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CAPSAICIN-SENSITIVE AFFERENTS ACTIVATE A SYMPATHETIC INTESTINOINTESTINAL INHIBITORY REFLEX IN DOGS

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SUMMARY

1. In urethane-anaesthetized dogs, an intra-arterial infusion of capsaicin (0·7–14 nmol min^{-1}) into a separated jejunal segment inhibited a vagally evoked cholinergic contraction of the other non-infused segments. The mechanism of this reflex was investigated.

2. The inhibition by capsaicin was abolished after bilateral splanchnic nerve section or cervical spinal cord transection (C5 or C6), but was unaffected by bilateral vagotomy. Decerebration partially reduced the inhibition.

3. The inhibition by capsaicin was abolished by pre-treatment with phentolamine or yohimbine, but was unaffected by prazosin or propranolol.

4. Sympathetic efferent discharge of the mesenteric nerve increased with capsaicin application, during which time vagally evoked contractions were inhibited.

5. Single-unit discharges of the major splanchnic and mesenteric afferents increased with capsaicin infusion to the loop which was innervated by the units.

6. Together the results implied that capsaicin stimulated canine intestinal primary afferents, resulting in the sympathetic intestinointestinal inhibitory reflex supraspinally. The inhibition of vagally evoked contractions may be due to a presynaptic inhibition via α_2-adrenoceptors, which are activated by the reflex.

INTRODUCTION

It is currently believed that capsaicin activates certain primary afferent neurones and induces a number of reflexes. By topical application or close intra-arterial injection of capsaicin, activation of visceral afferents located at various levels (stomach, small intestine, gall-bladder, pancreas, urinary bladder, etc.) produced a variety of responses of cardiovascular functions (Longhurst, Ashton & Iwamoto, 1980; Lembeck & Skofitsch, 1982; Ordway & Longhurst, 1983; Ordway, Longhurst & Mitchell, 1983; Longhurst, Stebbins & Ordway, 1984*a*; Giuliani, Maggi & Meli, 1988). Application of capsaicin to the outer surface of the urinary bladder activated the micturition reflex and induced phasic contractions of the urinary bladder (Maggi, Santicioli, Borsini, Giuliani & Meli, 1986). However, the intestinointestinal reflex evoked by capsaicin has not been determined in detail.

Thus the aim of this study was to investigate if capsaicin activates the

intestinointestinal reflex in a manner similar to that by distension in dogs. In addition, the pathway and centre of the reflex, and the mechanism of the inhibition by the reflex on vagally evoked contractions were studied.

METHODS

Thirty-nine mongrel dogs of both sexes weighing 5–13 kg were used. The animals were anaesthetized with an intramuscular injection of ketamine hydrochloride (10–20 mg kg^{-1}). A tracheal cannula was inserted. The femoral artery was cannulated and connected to a pressure transducer (Nihon Koden, MPU-0·5A) to record the arterial blood pressure. The femoral vein was also cannulated for the systemic administration of lactic Ringer solution (5 ml kg^{-1} h^{-1}), which contained 50 mg ml^{-1} urethane, 0·5 mg ml^{-1} gallamine and 50 mg ml^{-1} glucose, to compensate for the loss of body fluid and to maintain anaesthesia and immobilization of the animals. The end-tidal CO_2 pressure of expired gas was monitored by a capnometer (Hewlett Packard, 47210A) and was kept within a range of 4·5–5·7 kPa by means of regulating the rate and volume of ventilation. The body temperature was maintained at 36–38 °C by a heating pad placed under the body. The experiments were carried out in those animals whose mean blood pressure was kept above 80 mmHg.

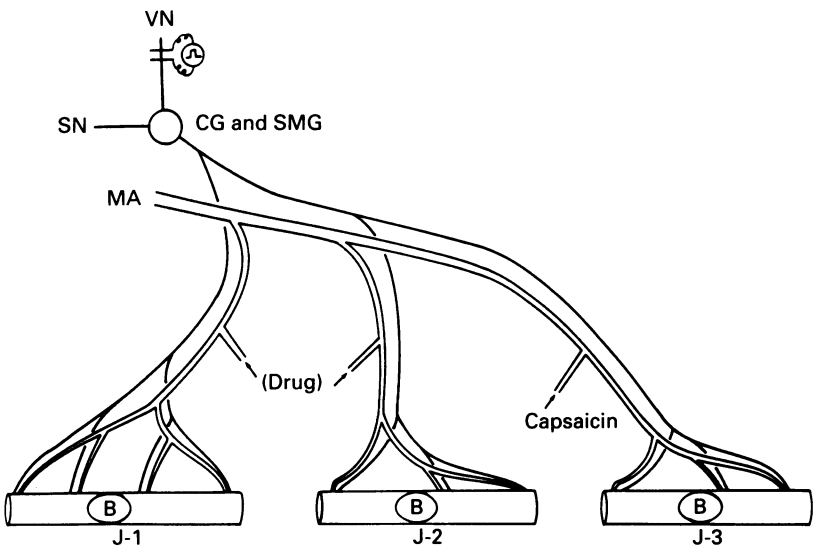

Fig. 1. Schematic drawing illustrating the arrangement for recording of intestinal motility and for administration of drugs. Jejunal motility was recorded as pressure change by intraluminal balloons (B) introduced into the isolated loops (J-1 to J-3). One loop was used for capsaicin infusion, and the others were used for observation of the reflex effect induced by capsaicin. In some experiments, drugs were administered to loops other than the capsaicin-infused loop. Vagal stimulation was performed at the neck. VN, vagus nerve; SN, splanchnic nerve; CG and SMG, coeliac and superior mesenteric ganglia; MA, mesenteric artery.

