J. Pharm. Pharmacol. 1990, 42: 546–552
Received December 4, 1989

Effects of Fedotozine on Gastrointestinal Motility in Dogs: Mechanism of Action and Related Pharmacokinetics

X. PASCAUD, C. HONDE, B. LE GALLOU, F. CHANOINE, F. ROMAN, L. BUENO* AND J. L. JUNIEN

*Jouveinal Laboratoires, 1 rue des Moissons, B.P. 100, 94265 Fresnes Cedex, France and * Laboratoire de Pharmacologie, I.N.R.A., 180 chemin de Tournefeuille, B.P.3, 31931 Toulouse Cedex, France*

Abstract—The effects of fedotozine, (+)-(1*R*)-1-phenyl-1-[(3,4,5-trimethoxy)benzyloxymethyl]-*N*,*N*-dimethyl-n-propylamine, on motility of the antrum and small intestine were investigated in dog. In fasted dogs, following i.v. administration, fedotozine at 1 and 2 mg kg^{-1} stimulated and at 5 mg kg^{-1} inhibited antral motility. Between 1 to 5 mg kg^{-1}, fedotozine exhibited a sustained and potent stimulatory effect on the small intestine inducing 1 to 4 phases III of the migrating motor complex (MMC) lasting up to 32 min in the duodenum and migrating to the jejunum. Following oral administration, fedotozine at 2·5 and 5 mg kg^{-1} constantly stimulated both antrum and small intestinal motility. In fed dogs, fedotozine i.v. (2 mg kg^{-1}) increased antral motility and induced phase III of MMC in the place of postprandial pattern. Naloxone (0·3 mg kg^{-1} i.v.) and naloxone methylbromide (2 mg kg^{-1} i.v.) inhibited the stimulatory effects of fedotozine on gastrointestinal motility indicating a peripheral opiate site of action of the drug whereas phentolamine, hexamethonium, propranolol and methysergide were inactive. In-vitro fedotozine showed submicromolar affinity for opiate receptors with a weak specificity for the μ-receptors in guinea-pig brain and myenteric plexus preparations. Plasma concentrations in dogs receiving fedotozine administered orally at 2·5 mg kg^{-1} (and in all dogs except one at 5 mg kg^{-1}) were below the detection limit (< 20 ng g^{-1}). In contrast, tissue concentrations in the muscle and mucosal layers of the gut were above 1 $\mu g\ g^{-1}$. Concentrations in gastric juice after i.v. injection of fedotozine were 10 to 20 times higher than in the plasma, indicating passage of the drug from the blood into the lumen. These results indicate that fedotozine between 1 to 5 mg kg^{-1} orally or i.v. stimulates gastrointestinal motility in dog through peripheral opiate receptors. This peripheral activity is related to a high distribution of the drug at the target organ.

The pattern of the digestive tract motility differs during the fed and the fasted state. In dogs, migrating motor complex (MMC) which characterize the fasted state (Szurszewski 1969) is replaced after feeding by a period of irregular contractile activity lasting several hours (Ruckebusch & Bueno 1976). Abnormal motor profile and, more precisely, a lack of phase III of the MMC has been reported in some pathological situations including bacterial overgrowth (Vantrappen et al 1977), intestinal pseudo obstruction (Summers et al 1983), gastric ulcers and duodenogastric reflux (Miranda et al 1985), diabetic gastroparesis (Abel et al 1985), diarrhoea (El Newihi et al 1985) and in patients suffering from irritable bowel syndrome under stress (Kumar & Wingate 1985). Opiates seem to play a physiological role in controlling gastrointestinal motility through μ and/or δ brain and peripheral receptors of the gut. Opiate drugs and endogenous opioids induce phase III-like activity in the fasted state and inhibit antral motility (Bueno & Ruckebusch 1978; Bueno et al 1985; Telford et al 1985). Whereas the stimulating effect of opioid drugs at the small bowel level seems to be locally mediated through μ-receptors (Fioramonti et al 1984; Sarna & Lang 1985; Telford et al 1985), their effects on antral motility appears to involve a central component (Bueno et al 1985).

This paper reports the effect of fedotozine [(+)-(1*R*)-1-phenyl-1-((3,4,5-trimethoxy)benzyloxymethyl)-*N*,*N*-dimethyl-n-propylamine, J01196] on digestive tract motility in dog. The results show that this drug activates gastrointestinal motility through peripheral opiate receptors which correlates with a high and specific distribution of the drug in the gut wall.

Correspondence to: X. Pascaud, Jouveinal Laboratoires, B.P. 100, 94265 Fresnes Cedex, France.

Materials and Methods

In-vivo studies

Ten mongrel dogs (5 males and 5 females), 8–22 kg, devoid of any apparent digestive disorder received a daily meal of a standard dog food (Viandoriz, Ets Bourgoin, France) at 1700 h for the studies carried out in fasted animals, or at 1130 h when tests were carried out during the postprandial period.

Ni/Cr electrodes (120 μm diameter, Johnson Matthey, London, UK) and strain gauge transducers (EA.06.090DH.350 SE from Micromeasurement, Raleigh, North Carolina, USA) were prepared as described by Pascaud et al (1978). Under halothane anaesthesia (Fluothane ND, Specia Labs, France) the electrodes were implanted within the muscular layer according to Ruckebusch (1973) and the strain gauge transducers were sewn on the serosa (Tevdec 4/10) to the circular muscle layer.

