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Functional characterization of the adenosine receptor mediating inhibition of peristalsis in the rat jejunum

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1 The non-selective adenosine agonist, 5'-N-ethylcarboxamidoadenosine (NECA), is a potent inhibitor of morphine withdrawal diarrhoea in rats. More recently we found that NECA exerts its antidiarrhoeal effect by inhibiting secretion in both the jejunum and ileum and also by inhibiting peristalsis in the ileum. The specific aim of this study was to characterize the receptor in the rat jejunum mediating inhibition of peristalsis via functional studies using a range of metabolically stable adenosine analogues based on the pharmacological criteria of relative agonist and antagonist potencies.

2 Peristalsis in the rat isolated jejunum was achieved by raising the pressure to between 7–11 cmH_2O for 3 min followed by a 3 min rest period (pressure at zero). The mean rate of peristalsis during inflation was 7.3 ± 0.1 peristaltic waves per 3 min and this rate remained consistent for up to 30 min, in 5 separate tissues. The inhibitory effects of the adenosine analogues were quantified by expressing their effects as a % reduction in the mean number of peristaltic contractions derived from the control tissues.

3 The rank order of agonist potency to reduce the rate of peristalsis was: N^6-cyclopentyladenosine (CPA) > NECA > **R**(–)-N^6-(2-phenylisopropyl)adenosine (**R**-PIA) > chloroadenosine (2-CADO) > **S**-PIA > 2-phenylaminoadenosine (CV-1808). This order complies well with the rank order of agonist potency that represents the activation of the A_1 receptor subtype (CPA > **R**-PIA = CHA = > NECA > 2-CADO > **S**-PIA > CV-1808).

4 The selective A_1 adenosine antagonist 1,3-dipropyl-8-cyclopentylxanthine (DPCPX) and the non-selective adenosine antagonist 8-phenyltheophylline (8-PT) at their respective concentrations of 10 nM and 2 μM caused parallel rightward shifts in the concentration-response curve to the non-selective A_1/A_2 agonist NECA. DPCPX was significantly more potent at inhibiting NECA than 8-PT as revealed by their apparent pA_2 values; DPCPX (9.5) and 8-PT (7.26). The high affinity of DPCPX relative to that of 8-PT suggests the presence of an A_1 and not an A_{2B} receptor. In addition, the high affinity of DPCPX (pA_2:9.37) against the selective A_1 agonist CPA, further confirms the presence of the A_1 receptor subtype.

5 In this study we found that the A_1 adenosine receptor is involved in regulating *in vitro* peristalsis which is different from the adenosine receptor regulating inhibition of secretion (A_{2B}) in the same region of intestine of the same species. We propose that A_{2B} adenosine agonists could be of clinical value in the management of diarrhoea that is due to microbiological organisms where antimotility effects are not desired.

Keywords: Adenosine receptors; rat intestine; peristalsis; antidiarrhoeal; A_1 receptor; NECA; *in vitro* technique

Introduction

It is now well established that adenosine has pharmacological actions on a variety of smooth muscle preparations and these effects are mediated via receptors which have been classified as P_1-purinoceptors (Burnstock, 1990; Kennedy, 1990). These receptors have been further subdivided into A_1, A_2 and A_3 receptors based on the orders of both agonist potency and antagonist affinity, G-protein coupling mechanisms, cellular responses and receptor cloning studies (Fredholm *et al.*, 1994). The A_1 receptor is differentiated from the A_2 receptor by the selective A_1 agonist, N^6-cyclopentyladenosine (CPA) which has a 2,500 fold A_1 selectivity vs A_2 in radioligand binding studies (Lohse *et al.*, 1988) and the selective A_1 antagonist 1,3-dipropyl-8-cyclopentylxanthine (DPCPX) which has a dissociation constant in the nanomolar range at A_1 and in the micromolar range at A_2 receptors (Bruns *et al.*, 1987a). The A_{2A} receptor is differentiated from the A_{2B} receptor by the selective A_{2A} agonist, CGS-21680 (Jarvis *et al.*, 1989) and the selective A_{2A} antagonist, PD-115199 (Bruns *et al.*, 1987b). At present there are no ligands available that are specific for the A_{2B} receptor. Consequently, the A_{2B} receptor is identified by the low affinity of A_{2A} selective ligands as highlighted in the agonist potency order and antagonist affinity order used to characterize adenosine receptor subtypes (Collis & Hourani, 1993). The recently described A_3 receptor can be identified by the selective agonist, N^6-benzyl-NECA which shows a 14 fold selectivity for A_3 receptors vs A_1 and A_{2A} (Van Galen *et al.*, 1994). This receptor is also insensitive to block by methylxanthines unlike the other adenosine receptor subtypes (Fredholm *et al.*, 1994) but is antagonized by 8-phenyl-substituted xanthines, such as BWA522, which is a potent antagonist in the nanomolar range (Fozard & Hannon, 1994).

