cocaine in any study to assess the potency of an agonist or antagonist.

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The interaction of cimetidine with various antihypertensive agents in the spontaneously hypertensive rat

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The H_2-receptor antagonist cimetidine (250 μg) administered intracerebroventricularly (i.c.v.) 15 and 30 min before clonidine (25 μg kg^{-1} i.v.), significantly antagonized clonidine-induced hypotension in anaesthetized spontaneously hypertensive rats. The hypertensive response of cimetidine was correlated with the inhibition of clonidine-induced hypotension. In addition, cimetidine (250 μg i.c.v.) counteracted the hypotensive effects of pentolinium (5·0 mg kg^{-1} i.v.), guanethidine (5·0 mg kg^{-1} i.v.) and minoxidil (1·0 mg kg^{-1} i.v.) These data do not support previous suggestions that the hypotensive action of clonidine is caused by stimulation of the H_2-receptor, but suggest that central administration of cimetidine causes peripheral vasoconstriction and this may offer resistance to the hypotensive action of different antihypertensive agents.

The interaction between clonidine and histamine H_2-receptor antagonists has indicated that histamine H_2-receptors are involved in the central hypotensive action of clonidine (Karppanen et al 1976, 1977; Finch et al 1978). However, there have been other findings which raise some doubt about this hypothesis, namely the induction of sleep in chicks, and the increase in locomotor activity in rats, are not inhibited by H_2-receptor antagonists (Vogt 1977; Nomura & Segawa 1979). Furthermore, Pilc et al (1979) and Timmermans et al (1980) have demonstrated that, in rat cerebral cortex, clonidine is bound specifically to sites different from the histamine binding sites. We have previously shown that in anaesthetized spontaneously hypertensive (SH) rats, central administration of cimetidine causes a sustained rise in perfusion pressure of the auto-perfused hindquarter (Dohadwalla & Dadkar 1981). In view of these observations, we have investigated the interaction between various antihypertensive agents and the H_2-receptor antagonist cimetidine, in anaesthetized SH rats.

* Correspondence.

Materials and methods

Male SH rats (230–250 g) the strain developed by Okamoto & Aoki (1963) were used. Permanent cannulation of lateral cerebroventricles was performed stereotaxically in pentobarbitone sodium anaesthetized animals as described by Dadkar et al (1984). These rats were allowed to rest for two to three days. On the day of experiment, they were anaesthetized with pentobarbitone sodium (50 mg kg^{-1} i.p.) and blood pressure was recorded on a Hellige physiological recorder using Statham P 23 Db pressure transducer. Mean blood pressure was calculated as diastolic pressure plus one third of pulse pressure. Intravenous administration of drug was carried out by cannulation of right jugular vein. Results were expressed as change in blood pressure over initial value in mmHg for each rat. The site of intracerebroventricular (i.c.v.) injection was confirmed at the end of the experiment by injection of 10 μl of Evans blue, and subsequent microscopic examination.

The drugs used were: clonidine hydrochloride (Boehringer Ingelheim), minoxidil (Upjohn), pentolinium tartrate (May & Baker) and guanethidine hydrochloride (Ciba-Geigy), dissolved in 0·9% sodium chloride and administered intravenously. Cimetidine

(SK&F) was dissolved in 0·1 M HCl and the pH was adjusted to 6 with 0·1 M NaOH and injected into the lateral ventricle in volume of 10 μl by means of a Hamilton microsyringe. The control animals were treated with vehicle. Results are expressed as mean ± s.e.m. Statistical analysis of the results was by Student's *t*-test. The hypotensive effect (0–60 min) of clonidine was expressed as area under the curve (trapezoidal rule) and examined using an analysis of variance. Differences were considered to be significant at $P < 0·05$ level.

