

AN INDIRECT SYMPATHOMIMETIC EFFECT OF BURIMAMIDE ON KITTEN ISOLATED ATRIA

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- 1 Burimamide (34-1080 μM) caused a concentration-dependent increase in the force and frequency of contraction of kitten isolated atria.
- 2 Metiamide (467 μM) had no stimulant action on kitten atria and did not modify the effects of burimamide.
- 3 The atrial stimulation produced by burimamide was reduced by (–)-propranolol (34-68 nM) and by cocaine (3 μM).
- 4 The atrial stimulant effect of burimamide was prevented by pretreatment of kittens with reserpine (1 mg/kg, 24 h before the experiment).
- 5 It is concluded that burimamide causes atrial stimulation by releasing catecholamines.

Introduction

Burimamide antagonizes the increase in gastric acid secretion induced by histamine, or penta-gastrin, by blocking histamine H_2 -receptors (Black, Duncan, Durant, Ganellin & Parsons, 1972). In addition, burimamide also stimulates acid secretion when injected into cats, possibly by acting as a partial agonist on the histamine H_2 -receptors in the stomach (Harris, Reed, Smy & Venables, 1975). In the present study, the effects of burimamide on kitten isolated atria have been investigated in an attempt to determine whether burimamide also stimulates other histamine H_2 -receptors in the cat. It has been found that burimamide possesses an appreciable cardiac stimulant action; this appears to be brought about by release of catecholamines, an effect previously reported in anaesthetized cats (Albinus & Sewing, 1973).

Methods

Kittens of either sex and weighing 400-900 g were anaesthetized with ether. Heparin (2500 u) was injected into a femoral vein. The heart was removed, washed in McEwen's solution (McEwen, 1956) at room temperature, and then transferred to the medium at 37°C. The atria were dissected from the ventricles and the preparation was placed in a 70 ml isolated organ bath. The McEwen's solution was maintained at 37 \pm 0.5°C and was bubbled with 95% O_2 and 5% CO_2 .

Contractions of the atria were monitored with an Ether ST202 force displacement transducer

connected to a Devices M2 recorder. Heart rate was recorded with a Devices Impulse Integrator (type 3210) which was triggered from the transducer signal. The atria were placed under an initial tension of 0.5-1.2 grams. Experiments were started after a 60 min equilibration period, during which time the bath fluid was changed six times.

Two types of experiment were performed: (a) concentration-response curve experiments, and (b) bracketing experiments consisting of single sub-maximal responses to noradrenaline, histamine, tyramine and burimamide. Concentration-response curves were constructed by giving an ascending series of doses, followed by washing out of each dose, this being continued until there were two or more identical increases in heart rate. The dosing procedure was as follows: noradrenaline, 3 min contact with the tissue on a 10 min cycle followed by six changes of bathing solution: histamine, 5 min contact on a 15 min cycle with 12 changes of bathing solution: burimamide, 5 min contact on a 20 min cycle with 18 changes of bathing solution. Each preparation in this series of experiments provided curves for all three agonist drugs.

In the bracketing experiments, submaximal responses of the atria to single doses of the agonists were matched. The three agonist drugs were then repeated in the presence of the modifying drug, propranolol and metiamide being added 1 min before each dose of agonist, whereas cocaine was allowed to act for 5 minutes.

Two kittens were pretreated with reserpine (1 mg/kg, i.p., 24 h before the experiment), the

drug solution being prepared by the method of Burack, Weiner & Hagen (1960).

Results were calculated as the mean \pm s.e. mean and analysed for significance of difference of means by Student's *t* test following an analysis of variance.

Drugs

The following drugs were used: tyramine hydrochloride and (–)-noradrenaline bitartrate (Koch-Light); histamine acid phosphate and reserpine (BDH); (–)-propranolol hydrochloride (ICI); cocaine hydrochloride (Mawson & Proctor); heparin (Evans); anaesthetic ether B.P. (May & Baker); burimamide and metiamide (S.K. & F.). Solutions of noradrenaline were protected from the light. Burimamide and metiamide were dissolved in the minimum volume of 10 N HCl, diluted with water, and taken to pH 7 with dilute NaOH.

