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Sympathoinhibition by adenosine A_1 receptors, but not P2 receptors, in the hamster mesenteric arterial bed

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Abstract

The aim of the present study was to determine whether there are prejunctional inhibitory P2 purine receptors on sympathetic nerves in the hamster isolated perfused mesenteric arterial bed. Adenosine 5′-*O*-(3-thiotriphosphate (ATPγS; 10 μM), adenosine 5′-*O*-(2-thiodiphosphate) (ADPβS; 100 μM) and AMP (10 μM) had no significant effect on neurogenic contractions to electrical field stimulation. In contrast, P1 receptor agonists attenuated sympathetic vasoconstriction with a potency order of N^6-cyclopentyladenosine (CPA) > 5′-(*N*-ethylcarboxamido)-adenosine (NECA) > adenosine. The pEC_{50} value for CPA was 7.5 ± 0.1 ($n = 7$). The concentration-inhibitory effect curve to CPA was shifted to the right by the adenosine A_1 receptor antagonist, 8-cyclopentyl-1,3-dipropyl-xanthine (DPCPX; 10 nM; apparent pK_B 9.6; $n = 6–7$). In methoxamine raised-tone mesenteries CPA (0.001–10 μM) did not elicit vasorelaxation, and NECA and adenosine were only weak vasorelaxants. These results indicate that adenosine A_1 receptors, but not P2 receptors, inhibit prejunctionally sympathetic neurotransmission in the hamster mesenteric arterial bed.

Keywords: Adenosine; ATP; Mesentery, hamster; Prejunctional neuromodulation; Purine receptor; Sympathetic neurotransmission

1. Introduction

Purines and pyrimidines are important modulators of cardiovascular function via actions at purine receptors characterized by their distinct molecular structures, pharmacological profiles and signal transduction mechanisms (see Abbracchio and Burnstock, 1994; Fredholm et al., 1994; Ralevic and Burnstock, 1998). There are two main families of purine receptors. P1 receptors are activated primarily by adenosine and are subdivided into A_1, A_{2A}, A_{2B} and A_3 receptors. P2 receptors are activated by ATP, ADP, UTP and UDP, and are divided into two families of ionotropic P2X receptors, and G protein-coupled P2Y receptors.

P2 receptors are expressed on the vascular smooth muscle and endothelium and can act to regulate blood vessel tone. In recent years, it has been reported that P2 receptors can also be expressed on the peripheral terminals of sympathetic nerves. Inhibition of sympathetic neurotransmission via prejunctional P2 receptors has been described in a number of tissues from rat and mouse, including the vas deferens, atria, tail artery, kidney, iris and pancreas (see Von Kügelgen et al., 1996; Koch et al., 1998). Although the subtype identity of these receptors is currently unclear, attenuation of the prejunctional sympathoinhibitory actions of P2 receptor ligands by pertussis toxin indicates that they are likely to be G protein-coupled P2Y receptors (Von Kügelgen et al., 1993, 1995). The existence of prejunctional P2Y receptors is, however, still controversial. Sympathetic perivascular nerves have long been known to express inhibitory adenosine P1 receptors, which are typically adenosine A_1 receptors (Brown and Collis, 1983; Illes et al., 1988, 1989).

The main aim of the present study was to determine whether there are P2 purine receptors that inhibit prejunctionally sympathetic neurotransmission in resistance vessels of the hamster mesenteric arterial bed. In addition to providing specific information about the possible role of P2 receptors in mesenteric blood vessels, this would tell us more about the regional distribution of P2 receptors in the cardiovascular system. In order to confirm that our pharmacological approach was an appropriate and sensitive assay for evaluation of prejunctional inhibitory neuromodulation, we additionally identified and characterized the subtype of adenosine P1 receptor modulating sympathetic

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neurotransmission in the hamster mesenteric arterial bed. In the context of the present study an advantage in the use of the mesenteric arterial bed of the hamster, compared to that of the rat, is that in the former there is only a minimal role of vasorelaxant $P2Y_1$-like receptors (Ralevic and Burnstock, 1996a,b). This allows the use of P2Y receptor ligands such as adenosine 5′-*O*-(2-thiodiphosphate (ADPβS) and 2-methylthio ATP in the characterization of prejunctional P2 receptors, whilst largely avoiding postjunctional relaxant effects.