It has been known for some time that adenosine and synthetic analogues have effects on the gut muscle of a variety of mammals, birds and amphibians and in particular the guinea-pig ileum (Drury & Szent-Gyorgi, 1929; Barsoum & Gaddum, 1935; Hayashi *et al.*, 1978; Collier & Tucker, 1983). The recent availability of adenosine analogues of known selectivities has facilitated the functional characterization of adenosine receptors. Some progress has been made in characterizing the adenosine receptors in the rat intestine. For example, it has been shown that the distal colon contains A_1 excitatory receptors in the muscularis mucosa (Bailey *et al.*, 1992) and A_2 inhibitory receptors in the longitudinal muscle (Bailey & Hourani, 1992) while the longitudinal muscle of the duodenum contains A_1 and A_{2B} receptor subtypes, both of which subserve relaxation (Nicholls *et al.*, 1992a). However, other regions of the small intestine, such as the jejunum and ileum have been

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relatively neglected except for our recent study which showed that the non-selective adenosine agonist, NECA, inhibits secretion and peristalsis in the rat ileum (Coupar & Hancock, 1994) and also inhibits secretion in the rat jejunum; this particular effect has been shown to be mediated via the A_{2B} receptor (Hancock & Coupar, 1995).

The specific aim of this study was to characterize the receptor mediating inhibition of peristalsis by use of a range of metabolically stable adenosine analogues based on the pharmacological criteria of relative agonist and antagonist potencies.

Methods

Male Hooded Wistar rats (250–350 g body weight) were stunned by a blow to the head and killed by exsanguination. A segment of proximal jejunum was excised close to the Ligament of Trietz and flushed of luminal contents with Krebs-Henseleit solution (composition mM: NaCl 118, KCl 4.7, $NaHCO_3$ 25, KH_2PO_4 1.2, $CaCl_2$ 2.5, $MgSO_4$ 1.2, D-(+)-glucose 11; bubbled with 95% O_2 and 5% CO_2). The segments were cut to approximately 7–10 cm lengths and were mounted vertically in a 30 ml organ bath containing Krebs-Henseleit solution maintained at 37°C and gassed with 95% O_2 and 5% CO_2. The open aboral end of each segment was secured over a glass tube connected to a reservoir containing Krebs-Henseleit solution. The reservoir contained a float which was linked to an isotonic transducer to measure luminal volume displacement as an index of circular muscle contraction. The oral end of each segment was closed off and connected to an isotonic transducer for recording longitudinal contractions under a load of 1 g. An intraluminal water pressure of 7–11 cm was required to elicit peristaltic activity. The tissues were allowed to equilibrate for 45 min during which time the Krebs-Henseleit solution was replaced every 15 min. Preliminary experiments showed that inflating the segments for 3 min followed by a 3 min rest period (water pressure at zero) produced regular peristaltic activity during the periods of inflation compared to a sustained elevated intraluminal pressure which produced variable and short lasting bursts of peristaltic activity. Consequently, the segments were inflated for 3 min followed by a 3 min rest period in all further experiments. The mean rate of peristalsis during this period was 7.3 ± 0.1 peristaltic waves per 3 min up to 30 min in 5 separate tissues. In these control tissues the rate of peristalsis in the first period of peristalsis was not significantly different from the final burst of peristalsis (Student's paired t test, $P < 0.05$).

