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Restoration of blood pressure and heart rate responses to tyramine by infusion of 5-hydroxytryptamine in reserpine-treated pithed rats

In the pithed rat, pretreatment with reserpine abolishes the cardiovascular effects of tyramine due to depletion of intraneuronal stores of noradrenaline (Burn & Rand, 1960; Torchiana, Wenger & others, 1966; Clarke & Leach, 1968; Clarke, 1970). An infusion of noradrenaline or one of its precursors, by repleting the tissue stores, restores responses to tyramine (Burn & Rand, 1960; Torchiana & others, 1966). This is easier to demonstrate if deamination of the infused (or formed) amine is prevented by prior injection of drugs possessing monoamine oxidase activity (Clarke & Leach, 1968).

In the pithed reserpinized rat, there is a tissue uptake process for low doses of 5-hydroxytryptamine (5-HT) (Fozard, 1969) similar to that described previously for noradrenaline (Muscholl, 1961; Weiner & Trendelenburg, 1962; Van Zwieten, Widhalm & Hertting, 1965). It was tentatively suggested that 5-HT and noradrenaline shared a common uptake pathway into the sympathetic nerves (Fozard, 1969). The ability of infusions of 5-HT to restore responses to tyramine in reserpinized pithed rats would support such a suggestion.

A total of 21 female Wistar rats weighing 190–230 g were used. Those pretreated with reserpine were given 5 mg/kg intraperitoneally 18–22 h before the experiment. Rats were pithed under pentobarbitone anaesthesia and set up for femoral intravenous injection and recording of carotid blood pressure (Clarke & Leach, 1968). In most experiments heart rate was also recorded (Clarke, Hiscoe & others, 1966). Drugs, dissolved in saline, were given in volumes of 0·1 ml and washed into the animal with 0·2 ml of saline. Infusions were administered into a femoral vein by a Palmer slow injection apparatus at a rate of 2·5 ml/20 min. Test doses of tyramine were not given until 30 min after the heart rate had returned to pre-infusion levels.

The blood pressure and heart rate responses to a 25 μg dose of tyramine were abolished after pretreatment with reserpine (Fig. 1). Infusions of 5-HT (0·5 mg/kg in 20 min) routinely caused an increase in both blood pressure and heart rate, but failed to restore the response to tyramine when this was injected 30 and 60 min after the end

of the infusion (Fig. 1A). Higher doses of 5-HT could not be used since they caused rapid deterioration of the preparations. If an injection of either bretylium (1 mg/kg) or nialamide (20 mg/kg) was given intravenously 10 min before the infusion of 5-HT, the tyramine response was restored when tested 30 and 60 min after completion of the infusion. This is illustrated for bretylium in Fig. 1B. In confirmation of earlier observations (Clarke & Leach, 1968), the same dose of bretylium or nialamide given alone failed to restore the responses to tyramine.

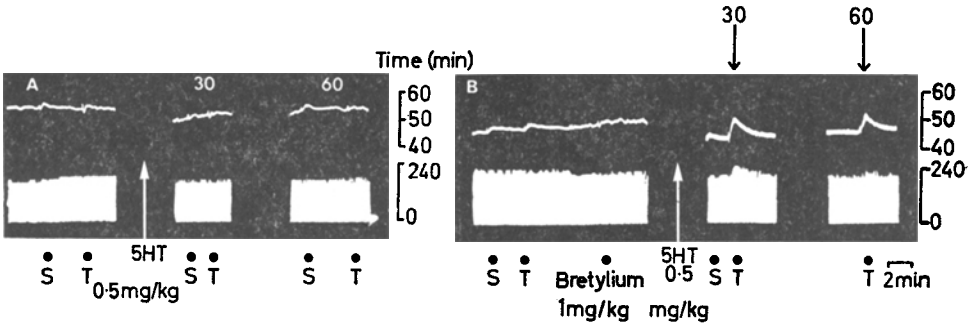

FIG. 1. Reserpine-treated pithed rats. Blood pressure (upper tracing; mmHg) and heart rate (lower tracing; beats/min) responses to tyramine 25 μg (T). A = Before and 30 and 60 min after an intravenous infusion of 5-HT (0·5 mg/kg in 20 min) at arrow. B = As for A except that bretylium (1 mg/kg) was injected 10 min before the infusion. S = Injection of 0·3 ml saline. Time 2 min.