A 6–8 cm long intestinal segment, with a suitable distribution of mesenteric vessels for the experimental arrangement shown in Fig. 1, was separated from the rest of the small intestine. The most centrally situated branch of the main mesenteric artery distributing to the loop was cannulated with a thin polyethylene tube (24G) for administration of drugs into the blood stream of the main mesenteric artery. After the luminal contents of the separated loop were thoroughly washed out with warm physiological saline, a 3 cm long rubber balloon was inserted into the lumen

from the oral end of the loop, and a vinyl tube for evacuation of secretory solution was placed at the anal end. Three to six of these separated loops with balloons were prepared. The intraballoon pressure during the relaxed intestinal phase was set at 0·8 kPa by infusing water into the balloon through a water manometer, which was connected to a low-range pressure transducer (Nihon Koden, LPU-0·1A). The pressure change of the balloon due to motility of the intestine was recorded on a pen-oscillograph (San-Ei, Recticorder-8S).

The right cervical vagus nerve trunk was exposed from the sheath and cut. The peripheral cut end of the vagus nerve was stimulated by a bipolar platinum electrode in a paraffin pool. Electrical stimulation with pulses of 10–20 Hz, 0·2–2·0 ms and 5–15 V was applied for 15–30 s at 5·5 min intervals.

Capsaicin (10–100 μM) was intra-arterially administered to only one separated loop through the mesenteric artery by means of an infusion pump (Truth, A-II) at the rate of 0·07 or 0·14 ml min^{-1}. This was done so that drugs would reach the mesenteric blood flow within 2–3 min after the infusion began, because of a 0·21 ml dead space of the cannula. To investigate the pharmacological properties, some blocking agents were intra-arterially administered as well as capsaicin. These drugs preceded the administration of capsaicin by 5 min.

Vagotomy was performed by cutting the remaining left cervical vagus nerve. Splanchnic nerve section was done extraperitoneally by means of bilateral division of the major and minor splanchnic nerves and bilateral extirpation of sympathetic chains from the 13th thoracic to 4th lumbar ganglia. Spinal cord transection and decerebration were carried out at the 5th and 6th cervical cord and at the mid-collicular level, respectively.

Distension of 10 cm long intestinal loops with 20–30 ml air was carried out in some experiments of spinal cord transection and decerebration. The pressure produced by distension ranged from 10 to 27 kPa.

Electrical activity was recorded from the mesenteric nerve and major splanchnic nerve by a bipolar platinum electrode in a paraffin pool. The central or peripheral cut end of the nerves was gently divided with fine forceps. Then, the units which responded to distension (10–15 ml air, 5–17 kPa) of suitable loops were identified: capsaicin was then administered while recording afferent activity. Control rates of discharge were averaged for the 1 min just before injection of capsaicin. Peak rates of discharge were averaged for 12 s if the fibre was stimulated. All rates of discharge are expressed as impulses per second.

An adequate level of anaesthesia was maintained throughout the experiments. This was assured by measuring the blood pressure continuously. Intra-arterial injection of capsaicin produced an increase in blood pressure of 10 mmHg in half the animals and no change in the others (see also Longhurst *et al.* 1984*a*). Furthermore in three experiments (not included in the present series), in which dogs were similarly anaesthetized but not paralysed, injections of capsaicin caused no movements that might be interpreted as responses to pain.

The drugs that were used included ketamine hydrochloride (Ketaral, Sankyo), urethane (Kanto), gallamine triethiodide (Sigma), atropine sulphate (Merck), hexamethonium bromide (Sigma), capsaicin (Sigma), phentolamine methanesulphonate (Regitin, Ciba), yohimbine hydrochloride (Sigma), prazosin hydrochloride (Tokyo Kasei), propranolol hydrochloride (Inderal, ICI), noradrenaline hydrochloride (Sigma), clonidine hydrochloride (Tokyo Kasei) and phenylephrine hydrochloride (Sigma). A stock solution of capsaicin (10 mM) was prepared with a mixture of 10 ml Tween 80, 10 ml ethanol and 80 ml physiological saline and then diluted with Tyrode solution before the experiments. The other drugs were dissolved in 0·9% saline.

Statistical comparisons were made using standard Student's paired and unpaired *t* tests to test for the significance of inhibitions caused by capsaicin application and to detect significant differences of the inhibition between treated and non-treated loops. All the data in the text are expressed as mean ± S.E.M. of the percentages of the response height to the control, and *n* represents the number of intestinal loops from which the data were obtained.