Experimental design and analysis of results

In separate tissues, the inhibitory effects of the adenosine analogues were quantified by expressing their effects as a % reduction in the mean number of peristaltic contractions derived from the experiments described above. The presence of peristalsis was confirmed in each of the 3 min periods by allowing two peristaltic contractions to occur before adding the test drug. The number of contractions was then recorded in the presence of the drug in the remainder of the 3 min period. This number was then expressed as a % of the possible 5 contractions (mean number of peristaltic contractions being 7 in 3 min. Refer to Figure 1 for further details). This method of analysis was chosen because the number of peristaltic waves in consecutive inflations was too variable to measure a before and after agonist effect in one tissue. The tissues were washed after each concentration of agonist. A higher concentration of the same agonist was tested when 2 peristaltic contractions were achieved in the subsequent 3 min period of elevated pressure. Each tissue was only functionally viable for up to 30 min which limited the number of concentrations of each agonist to four. The potency of each agonist (EC_{50} value) was calculated by linear regression analysis with 95% confidence intervals of the estimates.

The non-selective adenosine agonist, NECA and the A_1-selective agonist, CPA, were employed in the experiments designed to measure the affinity of the antagonists. In these experiments the antagonists were added 30 min prior to adding the first concentration of NECA or CPA to the bath and their affinities were estimated by measuring their apparent pA_2 values. The resultant dose ratios (DR) with associated CI were used to calculate the apparent pA_2 values from the K_B values derived from the equation-:

$$K_B = B/DR\text{-}1$$

where B is the concentration of the antagonist and the apparent pA_2 value is equal to -log K_B. Student's unpaired t test was used to compare single treatment means with their respective controls. The criterion for statistical significance was set at $P < 0.05$. Each rat is represented by $n = 1$.

Drugs

Agonists: chloroadenosine (2-CADO), N^6-cyclopentyladenosine (CPA),2-phenylaminoadenosine (CV-1808),5'-N-ethylcarboxamidoadenosine (NECA), **R**-(-)-N^6-(2-phenylisopropyl) adenosine (**R**(-)-PIA) and (**S**(+)-PIA). All were obtained from RBI, Natick, U.S.A. The agonists were dissolved in 0.9% w/v saline except CV-1808 and CPA which were dissolved in 6% and 1% ethanol in saline respectively and then diluted with saline to give the required concentration. The vehicle had no effect on the responses of the tissues.

Antagonists: atropine sulphate (Sigma, Castle Hill, Australia), 1,3-dipropyl-8-cyclopentylxanthine (DPCPX, RBI, Natick, USA) and 8-phenyltheophylline (8-PT, RBI, Natick, U.S.A.) were dissolved in 1% v/v dimethylsulphoxide (DMSO), 0.75% v/v 1 M NaOH in saline and further diluted with saline to give the required concentration. This vehicle had no significant effect on the peristaltic activity of the jejunum ($P > 0.05$, Student's unpaired t test).

Results

Atropine (100 nM, 10 min incubation) abolished peristaltic activity in the jejunum ($n = 4$). Each of the adenosine agonists caused a concentration-related reduction in the number of peristaltic waves occurring within 3 min with the following order of potency: CPA > NECA > **R**-PIA > 2-CADO > **S**-PIA > CV-1808 (Table 1, Figure 2). The inhibition elicited by each adenosine agonist was very rapid in onset, reaching its maximum within 30s and was readily reversible on washing (Figure 1).