2. Materials and methods

2.1. Isolated mesenteric arterial bed preparation

Adult male Syrian hamsters (100–140 g) were killed, after exposure to CO_2, by decapitation. Mesenteric beds were isolated and set up for perfusion as described previously (Hill et al., 1996; Ralevic et al., 1997). In brief, the abdomen was opened and the superior mesenteric artery exposed and cannulated with a hypodermic needle. The superior mesenteric vein was cut and blood flushed out from the preparation with approximately 0.5 ml of Krebs' solution. The gut was carefully dissected away from the mesenteric vasculature and the preparation mounted on a stainless steel grid (7×5 cm) in a humid chamber (custom-made at University College London). The preparation was perfused at a constant flow rate of 3 ml min^{-1} using a peristaltic pump (model 7554-30, Cole-Parmer Instrument, Chicago, IL). The perfusate was Krebs' solution of the following composition (mM): NaCl 133, KCl 4.7, NaH_2PO_4 1.35, $NaHCO_3$ 16.3, $MgSO_4$ 0.61, $CaCl_2$ 2.52 and glucose 7.8, gassed with 95% O_2–5% CO_2 and maintained at 37°C. Responses were measured as changes in perfusion pressure (mm Hg) with a pressure transducer (model P23XL, Viggo-Spectramed, Oxnard, CA) on a side arm of the perfusion cannula, and recorded on a polygraph (model 7D, Grass Instrument, Quincy, MA). Preparations were allowed to equilibrate for 30 min prior to experimentation. Agonists and antagonists were applied by addition to the perfusate reservoir. Electrical field stimulation was applied using a Grass 7D stimulator at the parameters indicated.

2.2. Experimental protocol

The effect of purines as modulators of the sympathetic neurogenic contractile response of the hamster mesenteric arterial bed was tested initially against responses to electrical field stimulation applied repetitively at a constant frequency of 64 Hz (90 V, 1 ms) for 5 s every 2 min. This frequency and duration of stimulation were chosen as they produce robust and reproducible contractile responses (10–30 mm Hg), which can be reversibly inhibited by certain purine compounds. Agonists were applied by perfusion for 10 min at each concentration, with a 10–15 min period of washout between each concentration. Ten minutes was more than sufficient for a maximal inhibitory effect of each concentration to be achieved. The effect of only a single agonist was investigated in each preparation, unless the agonist had no significant effect on the contractile response, in which case another agent was tested, after washout. The effect of 8-cyclopentyl-1,3-dipropyl-xanthine (DPCPX; 10 nM) on inhibitory-effect curves to N^6-cyclopentyladenosine (CPA) were investigated in separate preparations in which DPCPX was added to the perfusate 10 min after the onset of electrical field stimulation, and 15 min before co-perfusion with purine receptor agonist.

In a second experimental protocol, the effect of CPA on frequency response curves to electrical field stimulation were investigated. Three consecutive frequency–response curves to electrical field stimulation were generated at 12–64 Hz (90 V, 1 ms, applied for 5 s every 5 min), with 15 min separation. In time control experiments, the best reproducibility between contractile responses to electrical field stimulation was between the second and third frequency–response curves. Therefore, the second frequency–response curve was used as a control for the effect of CPA, tested against responses of the third frequency–response curve.

A third experimental protocol investigated the effect of adenosine (100 μM) on frequency–response curves to electrical field stimulation (4–32 Hz, 90 V, 1 ms) generated using a longer time of stimulation, namely 30 s, with stimulations applied at 5 min intervals. Thirty minutes separated each of three consecutive frequency–response curves. In time control experiments, the best reproducibility was found to be between the second and third frequency–response curves. Thus, the effect of adenosine was investigated against the third response curve and compared to the second, preceding, response curve.