RESULTS

Vagus nerve stimulation (10–20 Hz, 0·2–2·0 ms, 5–20 V) at 5·5 min intervals was applied to induce a contraction which developed approximately maximal pressure in every loop and which was fairly constant in response height in each of the loops. The

vagally induced contraction was inhibited to 36·5 ± 13·5 % of the control ($P < 0·01$, $n = 5$) by intra-arterial infusion of atropine (7 nmol min^{-1}, for 5 min) and reduced to 13·9 ± 1·8 % ($P < 0·001$, $n = 5$) by hexamethonium (7–70 nmol min^{-1}, for 5 min). These inhibitions were produced only in the drug-infused loops, but not in the other non-infused loops.

Effects of capsaicin on vagally induced contraction

At an infusion rate of 0·7–14 nmol min^{-1} for 5–10 min, capsaicin inhibited the vagally evoked contraction not only in the capsaicin-infused loop (7·9 ± 2·3 % of the control, $P < 0·001$, $n = 33$) but also in the other non-infused loops (26·6 ± 5·5 % of the control, $P < 0·001$, $n = 26$) (Fig. 2). The inhibitory action lasted for 8·6 ± 1·2 min

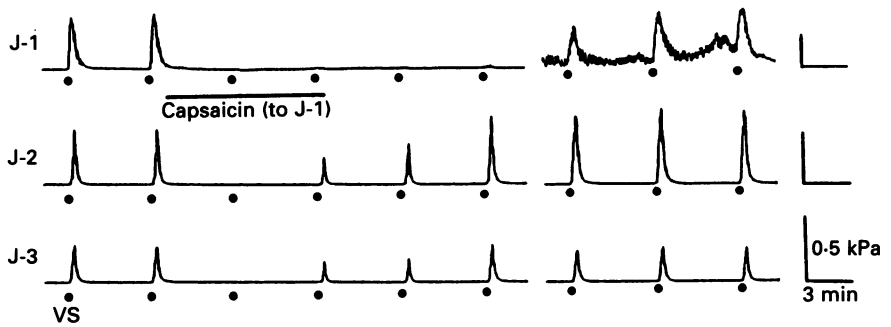

Fig. 2. Typical tracings showing the inhibitory action of capsaicin on the jejunal contraction induced by vagus nerve stimulation (VS; 10 Hz, 1 ms, 15 V, for 30 s, at 5·5 min intervals) at the capsaicin-infused loop (J-1) and non-infused loops (J-2 and J-3). The bar under the J-1 recording shows the intra-arterial infusion of capsaicin (0·7 nmol min^{-1}) to the J-1 loop. The right panel shows the recordings 40 min after the termination of capsaicin infusion.

($n = 32$) in the non-infused loops after the termination of the drug infusion, while in the capsaicin-infused loop it lasted for 49·0 ± 6·9 min ($n = 28$). Repeated applications of capsaicin on the same loop at intervals of less than 2 h could not produce significant inhibition in the non-infused loops, but could in the capsaicin-infused loop. An interval of more than 3 h was required to reproduce the inhibitory action on the same loop. However, the inhibition could be induced not only in the loops not previously treated with capsaicin but also in the loops which had previously been exposed to capsaicin, when capsaicin was administered to one of the non-treated loops within 2 h after the first application of capsaicin. Therefore, the loop to which capsaicin was applied was in turn changed in most animals. Intra-arterial infusion of a solvent of 100 μM-capsaicin (0·07–0·14 ml min^{-1}), 0·1 % Tween 80, 0·1 % ethanol–Ringer solution, had no significant effect.

In the following study, a mechanism of the inhibitory action of capsaicin on the vagally evoked contraction in the non-infused loops was determined.

Effects of extrinsic nerve sectionings

In dogs vagotomized bilaterally, capsaicin reduced any vagally evoked contraction, which was not significantly different from the inhibition in intact dogs

(Table 1). Application of capsaicin also produced an inhibition of vagally evoked contractions in the dog which had previously had the bilateral lumbar sympathetic nerves severed and the sympathetic chains extirpated (T13–L4). After successive division of the bilateral major and minor splanchnic nerves, the inhibitory response to capsaicin was abolished (Fig. 3), although the unilateral vagus remained intact. As shown in Table 1, splanchnicotomy and vago-splanchnicotomy abolished the inhibitory action of capsaicin significantly ($P < 0{\cdot}001$ in both). The results indicate that capsaicin activates the sympathetic intestinointestinal reflex.