The adenosine antagonists DPCPX and 8-PT at their respective concentrations of 10 nM and 2 μM caused parallel rightward shifts in the concentration-response curve to NECA. The respective dose-ratios were 30.46 ($n = 17$) and 37.46 ($n = 12$), corresponding to apparent pA_2 values of 9.47 (95% CI 9.41–9.54) for DPCPX and 7.26 (95% CI 7.20–7.33) for 8-PT using NECA as the agonist (Figure 3a and b). In addition,

Table 1 Potencies of adenosine agonists at reducing the number of peristaltic waves within 3 min

Adenosine agonist	EC_{50} (nM)	(n)	*95% CI*
CPA	2.3	(12)	2.9
NECA	25.4	(7)	3.8
R-PIA	41.2	(6)	3.7
2-CADO	150.9	(10)	2.8
S-PIA	239	(7)	1
CV-1808	7700	(7)	1500

For abbreviations, see text.

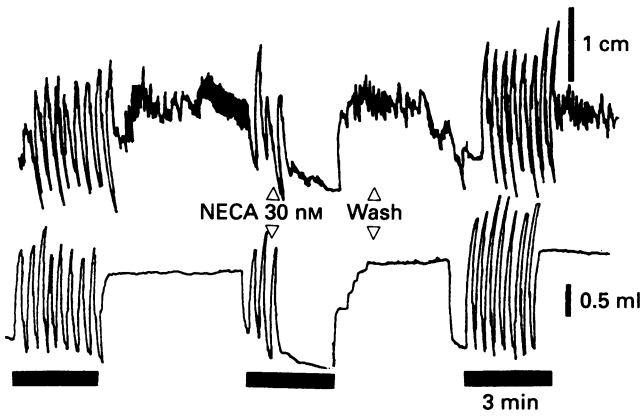

Figure 1 The inhibitory effect of 5'-N-ethylcarboxamidoadenosine (NECA) on peristalsis in a segment (10 cm in length) of rat jejunum. The top trace represents the contraction of the longitudinal muscle and the bottom trace shows the associated volume expulsion as a result of circular muscle contraction in response to increasing the intraluminal pressure to 10 cmH_2O. The 3 min periods of elevated intraluminal pressure are represented by the thick black bars. In the second period of peristaltic activity, NECA (30 nM) was administered after the second peristaltic contraction which produced an 80% reduction in the rate of peristalsis (i.e. one contraction out of a possible 5 contractions). The inhibition was very rapid in onset and was readily reversible on washing.

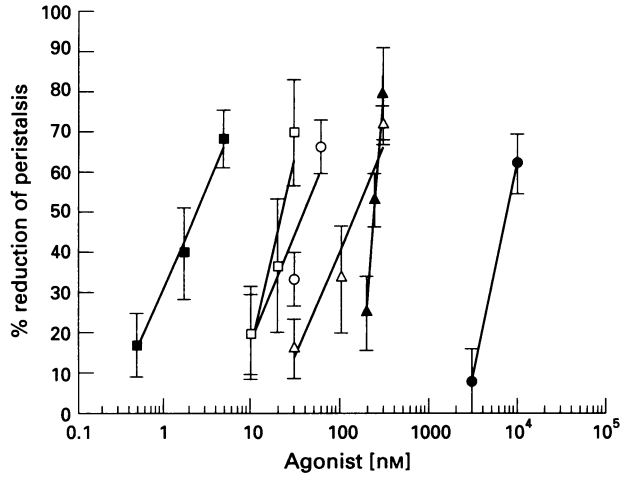

Figure 2 The concentration-effect relationships for a range of adenosine agonists at inhibiting peristalsis in the rat jejunum. The decreasing order of potency is as follows:- CPA (■), NECA (□), R-PIA (○), 2-CADO (△), S-PIA (▲), CV-1808 (●). Values are means ± s.e. means. For abbreviations, see text.

