Methixene: a non-competitive antagonist of bradykinin

SIR,—Garcia Leme & Rocha e Silva (1965) recently reported methixene [9-(1-methylpiperid-3-ylmethyl)thiaxanthen] to be a competitive antagonist of bradykinin in contrast to most of the similar tricyclic compounds they tested.

We have also studied the antagonistic effect of methixene, using the cumulative dose-response procedure according to Rossum (1963), Rossum & Brink (1963) and Ariëns Simonis (1964). Guinea-pig ileum was bathed in a 10 ml bath with a Tyrode solution containing 10^{-7} g/ml atropine sulphate and saturated with a mixture of oxygen 95% and carbon dioxide 5% at 37°.

Two cumulative dose response curves with bradykinin were made and if these curves differed less than 10% the actual experiment was begun. The guinea-pig ileum was incubated with methixene added to the medium to a final concentration of 5×10^{-8}, 10^{-7}, 5×10^{-7}, 10^{-6} g/ml for 30 min, after which period cumulative doses response curves with bradykinin in the presence of the inhibitor were made. The guinea-pig ileum was then washed several times with a fresh Tyrode solution during 30 min and a cumulative dose response curve with bradykinin was then made again. The same preparation was always used for three successive experiments with methixene, the sequence of methixene doses was chosen at random. Only when the last cumulative dose response curve with bradykinin without methixene was within 15% of the first two, was the experiment accepted.

The bradykinin curves were readily reproducible. The lowest dose of bradykinin which induced a concentraction of the guinea-pig ileum in these conditions had a final concentration in the bathing medium of 0·05 μg/ml. The maximum

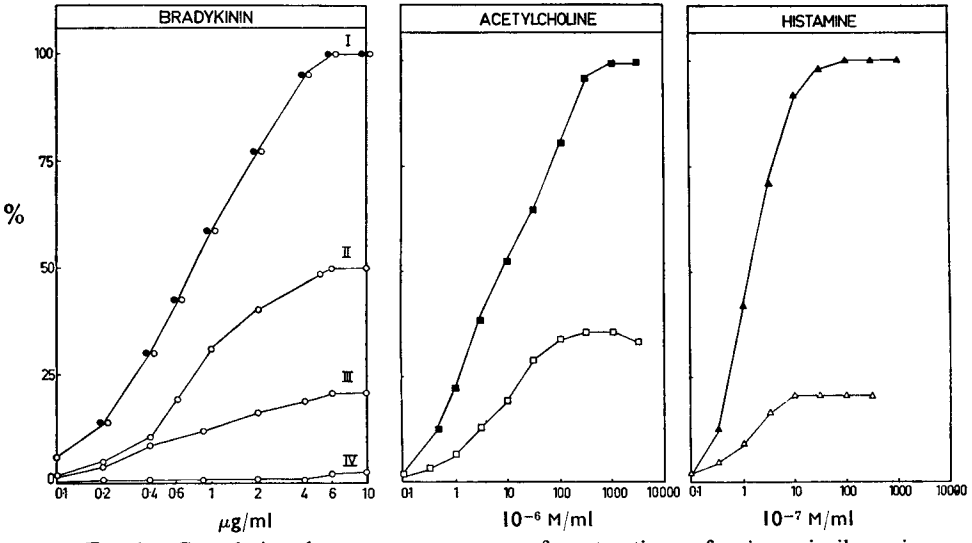

FIG. 1. Cumulative dose response curves of contractions of guinea-pig ileum in Tyrode solution saturated with oxygen 95% and carbon dioxide 5% at 37° (plus 10^{-7} g/ml atropine sulphate in the bradykinin experiments). All values means of 4 curves except where stated. ● Bradykinin controls (mean of 29 curves). ○ Bradykinin in the presence of I, 5×10^{-8}; II, 10^{-7}; III, 5×10^{-7}; IV, 10^{-6} (mean of 2 curves) g/ml of methixene. ■ Acetylcholine controls. □ Acetylcholine and 5×10^{-7} g/ml methixene. ▲ Histamine controls. △ Histamine and 5×10^{-7} g/ml methixene.

contraction, after which a tenfold increase of concentration did not induce a further increase in the contraction of the ileum, was found regularly at 10 μg/ml.

In the presence of 5×10^{-8} g/ml methixene no antagonistic effect was seen (Fig. 1). However, 10^{-7} g/ml methixene decreased the effect of bradykinin, but not the affinity. This was more pronounced with the 5×10^{-7} g/ml concentration of methixene. In the final concentration of 10^{-6} g/ml methixene the inhibition was almost completed (Fig. 1). These results suggest that methixene is a noncompetitive antagonist of bradykinin. This is not in agreement with the findings of Garcia Leme & Rocha e Silva who used a three dose technique on the guinea-pig ileum.

Additional experiments supported our findings of the non-specificity of the methixene blocking effect of bradykinin. In the urethane anaesthetised guinea-pig the effect of bradykinin on the bronchioles was examined with a modification of the method of Konzett & Rössler (1940). Methixene did not antagonise the bronchoconstrictor effect elicited by bradykinin in any of the doses given (0·01–10 mg/100 g i.v.).

TABLE 1. EFFECTS OF METHIXENE ON THE VASODILATOR RESPONSE OF RATS IN URETHANE ANAESTHESIA (1·1 ml 12·5% i.p./100 g) TO BRADYKININ INJECTED INTO THE TAIL VEIN. Pressure recorded from the carotid artery. Measurements to the nearest 5 mm Hg accurate with Statham P 23 transducers and Grass polygraph inkwriting recorder. Number of observations in brackets. Blood pressure given in mm Hg.

		Methixene (μg)			
Bradykinin μg	control	45	150	450	1500
0·25	5 (8)	10 (9)	15 (5)	25 (10)	40 (4)
0·5	10 (3)	—	—	25 (3)	40 (2)

The blood pressure decrease caused by the vasodilator effect of bradykinin recorded in the urethane anaesthetised rat was even potentiated by methixene in a dose dependent manner (Table 1). In addition, methixene was equally active as a non-competitive antagonist of the contractions of the guinea-pig ileum caused by cumulative doses of histamine and acetylcholine (Fig. 1) and also of the contractions caused by 10^{-5}M/ml 5-hydroxytryptamine and 1 U/ml substance P, which were 80% inhibited by 5×10^{-7} g/ml methixene. The contractions caused by 10^{-3} M/ml barium chloride on the guinea-pig ileum were also inhibited by the same concentration of methixene.

In conclusion methixene appears to be a non-competitive inhibitor of bradykinin and of several other agonists on the guinea-pig ileum.

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