Results

Effect of cimetidine (i.c.v.) on clonidine-induced hypotension. The hypotensive action of clonidine was investigated alone or in combination with cimetidine. Clonidine (25 μg kg^{-1} i.v.) elicited a marked hypotensive effect, with a maximum fall in mean blood presure (96·7 ± 6·9 mmHg) within 10 min which lasted for about 50–60 min. Cimetidine (250 μg i.c.v.) produced a pressor response. The effect began to develop gradually and the maximum was observed at 15–30 min (Fig. 1). Pretreatment with cimetidine at different time intervals significantly antagonized the hypotensive effect of clonidine. Furthermore, the maximum inhibition of the hypotensive effect of clonidine by cimetidine was correlated with its maximum pressor response (Fig. 1).

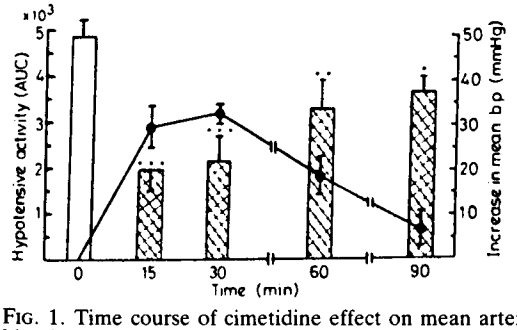

FIG. 1. Time course of cimetidine effect on mean arterial blood pressure and on hypotensive activity of clonidine in anaesthetized SH rats. Cimetidine was given 15–90 min before clonidine. Each point represents the mean ± s.e.m. of 8–10 observations. Cimetidine (250 μg i.c.v.) (●), clonidine (25 μg kg^{-1} i.v.) (open column) and cimetidine + clonidine (hatched columns). Results were evaluated for significance by an analysis of variance; (*$P < 0·05$, **$P < 0·01$, ***$P < 0·001$ compared with vehicle pretreated group).

Effect of cimetidine (i.c.v.) on minoxidil-, pentolinium- and guanethidine-induced hypotension. To investigate the specificity of the interaction between clonidine and cimetidine, we examined the interaction of cimetidine on established hypotensive action of different antihypertensive agents, which are known to elicit a blood pressure lowering effect either by direct action on the vascular smooth muscle (minoxidil) or by interruption of sympathetic tone (pentolinium and guanethidine).

Minoxidil (1 mg kg^{-1} i.v.) elicited a gradual fall in systemic blood pressure (69·4 ± 9·4 mmHg) in anaesthetized rats. The effect commenced within 1–2 min after the injection and persisted throughout the test period (60 min). When cimetidine (250 μg) was injected into the lateral cerebroventricles 15 min after the minoxidil injection, it significantly counteracted the established hypotensive action of minoxidil (Fig. 2). Pentolinium 5·0 mg kg^{-1} i.v.) and guanethidine (5·0 mg kg^{-1} i.v.) produced a fall in systemic blood pressure of 79·3 ± 6·6 and 73·7 ± 4·4 mmHg respectively in anaesthetized rats. The maximum hypotension was seen within 15–20 min and persisted throughout the period of observation (60 min). Injection of cimetidine (250 μg i.v.) 15 min after administration of these agents caused a significant reversal of pentolinium- or guanethidine-induced hypotension (Fig. 2).

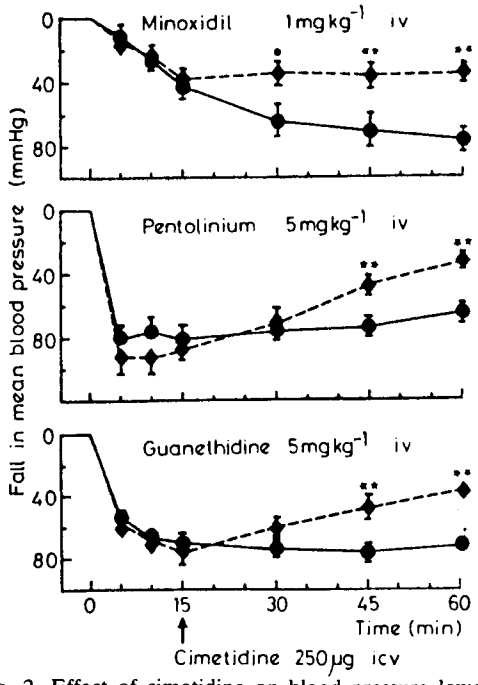

FIG. 2. Effect of cimetidine on blood pressure lowering activity of minoxidil, pentolinium and guanethidine in anaesthetized SH rats. Each point represents the mean ± s.e.m. of 10 observations. (●—●) Vehicle-treated animals, (● - - - ●) cimetidine-treated animals. Results were evaluated for significance by Student's *t*-test (*$P < 0·05$; **$P < 0·01$ compared with vehicle treated group).