DPCPX (10 nM) also produced a parallel rightward shift in the concentration-response relationship to CPA. The dose-ratio for DPCPX was 24.24 ($n=19$) using CPA as the agonist giving an apparent pA_2 value of 9.37 (95% CI 9.01–9.73) (Figure 3c). Neither antagonist altered the number of peristaltic contractions within 3 min that occurred during the first burst of peristalsis (control: 8.6 ± 1.2, $n=5$; DPCPX: 9.67 ± 0.94, $n=9$; 8-PT: 8.83 ± 0.98, $n=6$, Student's unpaired t test, $P>0.05$).

Discussion

The rank order of adenosine agonist potencies and antagonist affinities obtained in this study suggest that the A_1 adenosine receptor is involved in mediating inhibition of peristaltic activity in the rat jejunum based on the classification table described by Collis & Hourani (1993). These results, in conjunction with our previous findings on the rat jejunum

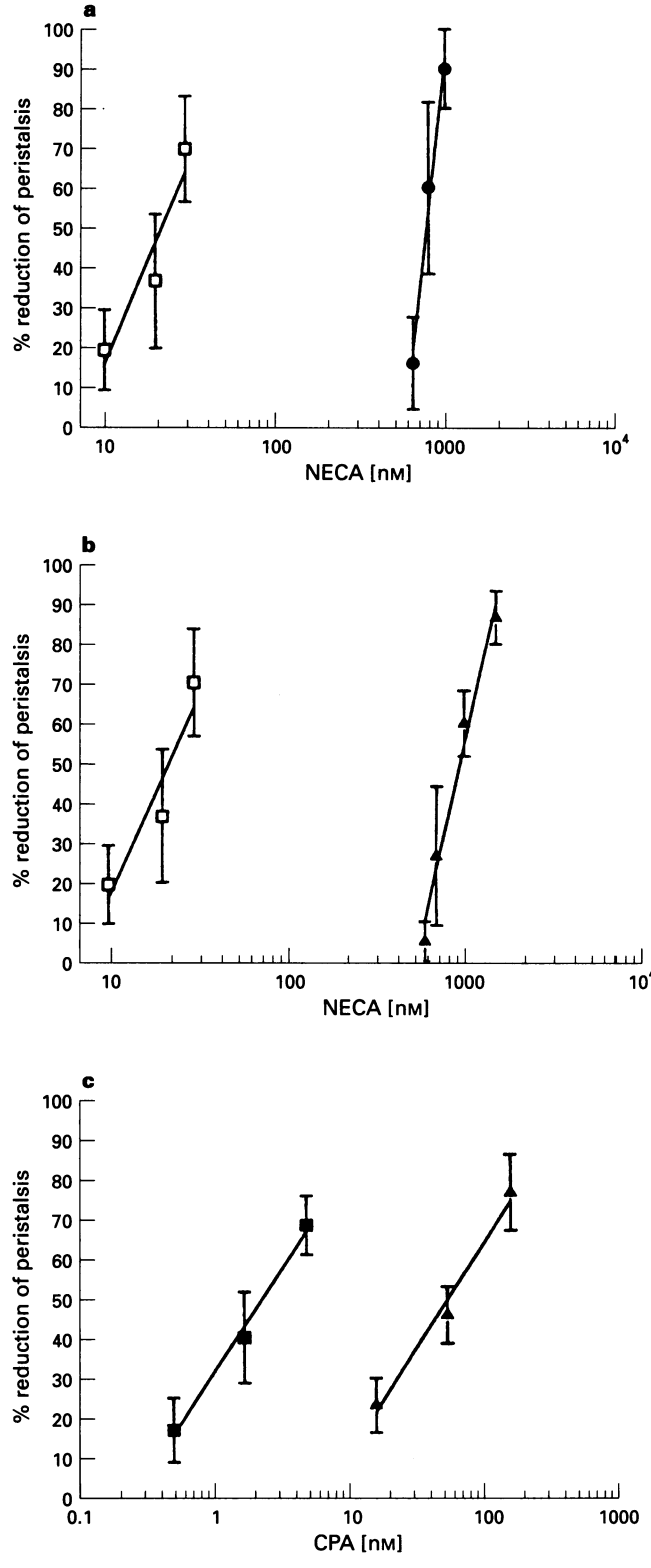

Figure 3 (a) Concentration-effect relationship of the non-selective adenosine agonist, NECA at inhibiting peristalsis in the rat jejunum in the absence (□) and in the presence (●) of the antagonist, DPCPX (10 nM). (b) NECA in the absence (□) and in the presence (▲) of the antagonist, 8-PT (2 μM). (c) The A_1 selective agonist, CPA at inhibiting peristalsis in the rat jejunum in the absence (■) and in the presence (▲) of the A_1 selective antagonist, DPCPX (10 nM). Values are means ± s.e. means. For abbreviations, see text.

(Hancock & Coupar, 1995), show that adenosine agonists exert their antidiarrhoeal action in rats (Dionyssopoulos *et al.*, 1992) by inhibiting both intestinal secretion via the A_{2B} ade-

nosine receptor (Hancock & Coupar, 1995) and intestinal motility or peristalsis via the A_1 adenosine receptor.

The agonists used in this study inhibited intestinal peristaltic activity in the order of CPA > NECA > **R**-PIA > 2-CADO > S-PIA > CV-1808 which almost coincides with the rank order of agonist potency that represents activation of the A_1 receptor: CPA > **R**-PIA = CHA = > NECA > 2-CADO > S-PIA > CV-1808 (Collis & Hourani, 1993). The only discrepancy in the order of agonist potencies is that