Confirmation of selective stimulation of perivascular nerves and of the identity of the neurotransmitter mediating the contractile response to electrical field stimulation was by addition of either guanethidine (5 μM) or prazosin (1 μM) at the end of the time control experiments, or after washout of agonists.

The direct effects of the purine compounds were assessed in a fourth experimental protocol in which the tone of the mesenteric arterial beds was increased by perfusion with methoxamine (10–80 μM). When a stable level of tone was achieved mesenteries were perfused with increasing concentrations (0.1 nM–100 μM) of purines for up to 10 min at each concentration until a stable effect on tone was achieved.

2.3. Drugs used

Methoxamine (hydrochloride), norepinephrine (arterenol bitartrate), adenosine (hemisulfate), ADPβS (trilithium salt), AMP (sodium salt), adenosine 5′-*O*-(3-thiotriphos-

phate (ATPγS; tetralithium salt), 5′-(*N*-ethylcarboxamido)-adenosine (NECA), DPCPX and prazosin (hydrochloride) were from Sigma. Guanethidine (Ismelin) was from Ciba Laboratories, Surrey. All drugs were dissolved in distilled water except for NECA and DPCPX, which were made up as stock solutions of 10 mM in dimethylsulphoxide and diluted in distilled water.

2.4. Data analysis

Contractile responses were measured as increases in perfusion pressure (mm Hg) above baseline. In the experimental protocol involving repeated contractions to electrical field stimulation at 64 Hz, three contractile responses were measured at each of the periods: (a) immediately before drug application; (b) at the time of maximal response during drug application, and (c) after washout. This was done at each concentration. For each of these periods the mean of the three consecutive contractions to electrical field stimulation was calculated. Although there was a decrease in contractile responses to electrical field stimulation with time, within each test period for each drug concentration responses were reasonably reproducible. Furthermore, inhibition by purines was considered significant only if contraction to electrical field stimulation after washout of purines was greater than in the presence of purines, indicating recovery of the response. Inhibition was calculated as the percentage decrease in the contractile response to electrical field stimulation compared to the control response at each concentration. The direct effects of purines on the tone of preparations perfused with methoxamine were calculated as the percentage decrease in perfusion pressure. Results are presented as means ± S.E.M., with the number of observations in parentheses (n). The EC_{50} value was taken to be the concentration of agonist that produced a 50% (half-maximal) response. pEC_{20} is the negative logarithm of the concentration of agonist required to elicit inhibition by 20% of the contractile response to electrical field stimulation or of methoxamine-raised tone. Apparent pK_B (negative log of K_B) was evaluated from $K_B = [B]/(DR - 1)$, where B = concentration of antagonist and DR (dose ratio) is the difference between the EC_{50} values in the absence and presence of antagonist. Comparisons were made by Student's t-test, with $P < 0.05$ considered significant.

3. Results

Electrical field stimulation elicited contractile responses of the hamster mesentery that were blocked by guanethidine (5 μM) or prazosin (1 μM) at all of the frequencies and durations of stimulation used in the present study. This indicates that noradrenaline is the principal mediator of the sympathetic contractile response to electrical field stimulation under these conditions, as reported previously (Hill et al., 1996; Ralevic and Burnstock, 1996a; Ralevic et al., 1997).

3.1. Effect of purine compounds on contractile responses to electrical field stimulation

Repetitive pulses of electrical field stimulation at 64 Hz (90 V, 1 ms, applied for 5 s every 2 min), elicited contractile responses of the mesentery: 20 ± 2.3 mm Hg at the onset and 15 ± 1.8 mm Hg at the end of experimentation ($n = 36$). Although responses were larger at the beginning than at the end of experimentation, they were reasonably reproducible within each test period for each concentration of purine, and recovery after washout was used as further evidence of an inhibitory effect. At basal tone the purine compounds had no direct effect on the tone of the preparations, with the exception of ATPγS, which elicited a small and short-lasting constriction at the highest concentration tested (10 μM) (9.4 ± 4 mm Hg, $n = 4$).