Results

Chronotropic and inotropic actions of burimamide

In 22 experiments the control heart rate was 157 ± 5 beats/min and the tension developed each beat was 760 ± 55 mg. Burimamide (34 – $1080 \mu\text{M}$) stimulated the atria, causing both chronotropic and inotropic effects. There was no tachyphylaxis to the cardiac effects of $270 \mu\text{M}$ burimamide over the first three applications ($n = 5$), although there was some loss of response after the seventh application of this dose in one preparation.

The cardiac stimulant effects of burimamide have been compared with those produced by noradrenaline and histamine. The positive chronotropic effects of the three drugs are shown in Figure 1. The maximum responses were: noradrenaline 130 (beats/min increase in heart rate), burimamide 84 and histamine 41. Burimamide and noradrenaline also exerted substantial positive inotropic effects, but histamine produced only a small negative inotropic response. However, the

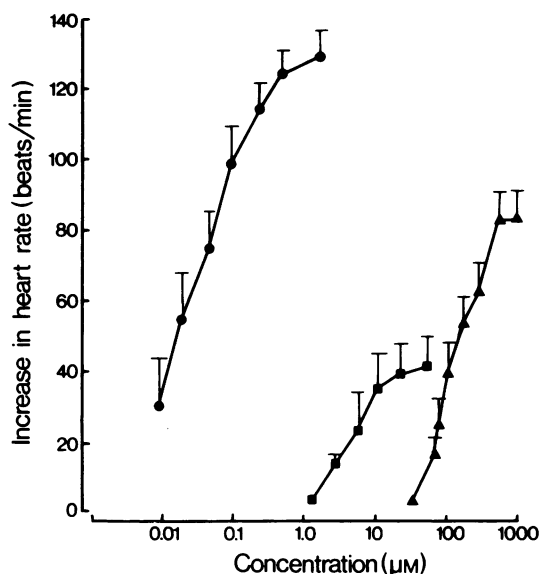


Figure 1 Concentration-response curves for the positive chronotropic response of kitten isolated atria to noradrenaline (●), histamine (▲) and burimamide (■). Vertical bars show s.e. mean of five experiments. Note that the maximum effect of burimamide was greater than that of histamine.

response of the atria to histamine was very variable; on some occasions a small positive inotropic effect was seen (see Figure 2 for example), and in some of the later experiments a positive chronotropic response was observed which was greater than the maximum in Figure 1.

Effects of various pretreatments on the cardiac response to burimamide

Metiamide Metiamide ($467 \mu\text{M}$), a more potent histamine H_2 -receptor antagonist than burimamide (Black, Duncan, Emmett, Ganellin, Hesselbo, Parsons & Wyllie, 1973), produced a small positive inotropic effect (<200 mg increase in tension,

Table 1 The effects of drugs on the positive chronotropic effects of agonists on kitten atria

Drugs	μM	n	Response as % control (s.e. mean)			
			Noradrenaline (17–170 nM)	Histamine (10–20 μM)	Tyramine (2 μM)	Burimamide (270 μM)
Metiamide	467	5	90 (10)	20 (10)*		104 (9)
(–)-Propranolol	0.034–0.068	6	40 (22)*	105 (10)		12 (4)*
Cocaine	3	5	134 (5)		28 (4)*	24 (7)*

* Signifies a significantly lower tachycardia after the application of a modifying drug ($P < 0.05$).