NECA is more potent than **R**-PIA in this study, whereas **R**-PIA is equipotent or more potent than NECA in the A_1 receptor classification. However, this difference in potency between the two agonists is relatively small. The order of agonist potency in this study is also similar to the A_{2B} receptor classification where the order of agonist potency is: NECA > 2-CADO > **R**-PIA = CHA > S-PIA > = CV-1808 > = CGS-21680 (Collis & Hourani, 1993). The only contrast is that **R**-PIA is more potent than 2-CADO in this study which is the reverse of that found in the A_{2B} classification. Unfortunately the A_{2B} receptor classification does not include our most potent agonist CPA but there is no evidence in the literature to indicate that CPA is without activity at the A_{2B} receptor. However, CPA is structurally and pharmacologically similar to CHA, so it is reasonable to assume that it will also be less potent than NECA at A_{2B} receptors.

The order of agonist potencies in this study to inhibit peristalsis is almost the opposite of the order that represents activation of the A_{2A} adenosine receptor where CGS-21680 = NECA > CV-1808 > = 2-CADO > **R**-PIA = CHA = CPA > S-PIA (Collis & Hourani, 1993). For instance, CV-1808 is the least potent agonist to inhibit peristalsis but is a potent agonist in the A_{2A} order and CPA the most potent agonist in this study is almost without activity in the A_{2A} receptor classification. The single exception is that NECA is a potent agonist in both this study and the A_{2A} receptor classification. CGS-21680, the selective A_{2A} agonist was not tested in this study since the agonists tested indicated the activation of an A_{2B} or A_1 receptor where CGS-21680 is the least potent in both classifications. Similarly the A_3 receptor was excluded because NECA was significantly more potent than **R**-PIA at inhibiting peristalsis in the rat jejunum unlike the equipotent activity of these agonists found in the A_3 classification (Collis & Hourani, 1993). In addition, xanthine antagonists, such as DPCPX and 8-PT as used in this study have low affinity at the A_3 receptor.