ATPγS (0.1–10 μM), ADPβS (100 μM) and AMP (10–100 μM) had no significant effect, or had effects only at the highest concentration, on the contractile response to electrical field stimulation (Fig. 1). In contrast, the adenosine P1 receptor agonists CPA, NECA and adenosine significantly attenuated the contractile response to electrical field stimulation in a concentration-dependent manner (Fig. 1). The selective A_1 receptor agonist CPA elicited maximal inhibition of 37.4 ± 4.3% and the pEC_{50} value was 7.5 ± 0.1 ($n = 7$). Inhibition by adenosine (0.1–100 μM) and the selective A_2 receptor agonist NECA (0.1–10 μM) did not reach a maximum, but the pEC_{20} values were 5.8 ± 0.1 ($n = 6$) and 7.6 ± 0.2 ($n = 5$), respectively. The pEC_{20} value for CPA was 8.2 ± 0.1 ($n = 7$), and confirmed the relative potencies of the agonists as CPA > NECA > adenosine.

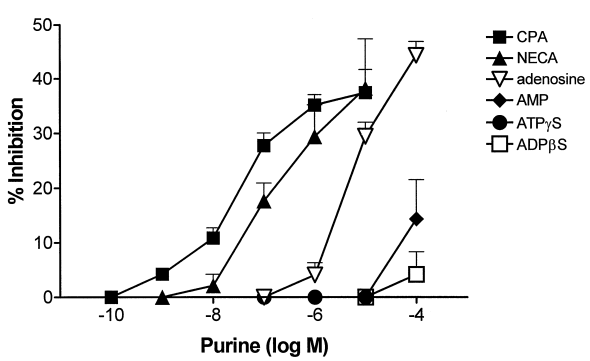

Fig. 1. Concentration inhibitory effect curves for N^6-cyclopentyl adenosine (■, CPA, $n = 7–10$), adenosine (▽, $n = 6$), 5′N-ethylcarboxamide adenosine (▲, NECA, $n = 5$), AMP (◆, $n = 4$), ATPγS (●, $n = 4$), and ADPβS (□, $n = 4$) on the contractile response to electrical field stimulation (64 Hz, 90 V, 1 ms, for 5 s) of the hamster isolated perfused mesenteric arterial bed. The contractile response elicited by electrical field stimulation prior to the addition of purine compound at each concentration was taken as 100% and the effect of the purine calculated as the percentage inhibition of this response. Means ± S.E.M. are shown.

3.2. Effect of DPCPX on CPA-mediated inhibition of responses to electrical field stimulation

The concentration-inhibitory effect curve for CPA on contractions to electrical field stimulation (64 Hz) was shifted to the right by an adenosine receptor antagonist, DPCPX, used at a concentration (10 nM) selective for the A_1 adenosine receptor (Fig. 2). The apparent pK_B value was 9.6 ($n = 6$–7). DPCPX alone had no effect on contractile responses mediated by electrical field stimulation of the mesenteric arterial beds.

3.3. Effect of CPA on frequency–response curves to electrical field stimulation

CPA (100 nM) inhibited contractile responses to electrical field stimulation, producing a rightward shift in the frequency response curve (12–64 Hz, 90 V, 1 ms, 5 s) (Fig. 3). The stimulation frequency required to elicit an increase in perfusion pressure of 10 mm Hg was significantly greater in the presence (44.3 ± 3.7 Hz; $n = 7$) than in the absence (32.7 ± 1.8 Hz; $n = 6$) of CPA ($P < 0.01$). There was no significant difference between the stimulation frequencies required to elicit an increase in perfusion pressure of 10 mm Hg between consecutive control response curves generated in the absence of drugs ($n = 6$).