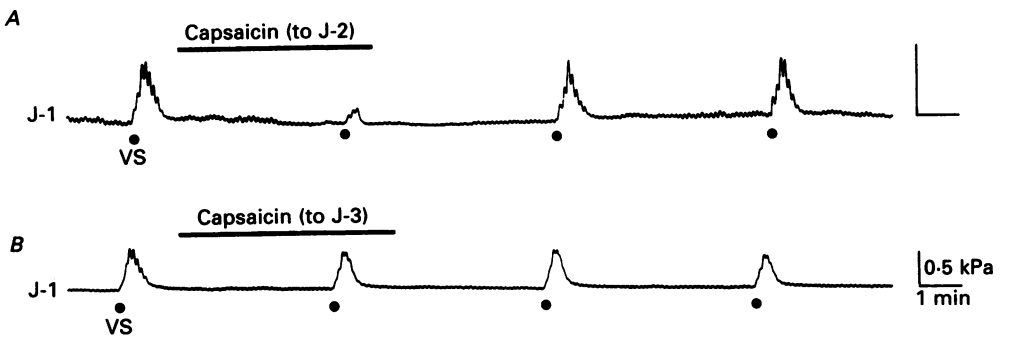

Fig. 3. Typical tracings showing the effect of bilateral splanchnic nerve section on the inhibitory action of capsaicin (1·4 nmol min^{-1}) on jejunal contractions (J-1) induced by vagus nerve stimulation (VS; 10 Hz, 1 ms, 8 V, for 15 s, at 5·5 min intervals). Sectioning of the right cervical vagus (for the stimulation) and extirpation of the bilateral sympathetic chains (T13–L4) had been previously performed (*A*). Successive division of the bilateral major and minor splanchnic nerves abolished the inhibitory action of capsaicin (*B*). Capsaicin was infused to J-2 loop in *A* and to J-3 loop in *B*.

Effects of cervical spinal cord transection

The spinal cord was transected at the 5th or 6th cervical level prior to or during the experiment, in order to examine the location of the centre of the inhibitory reflex by capsaicin. The inhibition of vagally evoked contractions by capsaicin was abolished by cervical spinal cord transection ($P < 0{\cdot}001$, Table 1), while the inhibition produced by distension with 20–30 ml air was unaffected by spinal cord transection. To avoid any influence of spinal shock, the experiment was performed more than 3·5 h after spinal cord transection.

Effects of decerebration

Decerebration was carried out at the mid-collicular level after recording the control response to capsaicin. The inhibition of vagally evoked contractions by capsaicin was obtained even after decerebration ($P < 0{\cdot}001$). However, the inhibition by capsaicin was significantly reduced when compared to that in the intact dog ($P < 0{\cdot}01$, Table 1). The inhibition by distension of vagally evoked contractions was unaffected by decerebration.

Effects of adrenergic blocking agents

Adrenergic blocking agents were intra-arterially administered to loops other than the capsaicin-infused loop (Fig. 1). As shown in Table 2, the inhibition of vagally evoked contractions by capsaicin did not occur in the loop which was treated with the α-adrenoceptor blocking agent phentolamine (70 nmol min^{-1}) ($P < 0.01$), although it did in the loop which was treated with the β-adrenoceptor blocking agent,

TABLE 1. Effects of capsaicin on vagally evoked contractions in each operated dog

Operation	Contraction (%)	n	Probability
Intact	26·6±5·5	26 (10)	$P < 0.001$
Vagotomy	31·5±3·1	61 (21)	$P < 0.001$
Splanchnicotomy	88·2±9·9	10 (3)	n.s.
Vago-splanchnicotomy	98·8±7·1	11 (3)	n.s.
Spinal cord transection	90·0±8·8	15 (4)	n.s.
Decerebration	52·0±6·9	36 (4)	$P < 0.001$

The data are expressed as mean±S.E.M. of the percentage of the amplitude of vagally evoked contractions to the control. n indicates the number of loops used in the experiment; the number in parentheses shows the number of animals. The probability was calculated by Student's paired t test against the height of the vagally evoked contraction before capsaicin infusion.

TABLE 2. Effects of capsaicin on vagally evoked contractions in the loop pre-treated with adrenergic blocking agents

Drugs	Contraction (%)	n	Probability
Non-treated loop	11·4±3·1	15 (11)	$P < 0.001$
Phentolamine	92·7±27·1	3 (3)	n.s.
Prazosin	3·8±1·9	5 (5)	$P < 0.001$
Yohimbine	78·6±8·3	4 (4)	n.s.
Propranolol	10·0±8·6	3 (3)	$P < 0.01$

The data are expressed as mean±S.E.M. of the percentage of the amplitude of vagally evoked contractions to the control. n indicates the number of loops used in the experiment; the number in parentheses shows the number of animals. The probability was calculated by Student's paired t test against the height of the vagally evoked contraction before capsaicin infusion.

propranolol (28–70 nmol min^{-1}) as well as in the non-treated loop. The α_2-adrenoceptor blocking agent yohimbine (7–70 nmol min^{-1}) abolished the inhibitory response to capsaicin ($P < 0.001$), but the α_1-adrenoceptor blocking agent prazosin (0·7–28 nmol min^{-1}) did not inhibit it (Fig. 4). Phentolamine (70 nmol min^{-1}) and prazosin (0·7 nmol min^{-1}) abolished the inhibition of vagally evoked contractions by noradrenaline and phenylephrine infused in the same loop (10 nmol in bolus), respectively. Yohimbine (7 nmol min^{-1}) reduced the inhibition by the α_2-adrenoceptor agonist clonidine (1 nmol in bolus).