The rank order of agonist potencies together with antagonist affinity orders are used to characterize functional adenosine receptors as outlined by Collis & Hourani (1993). The antagonists, DPCPX and 8-PT were investigated specifically to differentiate between A_1 and A_2 receptors since the order of agonist potencies suggests the presence of either an A_1 or an A_{2B} adenosine receptor that is responsible for mediating inhibition of peristaltic activity in the rat jejunum. The differences between the affinities of the two antagonists is evident in isolated tissues where 8-PT is non-selective for A_1 and A_2 receptors, whilst DPCPX has a 30–50 fold greater affinity for A_1 but is equi-effective with 8-PT at A_2 receptors (Collis *et al.*, 1989). In this study, DPCPX is significantly more potent at inhibiting NECA-evoked responses than 8-PT, as revealed by their apparent pA_2 values; DPCPX (9.47) and 8-PT (7.26). In addition, the apparent pA_2 value for DPCPX against the selective A_1 agonist, CPA was 9.53 which further confirms the presence of an A_1 receptor. These pA_2 values for both antagonists in the rat jejunum are in line with the pA_2 values reported in the rat and guinea-pig atria (Collis *et al.*, 1988) which are thought to possess the A_1 receptor (Collis, 1983; Haleen *et al.*, 1987). The apparent pA_2 for 8-PT against NECA in the rat jejunum (95% CI 7.20–7.33) is similar to the pA_2 value for 8-PT reported in the rat (95% CI 6.54–7.95) and guinea-pig atria (95% CI 5.89–7.23). In addition, the apparent pA_2 values for DPCPX in the rat jejunum against NECA (95% CI 9.41–9.54) and CPA (95% CI 9.01–9.73) are the same as the value for DPCPX in the rat atria (95% CI 7.33–9.42). However, the affinity values for DPCPX in the rat jejunum are significantly higher than that found in the guinea-pig atria (95% CI 7.67–8.65). One possible reason for the low affinity of DPCPX in the guinea-pig atria compared to that of the rat jejunum could be due to a species difference. For instance, Ukena *et al.* (1986) reported that the affinity of xanthine derivatives at A_1 binding sites in the brain are significantly higher in the rat than the guinea-pig. The high affinity values of DPCPX against NECA and CPA compared to the relatively low affinity of 8-PT against NECA found in this study discounts the possibility that adenosine agonists are inhibiting peristalsis in the rat jejunum via the A_2 adenosine receptor. This high antagonist potency of DPCPX along with the order of agonist potency indicates the presence of an A_1 adenosine receptor in the rat jejunum.

In the absence of exogenous adenosine agonists, neither DPCPX nor 8-PT altered the rate of peristalsis at the concentrations employed. Similarly, in a previous study these antagonists were free of intrinsic pro-secretory activity in the rat jejunum (Hancock & Coupar, 1995). It appears that under the conditions of both experiments, endogenous adenosine is not released in the rat jejunum to inhibit either peristalsis or fluid secretion. This situation is unlike that found in the guinea-pig atria, where both antagonists in the absence of exogenous adenosine increased the rate of beating of the isolated atria which was due to antagonism of the negative chronotropic action of endogenous adenosine (Collis *et al.*, 1989). The antagonist concentrations employed in the guinea-pig atria were considerably higher than those used in the rat jejunum which could explain the detection of endogenous adenosine activity in the atria.

In this study the intestine was radially distended with fluid which is thought to be detected by mechanoreceptors in the muscularis externa (Yokoyama & North, 1983; Smith *et al.*, 1990) to initiate peristalsis. The present study has not precisely determined the tissue location of the adenosine receptor involved in controlling peristalsis. However, our previous studies do suggest that the receptor is not located on intestinal smooth muscle or cholinergic neurones since high concentrations of NECA failed to affect transmurally stimulated cholinergic contractions of the rat jejunum (Coupar & Hancock, 1994). From these results, together with our more recent finding that atropine inhibits peristalsis in the jejunum it could be postulated that adenosine analogues act proximal to the final cholinergic neurone in the peristaltic reflex arc.