3.4. Effect of purine compounds on methoxamine-sustained vascular tone

The tone of a group of hamster mesenteric arterial beds was increased with methoxamine by a level comparable to that produced by electrical field stimulation (64 Hz) (by 29.6 ± 2.5 mm Hg above baseline; $n = 14$). CPA did not elicit vasorelaxation at concentrations (0.001–10 μM) at which it attenuated contractile responses to electrical field stimulation ($n = 4$) (Fig. 4). Adenosine and NECA both caused relaxation; relaxation to adenosine at 10 μM was $1.7 \pm 1.1\%$ ($n = 5$). NECA was more potent, causing relaxation at 10 μM of $14.9 \pm 3.6\%$ ($n = 5$) (Fig. 4). The pEC_{20} value for adenosine was between 3 and 4 and for NECA was < 5 (Fig. 4).

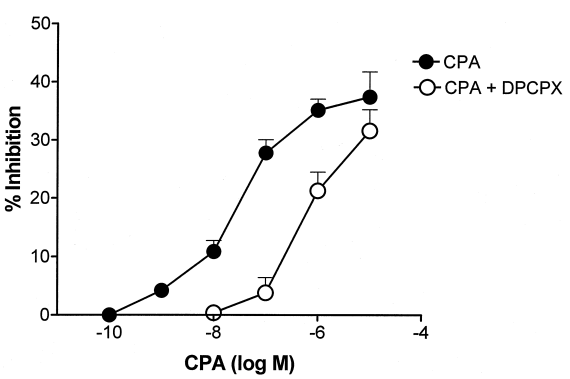

Fig. 2. Concentration inhibitory effect curves for N^6-cyclopentyl adenosine (CPA) in the absence and presence of the selective A_1 receptor antagonist DPCPX (10 nM) on the contractile response to electrical field stimulation (64 Hz, 90 V, 1 ms, for 5 s) of the hamster isolated perfused mesenteric arterial bed. CPA alone (●, $n = 7$), CPA in the presence of DPCPX (○, $n = 6$). Percentage inhibition of responses was calculated as in Fig. 1. Means ± S.E.M. are shown.

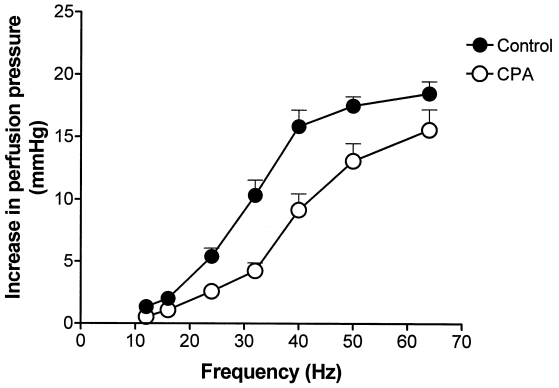

Fig. 3. Frequency-dependent vasoconstriction elicited by electrical field stimulation (12–64 Hz, 90 V, 1 ms, for 5 s) in the absence and in the presence of N^6-cyclopentyl adenosine (CPA; 100 nM). Data are from the second and third frequency response curves in a series of three per preparation. Frequency–response curves in the absence (●; second response curve) and in the presence (○; third response curve) of CPA ($n = 6$). Means ± S.E.M. are shown.

3.5. Effect of adenosine on frequency–response curves to electrical field stimulation

Electrical field stimulation (4–32 Hz, 90 V, 1 ms) applied for longer stimulation periods, of 30 s, evoked contractile responses which, at 32 Hz, were in excess of 100 mm Hg. There was no significant difference in the frequency required to elicit a contractile response of 60 mm Hg in the absence or presence of adenosine (100 μM) ($n = 6$), or between consecutive control frequency–response curves generated under similar conditions, but in the absence of drugs ($n = 6$).