The strain gauge transducers were placed on the antrum (6 cm above the pylorus) on the duodenum at 5 and 10 cm from the pylorus, on the jejunum at 20 cm below the ligament of Treitz. Two groups of electrodes were placed at 5 cm below the strain-gauge transducers on the antrum, the duodenum and the jejunum and another group was also implanted in the ileum at 20 cm above the ileo-caecal junction. Animals were allowed a two week rest before experimentation.

Recordings (8-channel recorder Dynograph R612, Beck-

man, USA) were obtained through a 9853A voltage/pulse/pressure coupler allowing the selection of either the electric signal given by electrodes or the mechanical activity of the strain gauge transducers which receive a 6 volt excitation current from the Dynograph. Each transducer was routinely calibrated for a 5–25 g tension. The signals produced by the strain gauge transducers were simultaneously recorded on a potentiometric 4 channel pen recorder (Linseis, Bioblock L6510, Strasbourg, France).

Experimental schedule. Recordings were begun at 0900 h. In a first series of experiments, fedotozine tartrate was dissolved in 0·9% NaCl (saline) for i.v. injection and given in a capsule for oral administration. The drug was given 10 min after the occurrence of a phase III of the MMC in the duodenum, or 2 h after a meal. The drug was injected either as a bolus or perfused at 10 mg kg^{-1} at a constant rate of 1 mL min^{-1} for 30 min.

In a second series of experiments, the mechanism of action was investigated by injecting putative antagonists intravenously 5 min before fedotozine infusion was begun. The antagonists used were naloxone HCl (0·30 mg kg^{-1}), naloxone methylbromide (2 mg kg^{-1}), atropine sulphate (0·10 mg kg^{-1}), hexamethonium (2·5 mg kg^{-1}), phentolamine (0·10 mg kg^{-1}), propranolol (0·10 mg kg^{-1}) and methysergide (10 μg kg^{-1}) to inhibit opiate receptors, M1 muscarinic, nicotinic, α- and β-adrenergic, and 5-HT receptors, respectively.

Data analysis. Visual analysis of electromyograms included evaluation of the characteristics of the motor profile and changes thereof, percentage of slow waves with spikes, frequency of the MMC cycle, and duration of each phase of the MMC. The mechanical activity was determined as a motility index calculated according to Walker et al (1972). Amplitude of contractions was scored as the maximal contractile amplitude, i.e. the maximum of contractions occurring during phase III was divided into 4 equal parts (class 4 being defined as the contractions of the highest amplitude). The motility index was then calculated (numbers of contractions x value of the class). Statistical comparisons of the various parameters were carried out using Student's *t*-test.

Binding studies

The capacity of fedotozine to displace opiate ligands from their receptors was studied on whole brain and myenteric plexus membrane preparations of male tricoloured guinea-pigs (Coblabo-Iffigniac, France), 300–325 g. Food but not water was withdrawn for 24 h before decapitation. The whole brain membranes were prepared according to Kosterlitz et al (1981) and the myenteric plexus membranes according to Monferini et al (1981). A sample of 0·10 mL of homogenate, corresponding to 100 mg of cerebral tissue adjusted to a final volume of 1 mL with a solution containing the drug, was used. Specific binding corresponded to the difference between total binding and non-specific binding obtained by addition of a high concentration of levallorphan (10 μM). The final concentrations of the following ligands (provided by New England Nuclear Corp.) were: [^{3}H]dihydromorphine ([^{3}H]DHM: 73·6 Ci $mmol^{-1}$), 0·7 nM; [^{3}H]naloxone ([^{3}H]NAL: 42·7 Ci $mmol^{-1}$) 1 nM; [^{3}H]D-Ala2-D-Leu 5 enkephalin ([^{3}H]DADLE: 46·9 Ci $mmol^{-1}$) 0·5 nM; and [^{3}H]ethylketocyclazocine ([^{3}H]EKC: 18·7 Ci $mmol^{-1}$) 0·5 nM. The binding studies with myenteric plexus membrane suspensions were performed according to Roman et al (1989) with 240 μL of homogenate adjusted to a final volume of 300 μL with a solution containing the product and the radioactive ligand at their final concentrations: [^{3}H]DAGO (57·5 Ci $mmol^{-1}$ Amersham, UK) 2 nM; [^{3}H]DADLE: 1 nM and [^{3}H]EKC: 2 nM. Tubes were incubated for 45 min at 25°C and then filtered on Whatman GF/B glass fibre filters. After repeated washings with Tris HCl buffer, the filters were dried and the radioactivity counted.

A selectivity index was calculated as described by Chavkin et al (1982). This represents the ratio of the lowest IC50 for a given compound to the IC50 of the subtype considered. The sodium shift was calculated according to Pert & Snyder (1974). This was calculated from the capacity of a given drug to displace [^{3}H]naloxone in the presence and in the absence of 100 mM NaCl (ratio of the two IC50 values).

Pharmacokinetic determinations

In a first series of experiments plasma concentrations of fedotozine were measured over 24 h after oral doses of 2·5 and 5 mg kg^{-1}, respectively.