Discussion

It has been reported previously, that, reduction in hypotensive activity of clonidine after intracerebroventricular (i.c.v.) administration of an H_2-receptor antagonist was much greater in established hypotension than that observed in pretreated rats (Karppanen et al 1976; Dohadwalla & Dadkar 1981). Furthermore, it has been demonstrated that the pressor action of central administration of cimetidine is mediated through an increase in efferent sympathetic outflow and released catechol-

amines from adrenal medulla (Dadkar et al 1984). These results suggest that pressor effect of cimetidine may counteract the hypotensive effect of clonidine.

In the present study, we have demonstrated that in anaesthetized SH rats, pretreatment with the H_2-receptor antagonist, cimetidine (i.c.v.) inhibited the fall in blood pressure induced by intravenous injection of clonidine. In addition, the inhibition of the hypotensive effect of clonidine by cimetidine is found to be dependent on its pressor effect. From this finding, it seems reasonable to assume that increased peripheral sympathetic activity by cimetidine may be responsible for inhibition of the hypotensive action of clonidine.

Furthermore, it has been demonstrated that central administration of cimetidine causes a sustained rise in perfusion pressure in autoperfused hind quarters of SH rats, and it was suggested that cimetidine might be increasing the vascular resistance due to alteration in the neurogenic tone of hindlimb vasculature (Dohadwalla & Dadkar 1981). To investigate the possibility of involvement of peripheral action of cimetidine in countering the hypotensive effect of clonidine, further interaction studies were performed using different antihypertensive agents which are known to elicit blood pressure lowering effect either by interruption of peripheral sympathetic tone or by direct action on the vascular smooth muscle. It was observed that cimetidine (250 μg i.c.v.) significantly counteracted the established hypotensive action of minoxidil, pentolinium and guanethidine, which are supposed to act mainly at the level of peripheral vasculature (DuCharme et al 1973: Willems & Bogaert 1978).

From these observations, it can be concluded that i.c.v. administration of cimetidine results in peripheral vasoconstriction and this may offer resistance to hypotensive actions not only of clonidine, but also to other hypotensive agents. Moreover, these findings also reject the hypothesis that clonidine initiates its central hypotensive effect via histamine H_2-receptors.

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Uptake of amino β-lactam antibiotics into rat intestinal brush border membrane vesicles

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Uptake of amino β-lactam antibiotics into rat intestinal brush border membrane vesicles has been examined for characterization of the transport of the antibiotics through the gut wall of the rat. The uptake of cephradine, cephalexin and ampicillin into membrane vesicles was similar, and there were no significant changes in the uptake in the presence of a NaCl or a KCl gradient. These results suggested that the carrier-mediated transport systems relating to amino acids and glucose were not concerned with the intestinal absorption mechanism of amino β-lactam antibiotics.

Amino β-lactam antibiotics are absorbed from the intestinal lumen after oral administration despite poor lipophilicity. Recent findings suggested that the intestinal absorption of antibiotics such as cephalexin and cephradine involved a carrier-mediated transport system relating to amino acids and dipeptides (Kimura et al 1978, 1982). However, in a preliminary experiment, we found that in the presence of several amino acids and dipeptides there were no changes in the absorption of these antibiotics. We have therefore used membrane vesicles for the investigation and characterization of amino β-lactam antibiotics transport through the brush border membranes of rat small intestine.

* Correspondence.

Method

Brush border membrane vesicles were isolated from the intestine of male Wistar rats (180–230 g) according to the calcium chloride precipitation method of Kessler et al (1978). The membranes were suspended to a final concentration of about 4 mg protein ml^{-1} with 20 mM