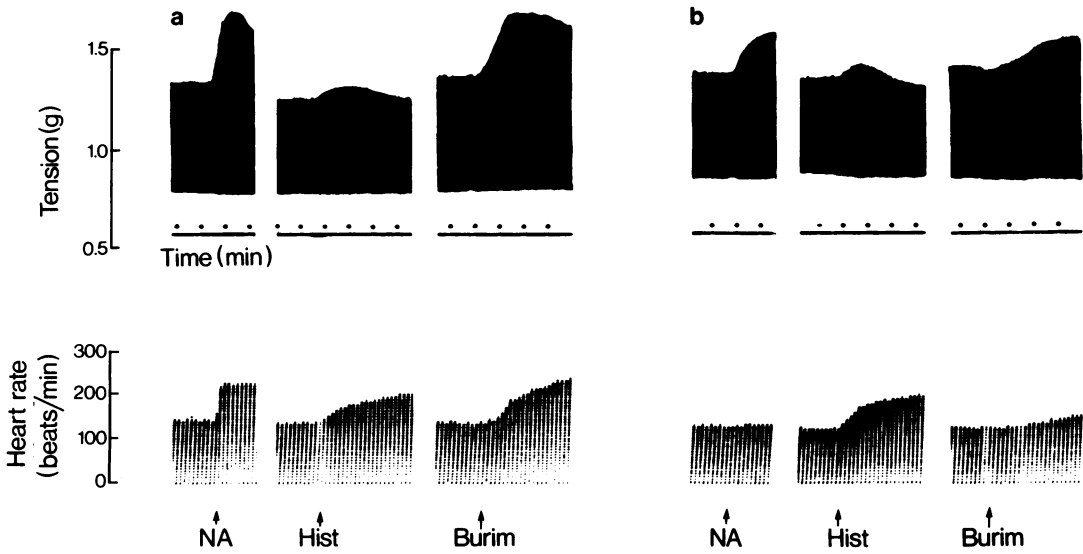


Figure 2 The effect of noradrenaline 85 nM (NA), histamine 10 μ M (Hist) and burimamide 270 μ M (Burim) on kitten isolated atria alone (a) and in the presence (b) of (–)-propranolol (68 nM added 1 min before each agonist). Note that the effects of noradrenaline and burimamide were reduced, but the sensitivity of the atria to histamine was unaffected.

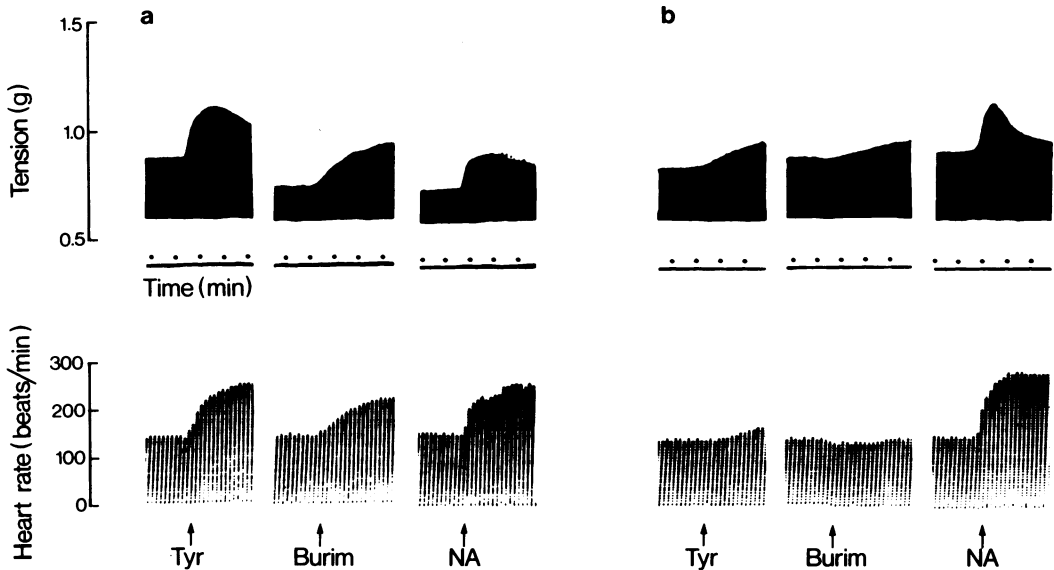


Figure 3 The effects of tyramine 2 μ M (Tyr), burimamide 270 μ M (Burim) and noradrenaline 42 nM (NA) before (a) and after (b) cocaine (3 μ M added 5 min before each agonist). Note that the effects of tyramine and burimamide were reduced after cocaine whereas the effect of noradrenaline was increased.

Table 2 Effect of reserpine on the positive chronotropic effect of agonists on kitten atria

	<i>Tachycardia (s.e. mean) beats/min</i>		
	<i>Noradrenaline (42 nM)</i>	<i>Tyramine (2 µM)</i>	<i>Burimamide (270 µM)</i>
Control group	82 (12)	96 (8)	71 (4)
Reserpine-treated kitten 1	60	26	3
Reserpine-treated kitten 2	113	0	0

Reserpine (1 mg/kg, i.p.) was administered 24 h before the isolated atria were prepared. There were five preparations in the control group.