Concentrations of fedotozine and its active metabolite norfedotozine in the intestinal wall were measured in two mongrel dogs dosed with 2·5 and 5 mg kg^{-1} [^{14}C]fedotozine given orally in the fasted state. Animals were killed at 1 and 3 h after drug administration and tissue fragments from various parts of the digestive tract were separated into muscle and mucosal layers. Tissue samples were homogenized in 0·1 M, pH 10 carbonate buffer and then extracted with ethyl acetate. TLC and autoradiographic development were carried out for tissue and plasma extracts. Zones of radioactivity were eluted from the silica gel plates for liquid scintillation counting of radioactivity.

The levels of fedotozine and its metabolite norfedotozine (*N*-desmethylfedotozine) in the gastric juice were measured in one mongrel dog with a gastric fistula following i.v. administration of fedotozine tartrate at 5 mg kg^{-1}. After a wait of approximately 15 min to ensure adequate distribution of the drug through the whole body, simultaneous gastric juice and venous blood samples were taken at regular intervals over 5 h. The concentrations of both compounds were determined by gas chromatography. Samples were extracted with hexane–isopropanol (99:1 v/v) at pH 12 and extracts were analysed with a Hewlett Packard instrument (5890 A) equipped with a nitrogen phosphorus detector. A 15 m × 0·53 mm Supelcowax 10 column was used. The oven temperature was 250°C and the injector and detector temperatures were 280°C. Hydrogen, air, carrier gas (helium) and auxiliary gas (helium) flow rates were 2·9, 88·9, 15 and 15·6 mL min^{-1}, respectively.

Results

Effects of i.v. vs p.o. administration of fedotozine in dogs

Fasted dogs. The effects of fedotozine after intravenous administration are summarized in Table 1 and illustrated in Figs 1, 2. The drug was tested between 1 and 5 mg kg^{-1}. At 1 mg kg^{-1} i.v., fedotozine increased antral motility in only a

Table 1. Effect of fedotozine on antral motility.

Fedotozine (mg kg^{-1})	Control	Motility index (arbitrary units) 1 h	2 h	3 h
Intravenous				
2	146±43	249±49**	220±32*	188±40
5	255±31	28±3**	198±22*	250±15
Oral				
2·5	84±26	79±22	159±55*	221±68*
5	107±55	122±74	150±84*	183±95*

The values represent the mean ± s.e.m. of the antral motility index which is the sum of (n contractions × class of amplitude) per hour (see Methods for more detail). The statistical comparison was made using Student's *t*-test for paired values. n = 3 to 5 dogs.

third of the animals tested, the motility index being increased by 27% in those dogs; in the other dogs no stimulation was observed and the overall effect was not significant. This dose could be considered as a threshold dose of the stimulatory activity of the drug. As shown in Fig. 1 and Table 1, fedotozine at 2 mg kg^{-1} i.v. strongly stimulated antral motility with the appearance of high amplitude contractions. Periods of regular spiking activity of longer duration than the normal MMC phase III (15·9±3 vs 7·1±0·6 min for the first period) appeared in the duodenum and was propagated to the mid jejunum (Fig. 1). Two periods of stimulation with 10 to 30 min intervals were often recorded (Fig. 2). At the higher dose (5 mg kg^{-1}), fedotozine inhibited antral motility whilst still stimulating the small intestine, inducing ectopic and prolonged phase III followed by 5 to 30 min of quiescence and then a 3 h disorganization of the motor profile.

As observed following i.v. administration, fedotozine at 2·5 and 5 mg kg^{-1} orally stimulated digestive motility at all levels tested (Fig. 3). The antral motility index was increased by about 100% for both doses up to 3 h (Table 1). One to four phases of regular spiking activity were observed in addition to the normal MMC cycle (Fig. 3). With respect to i.v. administration, these periods of spiking activity lasted longer than the normal MMC phase III (24±4·1 vs 7·0±0·5 min). These periods of intense stimulation were followed by a period of disorganization of the motor profile lasting up to 3 h after 2·5 mg kg^{-1} (Fig. 3).