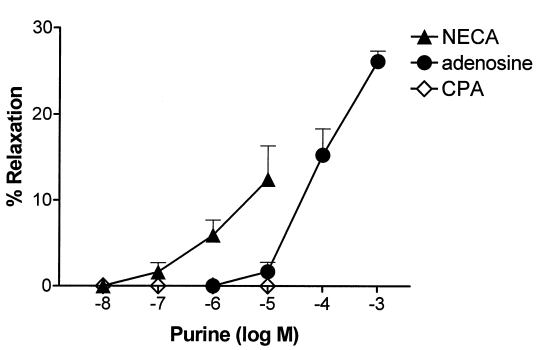

Fig. 4. Effect of NECA (▲, $n = 5$), adenosine (●, $n = 6$) and N^6-cyclopentyl adenosine (CPA) (◇, $n = 4$) on hamster mesenteries with tone raised with methoxamine. CPA had no direct effects on tone at up to 10 μM. Responses are evaluated as the percentage decrease in perfusion pressure. Means ± S.E.M. are shown.

4. Discussion

These results indicate that prejunctional adenosine P1 receptors of the A_1 subtype modulate sympathetic neurotransmission in hamster mesenteric arteries. By contrast, the lack of effect of ATPγS and ADPβS as modulators of contractile responses to electrical field stimulation indicates no role for P2 purine receptors in prejunctional inhibition of sympathetic neurotransmission in hamster mesenteric arteries.

ATPγS was ineffective in the hamster mesenteric arterial bed at a concentration (10 μM) at which it has a pronounced inhibitory effect on [^{3}H]noradrenaline overflow in rat atria (EC_{50} value of 4.8 μM) (Von Kügelgen et al., 1995). ADPβS (10–100 μM) was also virtually inactive as an inhibitor of contractions to electrical field stimulation in the hamster mesenteric arterial bed, although 2-methylthio ATP (1–100 μM), which can act at the same P2Y receptors as ADPβS (Schachter et al., 1996), has been shown to reduce noradrenaline overflow in the rat tail artery (Gonçalves and Queiroz, 1996). The lack of effect of ATPγS and ADPβS as modulators of sympathetic vasocontraction suggests that P2 receptors are not expressed by the peripheral terminals of sympathetic nerves in hamster mesenteric arteries. This is in contrast to reports in a number of other tissues of rat and mouse, including the vas deferens, atria, tail artery, kidney and iris, in which prejunctional inhibitory P2 receptors on sympathetic nerves have been described (Von Kügelgen et al., 1996). The existence of prejunctional P2Y receptors is, however, still controversial. The purinergic component of sympathetic neurotransmission varies greatly among different blood vessels (Burnstock and Ralevic, 1996) and does not contribute significantly to sympathetic contractile responses of the perfused hamster mesenteric arterial bed (Ralevic and Burnstock, 1996a, and present study). Thus, it is possible that inhibitory P2 receptors are expressed selectively on sympathetic nerves utilizing ATP as a cotransmitter. This is consistent with the concept that prejunctional P2 receptors on sympathetic nerve terminals are release-modulating autoreceptors (Von Kügelgen et al., 1993, 1995).

The majority of studies identifying and characterizing inhibitory P2 receptors on sympathetic nerves in the periphery have measured the effects of purine compounds on evoked [^{3}H]noradrenaline release in preloaded tissues (see Von Kügelgen et al., 1996; Koch et al., 1998). As we were able to detect adenosine A_1 receptor-mediated prejunctional inhibition of sympathetic neurotransmission in the hamster mesentery, this indicates that the lack of effect of ATPγS and ADPβS reported in the present study is not due to a lesser sensitivity of our assay technique. The difference between this and other studies is likely, therefore, to be due to tissue or species heterogeneity in expression of prejunctional P2 receptors. Pronounced species differences in the functional expression of P2 receptors (endothelial $P2Y_1$ receptors) has been observed previously for mesenteric arteries of the rat and hamster, as only in the former do endothelial $P2Y_1$-like receptors contribute significantly to vasorelaxation (Ralevic and Burnstock, 1988, 1996a,b).