$n = 8$) which was usually preceded by a small negative chronotropic effect ($n = 6/8$). The subsequent application of the cardiac stimulants, noradrenaline (42–85 nM), histamine (10–20 µM) and burimamide (270 µM), revealed a specific antagonism of histamine (Table 1).

Propranolol and cocaine Sub-maximal responses to doses of noradrenaline, histamine and burimamide were tested before and then 1 min after (–)-propranolol (34–68 nM). Results of a typical experiment are shown in Figure 2. Propranolol reduced both the chronotropic and inotropic effects of noradrenaline and burimamide, but did not affect the sensitivity of the atria to histamine (see Table 1).

Cocaine (3 µM) reduced the effects of tyramine and burimamide, whereas noradrenaline was more effective than in the controls (Figure 3 and Table 1).

Reserpine Pretreatment of two kittens with reserpine (1 mg/kg, i.p., 24 h before the experiment) reduced the sensitivity of the atria to tyramine and prevented the stimulant action of burimamide (Table 2). Responses to noradrenaline were unaffected by reserpine.

Discussion

In the present experiments on the isolated, spontaneously-beating atria of the kitten there is no evidence to suggest that burimamide is a partial agonist on histamine H_2 -receptors. The positive inotropic response to histamine of the atria was variable, and not always present; in contrast, burimamide consistently increased the frequency and force of contractions. The maximum effects of burimamide on both power and rate of beating were much greater than were those in the presence of histamine. The effects of burimamide were not blocked by metiamide, whereas those of histamine were antagonized. It can therefore be concluded

that the cardiac stimulant action of burimamide in these experiments was not due to an effect on histamine H_2 -receptors.

(–)-Propranolol significantly reduced the response to burimamide and noradrenaline without affecting sensitivity to histamine. It is unlikely that the 'non-specific' activity of propranolol (Papp & Vaughan Williams, 1969) contributed to the blockade of the effects of burimamide because of the low concentrations (34–68 nM) of the β -adrenoceptor blocking agent that were used. However, it was surprising that propranolol was more effective against burimamide than noradrenaline.

Experiments with cocaine and reserpine showed that the sympathomimetic effect of burimamide was an indirect effect, presumably caused by a release of noradrenaline from adrenergic neurones in the atria. Cocaine inhibits the active transport of monoamines into the neurone, thus preventing tyramine reaching the noradrenaline stores and by the same action increasing the concentration of exogenous noradrenaline in the vicinity of the receptors on the cardiac cells (MacMillan, 1959; Iversen, 1965). Since, in the present experiments, cocaine reduced the response of the atria to burimamide in a similar fashion to its effect on tyramine, it appears that burimamide can release catecholamines from the nerve endings by a tyramine-like action. This, considered with the insensitivity of the atria from the reserpine-treated kittens to both burimamide and tyramine, suggests that burimamide is an indirectly-acting sympathomimetic agent in this species.

Albinus & Sewing (1973) found that burimamide (4 mg/kg) increased blood pressure and heart rate in anaesthetized cats, and that these effects were antagonized by a mixture of α - and β -adrenoceptor blocking agents. The rise in blood pressure could also be prevented by acute adrenalectomy, but the tachycardia was not reduced. It is possible that a release of catecholamines in the heart, such as described in this paper, was responsible for the tachycardia which occurred in their experiments.

Cardiovascular effects following burimamide have also been seen in dogs (Lorenz, Thermann, Hamelmann, Schmal, Maroske, Reimann, Kusche, Schingale, Dormann & Keck, 1973). In man, Wyllie, Hesselbo & Black (1972) observed an increase in heart rate from 81 beats/min to 96 beats/min during infusion of burimamide. The plasma concentration of burimamide in the experiments in man reached a maximum of $40 \mu\text{M}$, a concentration that produced a tachycardia in the kitten isolated atria. However, although there are no data on the plasma concentration of burimamide accompanying histamine H_2 -receptor blockade in the kitten, it is probable that

appreciable indirect sympathomimetic activity occurs only at concentrations of burimamide greater than those needed for receptor blockade. In contrast with the present findings for burimamide, there was no evidence that metiamide released catecholamines from the heart. This represents yet another advantage of metiamide when compared with burimamide.

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