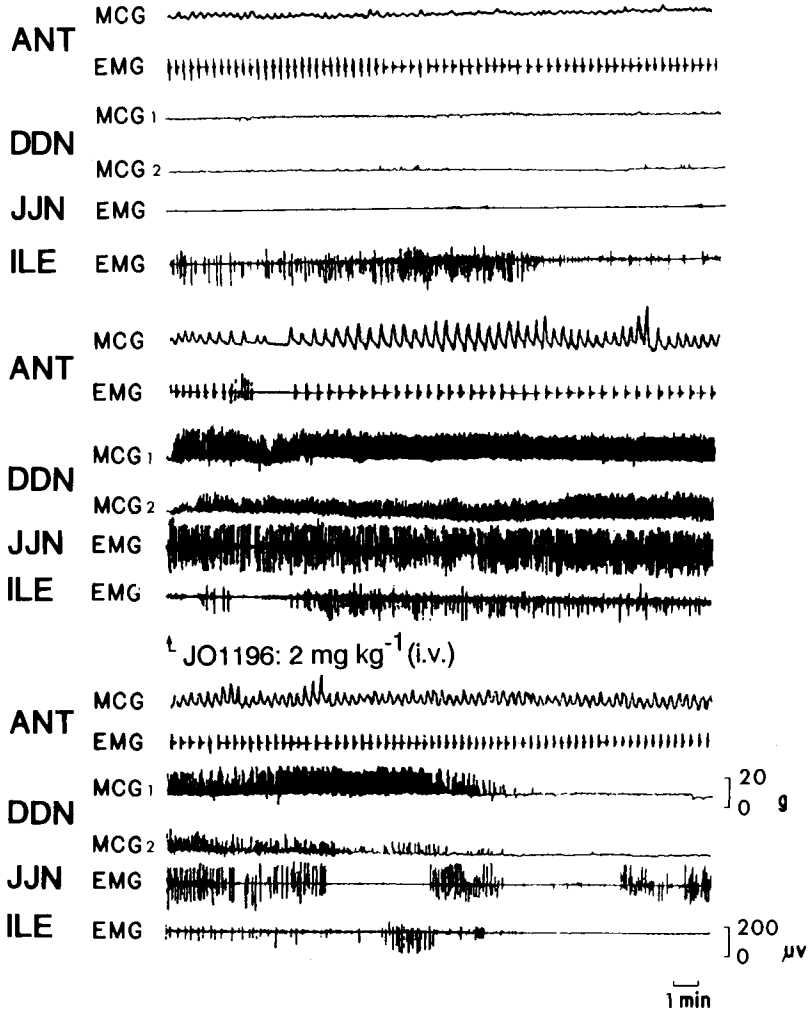

FIG. 1. Effect of fedotozine (2 mg kg^{-1} i.v.) on the electrical (EMG) and mechanical (MCG) activity of the antrum (ANT), duodenum (DDN), jejunum (JJN) and ileum (ILE) recorded in a fasted, unanaesthetized dog. Note the strong stimulatory effect observed at all levels and the very long phase III (32 min) induced by fedotozine at the duodenal and jejunal level.

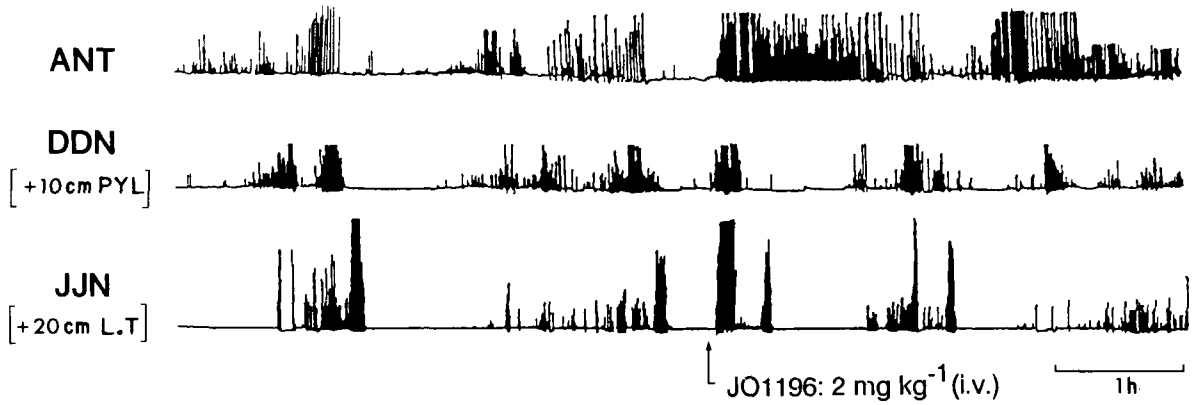

FIG. 2. Long term effect of 2 mg kg^{-1} i.v. of fedotozine on the pattern of myoelectrical activity (integrated record). Note the intense response at the antrum level and on the gut, the immediate appearance of a phase III type of activity on the duodenum in place of the silent pattern of the MMC cycle, migrating on the jejunum and followed by three other phase III type of activity before recovering the normal MMC pattern.

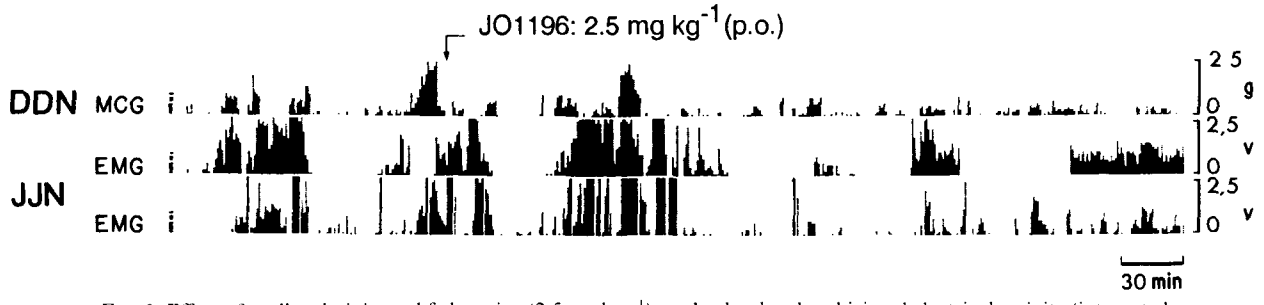

FIG. 3. Effect of orally administered fedotozine (2·5 mg kg^{-1}) on the duodenal and jejunal electrical activity (integrated record). The upper trace is the basal pattern and the lower trace shows the response to fedotozine. Note that increased activity appeared within 1 h after fedotozine and was followed by a long period of disorganization lasting up to 3 h post-treatment.