The restored response to tyramine in the presence of bretylium showed quite rapid tachyphylaxis, being reduced to pre-infusion levels between 90 and 120 min after the infusion. Injection of desipramine (0·1 mg/kg) abolished the restored response to tyramine, had no detectable effects on responses to injected 5-HT, and enhanced responses to noradrenaline. In contrast, injection of bromolysergide (0·04 mg/kg) abolished both the restored responses to tyramine and the pressor response to injected 5-HT (Fig. 2). In preparations not treated with reserpine, bromolysergide had no detectable effects on responses to noradrenaline or tyramine in doses up to 0·5 mg/kg.

The temporary restoration of tyramine responses in reserpinized tissues by exposure to noradrenaline, its precursors, or their α-methylated equivalents, is suggested to be due to refilling of transmitter stores (for references, see Iversen, 1967). In our experiments the restored responses to tyramine appear to be mediated indirectly, since they

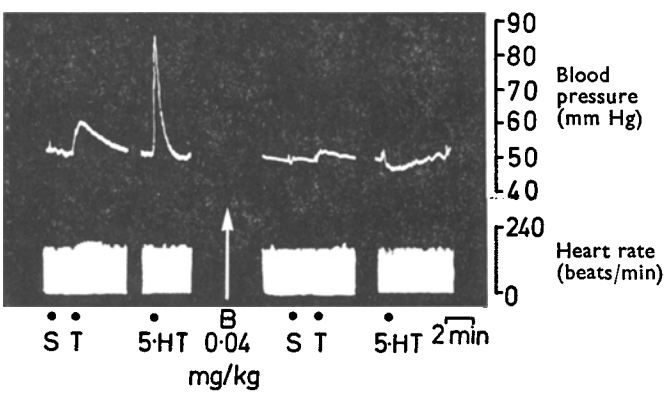

FIG. 2. Reserpine-treated pithed rat injected with bretylium (1 mg/kg) and given an infusion of 5-HT (0·5 mg/kg in 20 min) as in FIG. 1B. Blood pressure and heart rate responses to tyramine 25μg (T) and 5-HT 0·4μg (5-HT) before and after an intravenous injection of bromolysergide (0·04 mg/kg). Other details as in Fig. 1.

were antagonized by desipramine in doses which did not antagonize responses to either 5-HT or noradrenaline. Further, the mediator is likely to be 5-HT since the tryptamine D-receptor antagonist, bromolysergide, antagonized responses to 5-HT and responses to tyramine restored by 5-HT, but not responses to noradrenaline or tyramine mediated by noradrenaline.

The infusion dose of 5-HT used, only restored responses to tyramine when preceded by injection of drugs possessing monoamine oxidase activity. The results are analogous to those obtained by Clarke & Leach (1968), who demonstrated that pretreatment of reserpinized pithed rats with nialamide (20 mg/kg) or bretylium (1mg/kg) increased the efficacy of infusions of noradrenaline in restoring responses to tyramine. After bretylium, they observed an increased tissue retention of noradrenaline as a result of monoamine oxidase inhibition. The major route of inactivation of parenterally administered 5-HT in the reserpine-treated rat is also by oxidative deamination (Erspamer, 1956; Airaksinen, 1963; Axelrod & Inscoe, 1963). This occurs extremely rapidly after intravenous infusions (Fozard, 1969), and would explain the failure of infusions of 5-HT to restore responses to tyramine in untreated reserpinized preparations. Conversely, after nialamide or bretylium, deamination would be slowed and the restorative effects of 5-HT would be enhanced. In this connection, it has been shown that the tissue retention of [^{14}C]-5-HT after intravenous infusion in the pithed reserpinized rat was increased by prior injection of monoamine oxidase inhibitors, including bretylium (Fozard, 1969). The results would also indicate that 5-HT is penetrating to an intracellular site since monoamine oxidase is not located extracellularly (Blaschko, 1952; Kopin, 1964; Carlsson, 1965).

In conclusion these observations provide evidence for an uptake of 5-HT into an intracellular storage site accessible to tyramine. By analogy with similar results from experiments with noradrenaline (Clarke & Leach, 1968) this is likely to be the transmitter store within the sympathetic nerve endings.

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