Electrical activity of mesenteric efferents induced by capsaicin

Electrical efferent activity of the mesenteric nerve innervating the jejunal loop whose contractile activity was simultaneously monitored was recorded as multiunit

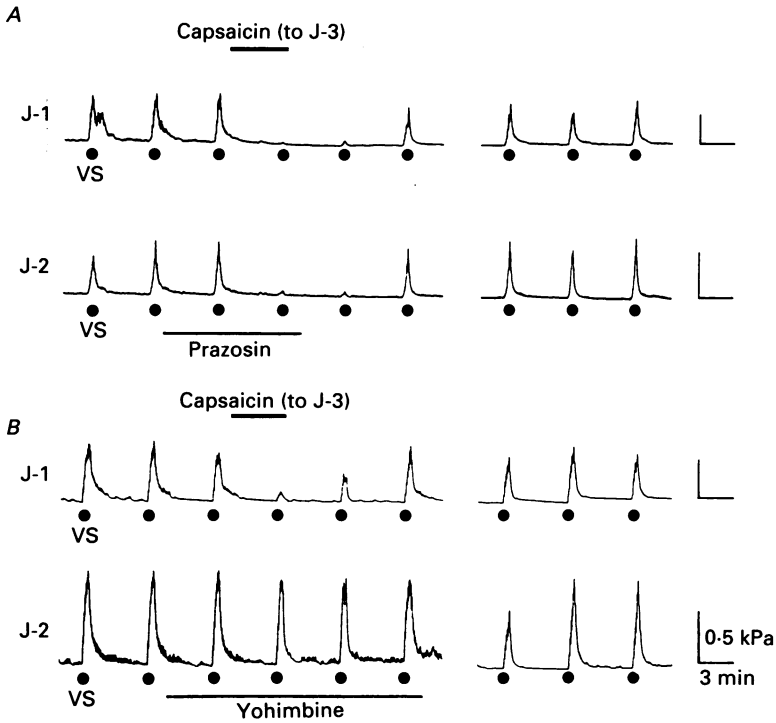

Fig. 4. Typical tracings showing the effect of prazosin (0·7 nmol min^{-1}, in *A*) and yohimbine (70 nmol min^{-1}, in *B*) on the inhibitory action of capsaicin (1·4 nmol min^{-1}) to jejunal contractions induced by vagus nerve stimulation(VS; 10 Hz, 1 ms, 5 V, for 30 s in *A* and 10 Hz, 1 ms, 10 V, for 30 s in *B*). Bilateral vagus nerves had previously been severed. Capsaicin was infused to J-3 loop in *A* and *B*. J-1 recordings in *A* and *B* show control at non-infused loops. Right panels show recordings 30 min after termination of capsaicin infusion.

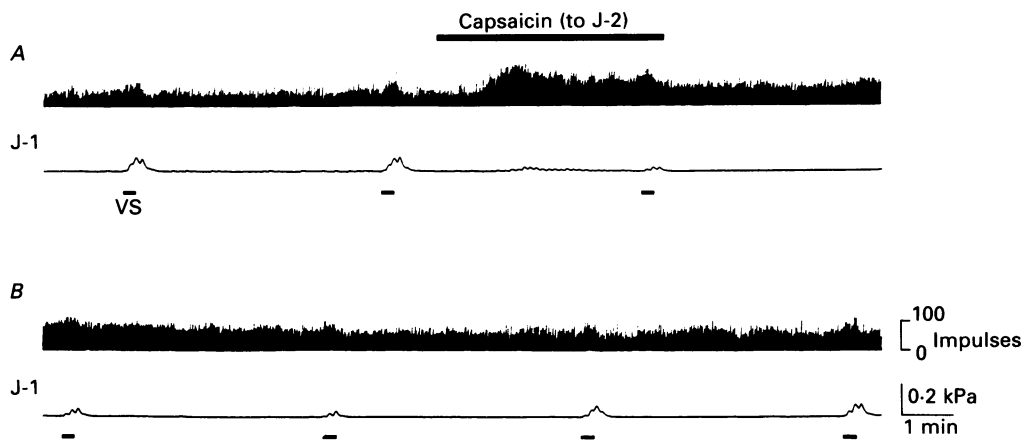

Fig. 5. Typical tracings showing the effect of capsaicin (1·4 nmol min^{-1}) on spontaneous and vagally evoked discharges of mesenteric efferents and motility recorded simultaneously. Electrical activity was recorded as multiunit discharges. Division of the bilateral vagus and extirpation of the sympathetic chains (T13–L4) had previously been performed. Spontaneous activity of the mesenteric efferents was responsive to capsaicin infused to J-2 loop with an increase in the frequency of the spikes, during which period the vagally induced contractions were reduced. *A* and *B* are continuous recording. VS: vagus nerve stimulation (10 Hz, 1 ms, 8 V, for 15 s).

discharges from the central cut end of one of the mesenteric nerve bundles running along the mesenteric artery. Spike frequency increased during efferent stimulation of the vagus and/or the major splanchnic nerve.