Fed dogs. Following i.v. administration, 2 h after feeding, fedotozine (2 mg kg^{-1}) induced a short-lasting inhibition followed by a progressive increase in the antral motility pattern. The motility index was increased from a basal value of 199 ± 57 to 210 ± 61, 276 ± 65 ($P < 0{\cdot}01$) and 308 ± 75 ($P < 0{\cdot}05$) at the first, second and third hours post-treatment, respectively. On the small bowel the drug induced a prolonged phase III type of activity ($8{\cdot}67 \pm 1{\cdot}32$ vs $5{\cdot}5 \pm 0{\cdot}24$ min in control animals) in the duodenum with migration to the jejunum (Fig. 4). The fasted type pattern was followed by a period of quiescence preceding the recovery of a normal fed pattern.

Mechanism of action

In-vivo studies. During a 30 min infusion of fedotozine (5 mg kg^{-1}), a constant increase of antral and duodenal motility occurred. Naloxone (0·30 mg kg^{-1}) injected 5 min before the start of the infusion reduced the fedotozine response at the antrum level and delayed and shortened phase III type activity due to fedotozine (Table 2). Atropine (0·10 mg kg^{-1}) also partly inhibited the effects of fedotozine in the antrum. No phase III-type period occurred in the duodeno-jejunum. Naloxone methylbromide had the same effect as naloxone. Atropine and naloxone administered together abolished the effect of fedotozine at all levels. Other antagonists were ineffective in blocking the effects of fedotozine.

Binding studies. Since naloxone was able to block the effect of fedotozine in-vivo, the capacity of fedotozine to displace various opioid ligands in-vitro was investigated. Table 3 shows that, at submicromolar concentrations, it inhibited the binding of tritiated dihydromorphine, DADLE and ethylketocyclazocine at μ-, δ- and κ- sites, respectively, in the brain and myenteric plexus preparations. The activity rates were 100:37:55 for μ-, δ and κ-receptors, respectively, in the brain and 87:100:46 in the myenteric plexus. Fedotozine was 3 to 16 times less active than morphine on μ-, δ- and κ-receptors but much more active than codeine. The IC50 ratio of fedotozine for μ brain and myenteric plexus receptors was 2·7 compared with 6·6 for morphine. In another set of experiments [^{3}H]naloxone was used to calculate the sodium shift produced by 100 mM NaCl. In such conditions, the IC50 of fedotozine increased from 400 ± 68 to 2600 ± 65 mM in the presence of a high concentration of NaCl, giving a sodium index of 6·5, whilst that for morphine and naloxone were 36 and 0·76, respectively.

In Table 3, the affinity of norfedotozine for opiate receptors is also reported. The metabolite has the same activity profile as fedotozine with a low specificity for μ-receptors. However, it has a lower affinity than fedotozine.

Pharmacokinetic measurements

Plasma and tissue concentrations of fedotozine. Plasma concentrations of fedotozine and norfedotozine were measured after oral doses of 2·5 and 5 mg kg^{-1}. At 2·5 mg kg^{-1}, fedotozine or norfedotozine were not detected in plasma samples at any time (< 20 ng mL^{-1}). At 5 mg kg^{-1}, three dogs had a plasma concentration lower than 20 ng mL^{-1} and one

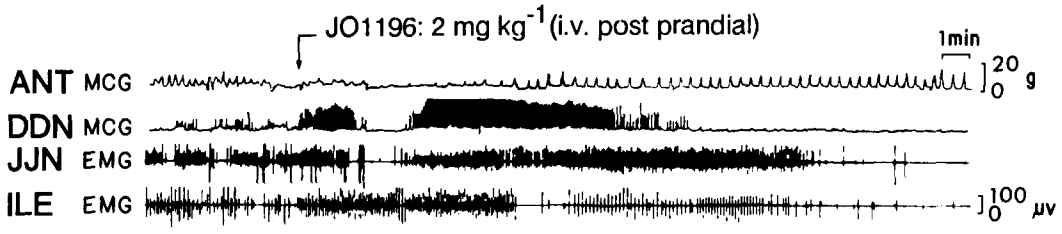

FIG. 4. Influence of fedotozine (2 mg kg^{-1}, i.v.) on the postprandial pattern of electrical and mechanical activity of the small intestine in fed dogs. Note the transient inhibition of the postprandial pattern and the occurrence of a long lasting stimulation of the duodenal and jejunal motility.

Table 2. Effects of naloxone, naloxone methyl bromide and atropine on the gastrointestinal motility induced by infusion of fedotozine in unanaesthetized dogs.

	n	Antrum C3, 4 contractions (h^{-1})	Jejunum phase III Activity profile	Delay (min)	Duration (min)
Fedotozine 5 mg kg^{-1} 30 min +saline 2 mL kg^{-1}	8	38·2±3·8	Phase III	8·2±1·4	13·4±2·7
Fedotozine+atropine 0·10 mg kg^{-1}	6	3·2±2·3 (***)	Absence of phase III (phase II)	—	—
Fedotozine +naloxone 0·30 mg kg^{-1}	6	13·0±4·1	Phase III (**)	25·0±3·5	7·7±0·7 (***)
Fedotozine +naloxone methyl bromide	4	18·0±6·3 (*)	Phase III	31·2±6·6	7·7±1·5
Fedotozine+naloxone 0·30 mg kg^{-1} +atropine 0·10 mg kg^{-1}	4	2·2±1·3 (***)	Absence of phase III (phase I)		

n: number of experiments, * $P<0.05$, ** $P<0.01$, *** $P<0.001$.
C3, 4: number of high amplitude (classes 3 and 4) contractions.
Statistical analysis (Student's t-test) compares the responses obtained with fedotozine+saline to fedotozine+antagonists.
Note that in the jejunum the antagonists either significantly delayed the appearance and decreased the duration of the phase III induced by fedotozine (naloxone and naloxone methylbromide) or totally inhibited the appearance of additional phase III. In the case of atropine, the phase III was replaced by a phase II pattern and the combination of both antagonists elicited inhibition of the motility pattern.

