBH activity in the other five. The maximal increase of the non-stimulated DBH activity was seen 1 h after the injection and reached from 15 to 125% of the initial DBH level, depending on the individual. On the other hand, the copper-stimulated DBH activity only increased from 23 to 36% of the initial value and there was a tendency to a further increase between 1 and 2 hours. For each individual, the rise of DBH activity when expressed in number of units was found to be the same in both copper-stimulated and non-stimulated DBH activities.

Discussion Our results show that theophylline induces an increase of DBH levels with a maximum occurring 1 h after the administration. The increase, however, was only observed in 50% of the subjects tested. It should be noted that Snider & Waldeck (1974) have shown that relatively high concentrations of aminophylline are required to induce the release of catecholamines from the isolated adrenal gland. Moreover, it should be emphasized that considerable variation of

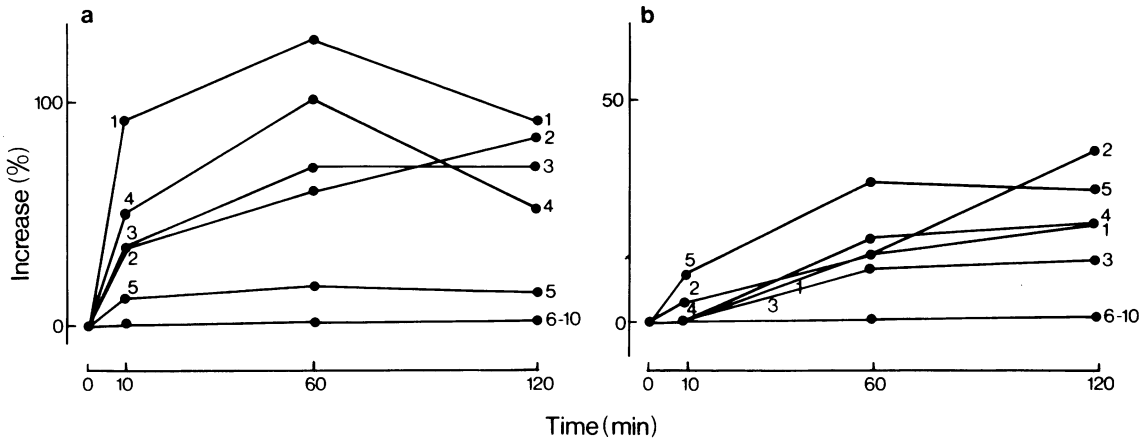

Figure 1 Percent increase in non-stimulated (a) and copper-stimulated (b) dopamine-β-hydroxylase activity after theophylline administration to ten human subjects (age range: 19-70; mean: 37). Values are recorded as percent change from initial values. Numbers on the figure refer to individual subjects. Samples of blood were taken at 10, 60 and 120 min after theophylline administration. The mean value of initial DBH activity of the ten patients was 36 i.u. ± s.e.mean 5, (range: 20-58), for the copper-stimulated activity and 11 i.u. ± s.e.mean 2, (range: 3-17), for the non-stimulated activity. In separate experiments, theophylline was demonstrated to have no activating or inhibiting effect upon DBH.

individual plasma DBH levels can occur (Freedman, Ohuchi, Goldstein, Axelrod, Fish & Dancis, 1972; Wetterberg, Aberg, Ross & Fröden, 1972; Horwitz, Alexander, Lovenberg & Keiser, 1973; Miras-Portugal, Aunis, Mandel, Warter, Coquillat & Kurtz, 1975). Thus it is possible that the theophylline susceptibility differs from one individual to another and that the dose used in our experiments (which was limited by the toxic effect of higher doses of theophylline) was not sufficient to induce DBH release in some subjects.

Variations of DBH activity are determined by changes in its rate of release but also by changes in its rate of clearance from plasma and presumably by the action of endogenous inhibitors (not all of them being reversible by copper, e.g. plasma proteinases). The rise of DBH activity lasted for 1 h or longer. After 1 h, the haemodynamic changes provoked by the theophylline are no longer observable. Therefore, plasma volume changes which might cause variations of plasma DBH activity over a short time interval (Stone, Kirshner, Gunnells & Robinson, 1974; Miras-Portugal *et al.*, 1975) are presumably not responsible for the effect we observed. Thus the rise of plasma DBH activity described in this paper may reflect accurately the increased peripheral catecholamine release induced by theophylline.

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