In the vagotomized dog, capsaicin caused an increase in the frequency of the spike activity (6·2 ± 1·0 times, $n = 15$) of the mesenteric efferents during infusion of the

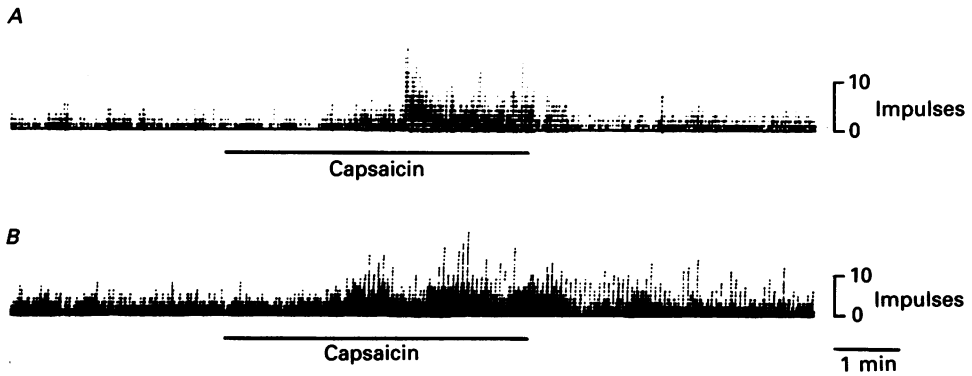

Fig. 6. Typical tracings showing the effects of capsaicin on electrical activity of mesenteric and major splanchnic afferents. *A*, single-unit discharge of the mesenteric afferents responding to capsaicin (7 nmol min^{-1}) infused to the loop which was supplied by the recorded mesenteric nerve. *B*, single-unit discharge of the major splanchnic afferents responding to capsaicin (1·4 nmol min^{-1}) infused to the loop which was responsive to distension.

drug with some delay. The vagally evoked contractions in the loop were inhibited by capsaicin during the increase of the mesenteric efferent activity (Fig. 5). The increase of the efferent activity by capsaicin disappeared after bilateral splanchnic nerve section. An increase by vagus nerve stimulation of mesenteric efferent activity was not affected by capsaicin, indicating that capsaicin at the concentration that was used did not have any conduction blocking action on the nerve (Jancsó & Such, 1983; Baranowski, Lynn & Pini, 1986).

Electrical activity of splanchnic and mesenteric afferents induced by capsaicin

Afferent electrical activity was recorded to confirm the afferent limb of the intestinointestinal inhibitory reflex induced by capsaicin. A single-unit discharge responding to distension of an intestinal loop was recorded from the peripheral cut end of the major splanchnic nerve, in order to detect the unit with receptive field on the loop to apply capsaicin. Eight out of nine units were responsive to infusion of capsaicin into the same loop as the distended one (Fig. 6*B*), but one unit was not responsive to capsaicin. The frequency of the spikes increased from 1·1 ± 0·2 to 5·3 ± 1·2 impulses s^{-1} ($n = 8$, $P < 0·01$) by capsaicin infusion and the increase continued for 5–15 min. Afferent discharge of the mesenteric nerve was also recorded as a single unit. Five units responding to distension were also responsive to capsaicin infusion with an increase in frequency of the spike activity (from 0·7 ± 0·2 to 6·0 ± 1·7 impulses s^{-1}, $P < 0·02$) (Fig. 6*A*). Because of difficulty of repeated application of capsaicin, the unit that did not respond to distension was not examined further.

DISCUSSION

Although intra-arterial infusions of hexamethonium and atropine reduced vagally evoked contractions of the canine small intestine only in the drug-infused loop, capsaicin inhibited the contraction not only in the capsaicin-infused loop but also in the other non-infused loops. This inhibitory action of capsaicin on the non-infused loops was simultaneously produced with a similar duration and extent in all the prepared loops. In addition, capsaicin should activate sensory nerves in the infused loop, since a second application of capsaicin within 2 h in the same loop did not produce respective inhibition, indicating desensitization which is characteristic of the specific action of capsaicin on sensory nerves (Maggi, Meli & Santicioli, 1987; Maggi & Meli, 1988). It seems therefore that the inhibitory response to capsaicin in the non-infused loop is due to activation of the extrinsic intestinointestinal reflex.

The inhibitory action of capsaicin on vagally evoked contractions in the non-infused loops was abolished by bilateral splanchnic nerve section but was unaffected by bilateral vagotomy. This finding suggests that capsaicin activates a reflex similar to the intestinointestinal reflex evoked by distension (Bayliss & Starling, 1899; Hermann & Morin, 1934; Chang & Hsu, 1942; Hukuhara, Nakayama, Yamagami & Miyake, 1959). This was also supported by the results of the recording of electrical activity from the mesenteric and splanchnic nerves. An increase in single-unit discharges of the major splanchnic and mesenteric afferents was induced by capsaicin application. In the vagotomized dog, capsaicin increased the frequency of multiunit discharges of mesenteric efferents, during which time vagally evoked contractions recorded simultaneously in the same loop were inhibited. Furthermore, the mesenteric efferent activity was not increased by capsaicin application after splanchnic nerve section. The sympathetic intestinointestinal reflex induced by capsaicin has been presumed by findings that transient relaxation of the distal colon by systemic administration of capsaicin was reduced by tetrodotoxin, hexamethonium or guanethidine as well as by systemic capsaicin desensitization in guinea-pigs (Maggi *et al.* 1987), and that a delay of gastrointestinal propulsion in capsaicin-treated rat was abolished by guanethidine (Holzer, 1986).