Table 3. Affinity of fedotozine, norfedotozine, morphine and codeine for different opiate receptor subtypes in guinea-pig brain and ileum myenteric plexus preparations.

	IC50 (nM)						
	Brain				Myenteric plexus		
	[^{3}H]DHM	[^{3}H]DAGO	[^{3}H]DADLE	[^{3}H]EKC	[^{3}H]DAGO	[^{3}H]DADLE	[^{3}H]EKC
Fedotozine	105±5·6	197±12	543±63	358±53	536±37	466±47	1017±70
Norfedotozine	164±37	499±73	1282±120	1376±235	626±114	1657±185	2776±325
Morphine	6·32±0·91	8·6±1·5	152±51	128±15	57·1±10·5	41·2±6·8	246±20
Codeine	419±91	960±90	4187±195	13007±2131	3559±845	15142±3562	36097±1979

The values are mean (±s.e.m) of 4 to 8 assays.

dog had a C_{max} (1 h) of 126 ng mL^{-1}. Tissue concentrations in the mucosal and muscle layers of two dogs after 5 mg kg^{-1} at 1 and 3 h are shown in Table 4. Concentrations representing even more than 60 times the highest plasma concentration (126 ng mL^{-1}) were reached (ileum). Concentrations in the mucosa were in general higher than in the muscle layers. Mean concentrations were 2·5 µg g^{-1} in the mucosa and 1·8 µg g^{-1} in muscle layers for fedotozine (1·8 and 0·6 µg g^{-1} for norfedotozine).

Presence of fedotozine in the gastric juice. Fedotozine is rapidly excreted into the gastric juice (Fig. 5). At 30 min after i.v. injection, 4·37% of the administered dose was present in the gastric juice as unchanged fedotozine (2·6 mg). Concentrations were 10 to 20 times higher in the gastric juice than in the plasma. At 5 h, about 7% of the dose had been recovered in the gastric juice as unchanged drug. Norfedotozine achieved lower concentrations in plasma (C_{max} = 56 ng mL^{-1}) and in the gastric juice (C_{max} = 300 ng mL^{-1}).

Table 4. Tissue concentrations of fedotozine in the upper digestive tract after oral administration (5 mg kg^{-1}). Results are expressed in ng g^{-1} fresh tissue.

	1 h				3 h			
	Mucosa		Muscle		Mucosa		Muscle	
Tissue	F	Nf	F	Nf	F	Nf	F	Nf
Stomach	2503	521	1565	491	1600	1612	910	589
Duodenum	1114	1560	719	570	696	1191	659	473
Jejunum	1636	1933	1235	718	651	1448	761	716
Ileum	6090	3748	1457	565	275	1436	576	504

F = Fedotozine Nf, Norfedotozine.

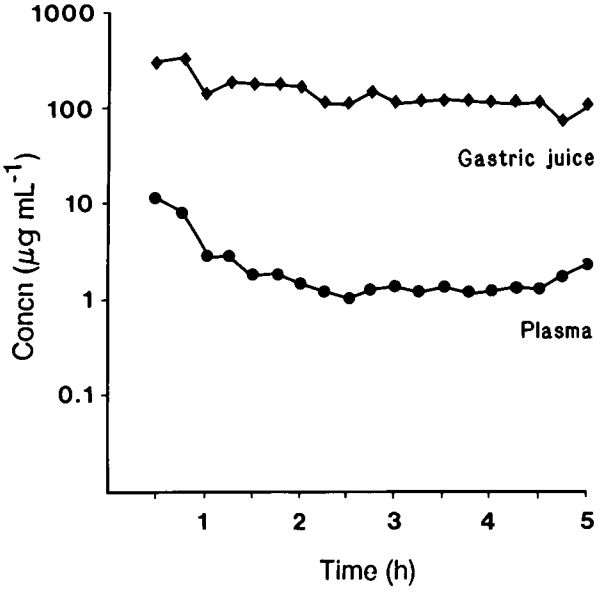

FIG. 5. Plasma and gastric levels of fedotozine following its intravenous administration (5 mg kg^{-1}).

Discussion

The present study shows that fedotozine induces a long-lasting stimulation of antral and small intestinal motility at doses ranging from 1 to 5 mg kg^{-1} by either the i.v. or oral route. These effects are inhibited by naloxone HCl, naloxone methylbromide and atropine, but not by hexamethonium, phentolamine, propranolol or methysergide. The results suggest that fedotozine acts on gut motility through opiate and cholinergic pathways. Nicotinic, α- or β-adrenergic and 5-HT receptors are not involved.