There is evidence from other functional studies that adenosine receptors do exist in the rat intestine. For instance, CPA and NECA have been shown to relax the duodenum by activating A_1 and A_{2B} receptors respectively (Nicholls *et al.*, 1992a). Additionally, A_1 agonists caused contraction of the muscularis mucosa (Bailey *et al.*, 1992) and A_2 agonists cause relaxation of the longitudinal muscle of the distal colon (Bailey & Hourani, 1992). There is also evidence from molecular biology that adenosine receptors (A_1, A_2, A_{2B}) are expressed in numerous rat tissues such as the brain, spinal cord, heart, lung, urinary bladder and in particular the large intestine (A_{2B}) but not in the small intestine (Stehle *et al.*, 1992). A recent binding study has established the existence of A_1 binding sites using the A_1-selective ligand, DPCPX in the duodenum and colon (Peachey *et al.*, 1994). In another binding study the non-selective ligand, NECA, was shown to bind to both A_1 and A_2 receptors in the rat brain, but did not exhibit any specific binding in the small intestine (Bruns *et al.*, 1986). This emphasizes the importance of functional studies to reveal and characterize receptor subtypes. From the above studies, it is apparent that the duodenum is the only section of the small intestine previously shown to contain adenosine receptors.

The guinea-pig has been the only species used to date to investigate the effect of adenosine or its analogues on intestinal peristalsis (Okwuasaba & Hamilton, 1975; Van Nueten *et al.*, 1976). This seems surprising since it is well accepted that the effects and mechanisms of drug action, particularly of opiates, is species-dependent. This situation is highlighted by our finding that the adenosine agonist, NECA inhibits transmu-

rally stimulated contractions of the guinea-pig ileum but not of the rat ileum or jejunum (Coupar & Hancock, 1994). In this recent study we also tested the effect of NECA on the peristaltic reflex in the rat ileum to find that NECA at 10 nM slowed the reflex while 30 nM virtually abolished it (Coupar & Hancock, 1994). The potency of NECA in the ileum is in line with the potency of NECA (EC_{50} = 25.4 nM) found in the rat jejunum in the present study, despite the finding that the rate of peristalsis is faster in the jejunum (1 contraction per 25.7 s) than that previously found in the rat ileum (1 contraction per min). In the guinea-pig, peristalsis was also found to be faster in the upper than the lower portion of the small intestine (Kromer *et al.*, 1981). It has been suggested that regional differences in the amounts of inhibitory neuromodulators released, such as endogenous opioids, are responsible for regulating the decrease in rate of peristaltic activity observed aborally in the guinea-pig small intestine (Kromer *et al.*, 1981). The endogenous inhibitory neuromodulator could also be adenosine formed from ATP. It has been known for some time that ATP is taken up and can be released in response to stimulation of non-adrenergic inhibitory fibres that innervate the gut (Burnstock *et al.*, 1970; Satchell & Burnstock, 1971; Su *et al.*, 1971). In the rat duodenum it has been shown that ATP is rapidly metabolized to ADP, AMP and inosine, a metabolite of adenosine which is inactive at purinoceptors (Nicholls *et al.*, 1992b). Adenosine itself is rapidly inactivated in this tissue by adenosine deaminase (Franco *et al.*, 1988). In addition, studies by Reiter *et al.* (1989) detected adenosine deaminase in the human intestinal mucosa. However, in this study we were unable to reveal the presence of endogenous adenosine as indicated by a lack of enhancement of the peristaltic reflex at the antagonist concentrations employed.

It must be emphasized that the results of this study are only an approximation of the inhibitory effect of adenosine analogues on intestinal motility because of the *in vitro* technique employed. *In vivo* studies in this area need to be performed to confirm that inhibition of peristalsis seen *in vitro* leads to decreased transit in the whole animal. However, in this study we have found that the adenosine receptor involved in regulating *in vitro* peristalsis in the rat jejunum (A_1) is different from that regulating secretion (A_{2B}) (Hancock & Coupar, 1995) in the same region of intestine of the same species. Our recent characterization of adenosine receptors in the rat jejunum provides the opportunity to inhibit motility selectively without affecting secretion and *vice versa* by the use of selective adenosine agonists in the management of diarrhoea. On this basis we postulate that A_{2B} adenosine agonists could be of clinical value in the management of diarrhoea that is due to microbiological organisms where antimotility effects are not desired.

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