The centre of the intestinointestinal inhibitory reflex by capsaicin could be located supraspinally, since the reflex persisted in decerebrated dogs but was abolished in dogs that had undergone cervical spinal cord transection. However, the inhibitory reflex induced by distension persisted in dogs that had undergone decerebration and spinal cord transection, indicating a spinal reflex centre as suggested in early reports (Morin & Vial, 1934; Chang & Hsu, 1942). It appears that the difference of location between the capsaicin- and distension-induced reflex centre is due to the size and the number of sensory afferents stimulated by them: there was no significant difference between A and C fibre afferents on the threshold to distension (2–13 kPa) of the feline colon (Blumberg, Haupt, Jänig & Kohler, 1983). Capsaicin stimulated all C fibres but stimulated only 38% of A fibres which responded by mechanical stimulations (Longhurst, Kaufman, Ordway & Musch, 1984*b*). It is therefore possible that capsaicin could stimulate most C fibres and a small number of A fibres, although afferent units of splanchnic nerve activated by both distension (5–17 kPa) and capsaicin in the present experiment may be both A and C fibres or only A fibres.

Taken together, it is likely that the inhibitory reflex caused by capsaicin was mainly mediated through sensory C fibres and supraspinal centres but partially through A fibre afferents and the spinal centre, and that the reflex elicited by distension was mediated through both centres. However, the reflex effect through supraspinal centres by distension may always be masked by the reflex effect through the spinal centre. In contrast, the reflex evoked by capsaicin through the spinal cord may not be sufficient to induce a significant inhibition of the vagally evoked contractions because vagus efferents were fully activated by electrical stimulation, so that, the reflex effect was apparently abolished after the spinal cord transection. The centre of most reflexes, e.g. the depressor cardiovascular reflex from the jejunum (Lembeck & Skofitsch, 1982), the femoral artery (Donnerer & Lembeck, 1983) and the urinary bladder (Giuliani *et al.* 1988), and the micturition reflex from the urinary bladder (Maggi *et al.* 1986), induced by activation of sensory C fibres by capsaicin is located in the supraspinal structure.

In the present results, it is suggested that the location of the supraspinal reflex centre is in both the brain stem and higher brain, because of the intermediate inhibitory response to capsaicin after decerebration (Table 1).

The inhibitory response to capsaicin on the vagally evoked contraction which was sensitive to atropine, hexamethonium and noradrenaline was diminished by pretreatment with phentolamine or yohimbine, but was unaffected by prazosin or propranolol. In the isolated guinea-pig ileum, α_2-adrenoceptor agonists inhibit acetylcholine release, resulting in a reduction of the amplitude of electrically evoked contractions, and these inhibitions are antagonized by yohimbine (Kilbinger & Wessler, 1979; Andréjak, Pommier, Mouillé & Schmitt, 1980; Fagbemi & Salako, 1980). Yohimbine also antagonizes the inhibition of field-stimulated contractions induced by peri-arterial nerve stimulation (Fagbemi & Salako, 1980). Such presynaptic inhibition was also reported in the isolated canine small intestine (Nakahata, Nakanishi & Suzuki, 1982). Therefore, the present results suggest that inhibition of vagally induced contractions by capsaicin is due to a presynaptic inhibition via α_2-adrenoceptors by activation of the sympathetic reflex.

Capsaicin also stimulates sensory neurones antidromically, as a result of which the released tachykinins excite intestinal motility and calcitonin gene-related peptide inhibits intestinal motility by activation of non-adrenergic, non-cholinergic inhibitory neurones or by direct action on the smooth muscles. Furthermore, capsaicin itself inhibits the motility by acting directly on the smooth muscle cells (see reviews by Buck & Burks, 1986 and Maggi & Meli, 1988). In the present results, the inhibitory response to capsaicin of the vagally evoked contraction in the capsaicin-infused loop was larger and longer lasting than the reflex response in the non-infused loop. This response in the capsaicin-infused loop may be due to a sum of the various inhibitory actions described above.

In conclusion, capsaicin activates the canine sympathetic intestinointestinal reflex whose centre is located in the supraspinal region, which is different from the intestinointestinal reflex activated by distension. The inhibition by capsaicin of vagally evoked contractions is due to presynaptic inhibition via α_2-adrenoceptors, which are activated by the reflex.

REFERENCES

ANDRÉJAK, M., POMMIER, Y., MOUILLÉ, P. & SCHMITT, H. (1980). Effects of some α-adrenoceptor agonists and antagonists on the guinea-pig ileum. *Naunyn-Schmiedeberg's Archives of Pharmacology* **314**, 83–87.

BARANOWSKI, R., LYNN, B. & PINI, A. (1986). The effects of locally applied capsaicin on conduction in cutaneous nerves in four mammalian species. *British Journal of Pharmacology* **89**, 267–276.

BAYLISS, W. M. & STARLING, E. H. (1899). The movements and innervation of the small intestine. *Journal of Physiology* **24**, 99–143.