Opiate drugs are known to induce phase III-type activity in different species (Bueno & Ruckebusch 1978; Sarna & Lang 1985; Telford et al 1985). It is now well established that the activity front induced by opiates is a peripheral effect (Fioramonti et al 1984; Sarna & Lang 1985). This effect of opiates is blocked by naloxone as well as by naloxone methyl-bromide which does not cross the blood brain barrier (Fioramonti et al 1984) as reported here for fedotozine. Morphine and other opiates generally induce premature phase III at low doses and the appearance of the normal activity front (Sarna & Lang 1985). However, fedotozine given either i.v. (2 mg kg^{-1}) or orally (2·5 mg kg^{-1}) was able to induce a repetitive ectopic phase III superimposed on the normal MMC cycle. The activation by opiates of the small intestine motility in dog involves, at least in part, a stimulation of the cholinergic pathway, since it is inhibited by atropine (Fox & Daniel 1987a, b). As expected, the effects of fedotozine were also depressed by atropine. Since this drug or its main metabolite have no significant affinity for M1 and M2 receptors (data not shown) its effects as with other opiates probably involve an indirect interaction with cholinergic nerves.

A feature of further interest is the strong stimulation of antral motility produced by low doses of fedotozine, while high intravenous doses induced some inhibition only at this level. If this inhibitory activity corresponds to the classical response observed with opiates which is of central origin (Bueno & Fioramonti 1988) the stimulatory effect observed with low doses requires further explanation. Each of the subclasses of opiate receptors is present in the dog antrum (Allescher et al 1987) and because of fedotozine's lack of binding specificity it could act with an almost equal potency at all these sites. In unanaesthetized fasted dogs, intravenous opiates such as morphine induce strong antral contractions for about 15 min followed by a period of disorganization (Fioramonti et al 1984); i.v. and i.c.v. administered DALAMIDE, (D-Ala-2-Met-5-enkephalinamide) disrupted the MMC pattern for 4 h without a preceding transient stimulation (Bueno et al 1985) and orally administered loperamide produced a long-lasting period of rhythmic and sustained contractions (Fioramonti et al 1987). These different kinds of patterns could be explained in terms of central vs peripheral activity. However, the effects of morphine are both centrally and peripherally mediated whilst that of DALAMIDE (i.v.) and loperamide (p.o.) are peripheral since they are believed to permeate the blood brain barrier poorly (Wüster & Herz, 1978; Rapoport et al 1980). The antrum stimulatory effect of fedotozine resembled that of loperamide more than that of DALAMIDE or morphine. Therefore, it may be suggested that the antrum pattern elicited by low doses of fedotozine or loperamide could be related to a preferential affinity of these molecules for one of the μ-receptor subtypes (μ_1 or μ_2) described by Pasternak & Wood (1986). Receptors of the μ-subtypes have been shown to possess discriminative properties at different levels of the opiate range of activities, such as analgesia, hormone release, hypothermia or respiratory depression. In this report, the antral responses to fedotozine and loperamide could be related to a μ_2 activity but further experiments are needed to strengthen this hypothesis.

In-vitro binding studies also support the in-vivo data since fedotozine and norfedotozine displace opiate ligands from

their binding sites. Although the drug is about 16 to 23 times less active than morphine on brain μ-receptors, depending on the tritiated ligand used ([^{3}H]DHM or [^{3}H]DAGO), it is only 10 times less active in myenteric plexus preparations. This change in relative binding capacity between morphine and fedotozine in these preparations might be due to the heterogenicity of the sites on the two organs (Leslie et al 1980). With respect to codeine, fedotozine is 5 to 7 times more potent on μ-receptors and 35 times more effective on the κ-receptors in both preparations. In contrast to morphine and codeine, fedotozine does not show high selectivity for μ-receptors. The affinity for κ-receptors is only 1·8 and 1·9 times less in the brain and ileum, respectively, than for μ-receptors. The sodium shift of 6·2 observed with fedotozine corresponds to an agonist/antagonist (Pert & Snyder 1974), whereas morphine and naloxone can be classified as either a full agonist or antagonist, respectively. Norfedotozine exhibits the same activity profile and a weaker affinity than fedotozine.

The pharmacokinetic data reported in this study shows that the effects of fedotozine on motility occur at oral doses (2·5, 5 mg kg^{-1}) for which plasma concentrations (<20 ng mL^{-1}) cannot be detected. However, tissue levels after administration of fedotozine at 5 mg kg^{-1}, orally, indicate that the drug and its main metabolite are highly concentrated in the target organ. The estimated tissue/plasma ratio can reach about 15 in the muscle layers (antrum, jejunum) and more than 15 in the mucosa (antrum, ileum). When administered i.v., the drug is recovered in the gastric juice at much higher concentrations than in the plasma (10 to 20 times). The passage of drugs into the gastric juice has been observed for several compounds and may be explained by absorption of undissociated drug from the blood through the lipid membrane of the stomach. Therefore the concentration of drug in gastric juice would depend mainly on the ionization constant. These results indicate that after either oral treatment or i.v. injection, high concentrations of fedotozine and its active metabolite are found within the gastrointestinal walls and the lumen due to local and gastric absorption from the blood where concentrations are very low, in relation to an extensive first pass effect. In the case of fedotozine these properties allow the drug to be present in high concentrations in the target organ and may provide an explanation of the peripheral opiate activity of this compound.

References

Abel, M., Camilleri, M., Malagelada, J. R. (1985) High prevalence of gastric electrical dysrythmias in diabetic gastroparesis. Gastroenterology 88: 1299 (abstract)

Allescher, H. D., Ahmad, S., Kotska, P., Kwan, C. Y., Daniel, E. E. (1987) Distribution of opioid receptors in canine small intestine: implications for function. Am. J. Physiol. 256: G966–G974

Bueno, L., Ruckebusch, Y. (1978) Origine centrale de l'action excito-motrice de l'intestin par la morphine. Compte-rendus de la Société de Biologie 172: 972–977

Bueno, L., Fioramonti, J. (1988) Action of opiates on gastrointestinal function. In: Read, N. V. (ed.) Gastrointestinal Neurophysiology. Clinical Gastroenterology, Vol. 2, No. 1, Baillière Tindell Ltd pp 123–139

Bueno, L., Fioramonti, J., Hondé, C., Fargeas, M. J., Primi, M. P. (1985) Central and peripheral control of gastrointestinal and colonic motility by endogenous opiates in conscious dogs. Gastroenterology 88: 549–556

Chavkin, C., James, I. F., Goldstein, A. (1982) Dynorphin is a specific endogenous ligand of the opioid receptors. Science 215: 413–415

El Newihi, H., Dooley, C., Lohrmann, A., Zeidler, A., Valenzuela, J. E. (1985) Pathophysiologic studies in diabetic diarrhoea. Gastroenterology 88: 1374, 1517 (abstracts)

Fioramonti, J., Fargeas, M. J., Bueno, L. (1984) Comparative effects of morphine and cyclazocine on gastrointestinal motility in conscious dog. Arch. Int. Pharmacol. Ther. 270: 141–170

Fioramonti, J., Fargeas, M. J., Bueno, L. (1987) Stimulation of gastrointestinal motility by loperamide in dogs. Dig. Dis. Sci. 32: 641–646

Fox, J. E. T., Daniel, E. E. (1987a) Exogenous opiates: their local mechanisms of action in the canine small intestine and stomach. Am J. Physiol. 253: G179–G188

Fox, J. E. T., Daniel, E. E. (1987b) Activation of endogenous excitatory opiate pathways in the canine small intestine by field stimulation and motiline. Ibid. 253: G189–G194

Kosterlitz, H. W., Paterson, S J., Robson, L. E. (1981) Characterization of the κ-subtype of the opiate receptor in the guinea-pig brain. Br. J. Pharmacol. 29: 595–602

Kumar, D., Wingate, D. L. (1985) The irritable bowel syndrome: a paroxysmal motor disorder. Lancet ii: 973–977

Leslie, F. M., Chavkin, C., Cox, B. H. (1980) Opioid binding properties of brain and peripheral tissues: evidence for heterogenicity in opioid ligand binding site. J. Pharmacol. Exp. Ther. 214: 395–402

Miranda, M., Defilippi, C., Valenzuela, J. E. (1985) Abnormalities of the interdigestive motility complex and increases in duodenogastric reflux in gastric ulcer patients. Dig. Dis. Sci. 30: 16–21

Monferini, F., Strada, D., Maara, L. N. (1981) Evidence for opiate receptor binding in small intestine. Life Sciences 29: 595 602

Pascaud, X., Genton, M. J., Bass, P. (1978) A miniature transducer for recording intestinal motility in unrestrained chronic rats. Am. J. Physiol. 235: E532

Pasternak, G. W., Wood, P. J. (1986) Multiple mu opiate receptors. Life Sci 38: 1889–1898

Pert, C. B., Snyder, S. H. (1974) Opiate receptor binding of agonists and antagonists affected differentially by sodium. Mol. Pharmacol. 10: 868–879

Rapoport, S., Klee, W. A., Pettigrew, K. D. et al (1980) Entry of opioid peptides into the central nervous system. Science 207: 84–86

Roman, F., Pascaud, X., Vauché, D., Junien, J. L. (1989) Characterization of 3H.DAGO, 3H.DSLET and 3H.EKC binding in the guinea pig myenteric plexus. Advance in Biosciences, Vol. 75, Pergamon Press, pp 45–48

Ruckebusch, Y. (1973) L'électromyographie globale des muscles lisses à partir d'électrodes chroniques intrapariétales souples. Rev. Méd. Vét. 124: 1407–1434

Ruckebusch, Y., Bueno, L. (1976) Effect of feeding on the motility of the stomach and small intestine in the pig. Br. J. Nutr. 37: 397–405

Sarna, S. K., Lang, I. M. (1985) Dose- and time-dependent biphasic response to morphine on intestinal migrating myoelectric complex. J. Pharm. Exp. Ther. 234, No. 3:814–820

Summers, R. W., Anuras, S., Green, J. (1983) Jejunal manometry patterns in health, partial intestinal obstruction and pseudo obstruction. Gastroenterology 85: 1290–1300

Szurszewski, J. H. (1969) A migrating electric complex of the canine small intestine. Am. J. Physiol. 217: 1757–1763

Telford, G. L., Hoshmonai, M., Moses, A. J., Szurszewski, J. H. (1985) Morphine initiates migrating myoelectric complexes by acting on peripheral opioid receptors. Ibid. 249: G557–G562

Vantrappen, G., Janssens, J., Hellemans, J., Ghoos Y. (1977) The interdigestive motor complex of normal subjects and patients with bacterial overgrowth of the small intestine. J. Clin. Invest. 59: 1158–1166

Walker, G. D., Stewart, J. J., Bass, P. (1972) The effect of secretion on the fed pattern of gastric and duodenal contractile activity. Surg. Gynecol. Obstet. 134: 807–809

Wüster, M., Herz, A. (1978) Opiate agonist action of antidiarrhoeal agents in vitro and in vivo. Finding in support for selective action. Naunyn-Schmiedeberg's Arch. Pharmacol. 301: 187–194