BLUMBERG, H., HAUPT, P., JÄNIG, W. & KOHLER, W. (1983). Encoding of visceral noxious stimuli in the discharge patterns of visceral afferent fibres from the colon. *Pflügers Archiv* **398**, 33–40.

BUCK, S. H. & BURKS, T. F. (1986). The neuropharmacology of capsaicin: review of some recent observations. *Pharmacological Reviews* **38**, 179–226.

CHANG, P. Y. & HSU, F. Y. (1942). Localization of intestinal inhibitory reflex arc. *Quarterly Journal of Experimental Physiology* **31**, 311–318.

DONNERER, J. & LEMBECK, F. (1983). Capsaicin-induced reflex fall in rat blood pressure is mediated by afferent substance P-containing neurones via a reflex centre in the brain stem. *Naunyn-Schmiedeberg's Archives of Pharmacology* **324**, 293–295.

FAGBEMI, S. O. & SALAKO, L. A. (1980). The effect of prazosin on the guinea-pig ileum. *British Journal of Pharmacology* **70**, 395–402.

GIULIANI, S., MAGGI, C. A. & MELI, A. (1988). Capsaicin-sensitive afferents in the rat urinary bladder activate a spinal sympathetic cardiovascular reflex. *Naunyn-Schmiedeberg's Archives of Pharmacology* **338**, 411–416.

HERMANN, H. & MORIN, G. (1934). Mise en évidence d'un réflexe inhibiteur intestino-intestinal. *Comptes rendus hebdomadaires des séances et mémoires de la Société de biologie et de ses filiales et associées* **115**, 529–531.

HOLZER, P. (1986). Capsaicin-sensitive afferent neurones and gastrointestinal propulsion in the rat. *Naunyn-Schmiedeberg's Archives of Pharmacology* **332**, 62–65.

HUKUHARA, T., NAKAYAMA, S., YAMAGAMI, M. & MIYAKE, T. (1959). On the intestinal extrinsic reflexes elicited from the small intestine. *Acta medica okayama* **13**, 113–121.

JANCSÓ, G. & SUCH, G. (1983). Effects of capsaicin applied perineurally to the vagus nerve on cardiovascular and respiratory functions in the cat. *Journal of Physiology* **341**, 359–370.

KILBINGER, H. & WESSLER, I. (1979). Increase by α-adrenolytic drugs of acetylcholine release evoked by field stimulation of the guinea-pig ileum. *Naunyn-Schmiedeberg's Archives of Pharmacology* **309**, 255–257.

LEMBECK, F. & SKOFITSCH, G. (1982). Visceral pain reflex after pretreatment with capsaicin and morphine. *Naunyn-Schmiedeberg's Archives of Pharmacology* **321**, 116–122.

LONGHURST, J. C., ASHTON, J. H. & IWAMOTO, G. A. (1980). Cardiovascular reflexes resulting from capsaicin-stimulated gastric receptors in anesthetized dogs. *Circulation Research* **46**, 780–788.

LONGHURST, J. C., KAUFMAN, M. P., ORDWAY, G. A. & MUSCH, T. I. (1984*b*). Effects of bradykinin and capsaicin on endings of afferent fibers from abdominal visceral organs. *American Journal of Physiology* **247**, R552–559.

LONGHURST, J. C., STEBBINS, C. L. & ORDWAY, G. A. (1984*a*). Chemically induced cardiovascular reflexes arising from the stomach of the cat. *American Journal of Physiology* **247**, H459–466.

MAGGI, C. A. & MELI, A. (1988). The sensory-efferent function of capsaicin-sensitive sensory neurons. *General Pharmacology* **19**, 1–43.

MAGGI, C. A., MELI, A. & SANTICIOLI, P. (1987). Four motor effects of capsaicin on guinea-pig distal colon. *British Journal of Pharmacology* **90**, 651–660.

MAGGI, C. A., SANTICIOLI, P., BORSINI, F., GIULIANI, S. & MELI, A. (1986). The role of the capsaicin-sensitive innervation of the rat urinary bladder in the activation of micturition reflex. *Naunyn-Schmiedeberg's Archives of Pharmacology* **332**, 276–283.

MORIN, G. & VIAL, J. (1934). Sur les voies et les centres du réflexe inhibiteur intestino-intestinal. *Comptes rendus hebdomadaires des séances et mémoires de la Société de biologie et de ses filiales et associées* **116**, 536–538.

NAKAHATA, N., NAKANISHI, H. & SUZUKI, T. (1982). Prejunctional inhibition mediated via α_2-adrenoceptors in excitatory transmission of canine small intestine. *Journal of Pharmacy and Pharmacology* **34**, 510–513.

ORDWAY, G. A. & LONGHURST, J. C. (1983). Cardiovascular reflexes arising from the gallbladder of the cat: effects of capsaicin, bradykinin, and distension. *Circulation Research* **52**, 26–35.

ORDWAY, G. A., LONGHURST, J. C. & MITCHELL, J. H. (1983). Stimulation of pancreatic afferents reflexly activates the cardiovascular system in cats. *American Journal of Physiology* **245**, R820–826.