Both the pEC_{50} value for CPA and the apparent pK_B value for DPCPX as an antagonist of responses to CPA are similar to those reported for prejunctional adenosine A_1 receptors in other tissues (Kurz et al., 1993; Von Kügelgen et al., 1995), consistent with an action of CPA at adenosine A_1 receptors. The potency order of the adenosine analogs tested as inhibitors of sympathetic neurotransmission (CPA > NECA > adenosine) is further consistent with a pharmacological profile of adenosine P1 receptors of the A_1 subtype. These findings are in line with previous reports of adenosine-mediated inhibition of sympathetic neurotransmission. In the rat mesenteric arterial circulation adenosine exerts pre- and postjunctional inhibitory control of vasoconstrictor sympathetic neurotransmission via actions at P1 receptors (Kamikawa et al., 1980; Lukacsko and Blumberg, 1982; Kuan and Jackson, 1988). The subtype of prejunctional P1 receptor has been characterized in other vessels and species; adenosine A_1 receptors modulate sympathetic neurotransmission in rabbit mesenteric arteries (Illes et al., 1988), rabbit portal vein (Brown and Collis, 1983) and rat tail artery (Illes et al., 1989), and sensory–motor neurotransmission in the rat mesenteric arterial bed (Rubino et al., 1993).

The lack of a direct effect on vascular tone by CPA, the most potent of the adenosine analogues mediating inhibition of neurotransmission, shows unequivocally that the inhibition is pre- and not postjunctional. Specifically, it rules out the possibility of an inhibitory action of CPA on sympathetic neurotransmission mediated by vasorelaxation or by an interaction with α_1 adrenoceptors. The prejunctional inhibitory effect of CPA was observed across the frequency response curve, although a clear inverse relationship between frequency of nerve stimulation and the effectiveness of prejunctional inhibition, which is characteristic of neuromodulators, was not seen. As adenosine and NECA were more potent as inhibitors of sympathetic neurogenic contraction than as mediators of vasorelaxation, this indicates that their actions are also mediated, at least in part, prejunctionally. Adenosine has a greater affinity for A_1 receptors vs. other adenosine receptors, which may account for its greater potency as a prejunctional inhibitor of sympathetic neurotransmission. The postjunctional action of adenosine is likely to be mediated by an adenosine A_2 receptor, as NECA, but not CPA, caused vasorelaxation. In the rat mesentery the principal postjunctional adenosine receptor is an adenosine A_{2B} receptor located primarily on the smooth muscle (Rubino et al., 1995; Prentice et al., 1997), but its identity in hamster mesenteric arteries is not known. At higher concentrations of adenosine it is likely that both pre- and postjunctional actions contribute to inhibition of sympathetic neurotransmission.

With a longer time of electrical field stimulation, for 30 s, there was no significant modulation by adenosine of the frequency–response curve, at a concentration at which it inhibits markedly both responses to electrical field stimulation applied for 5 s, and methoxamine-induced tone. This cannot be attributed to the different amplitudes of the responses evoked with long and short times of stimulation, because low frequency stimulation for 30 s elicited contractions of similar amplitude as those at 5 s (64 Hz). This indicates that a longer duration of neurogenic stimulation can override the pre- and postjunctional inhibitory effects of adenosine, which has implications for the role of adenosine in modulation of sympathetic neurotransmission.

Although excitatory P2 receptors are expressed in sympathetic ganglia, the sympathetic nerve terminals possess no, or only a small number of, excitatory P2 receptors (Von Kügelgen et al., 1996). The possibility that excitatory prejunctional P2 receptors are expressed on sympathetic nerve terminals in hamster mesenteric arteries was not investigated in the present study. Our study also does not investigate the possibility of other, non-P2Y, prejunctional inhibitory purine receptors.

There are several possible endogenous sources of adenosine that might act at prejunctional adenosine A_1 receptors on sympathetic nerve terminals. ATP and adenosine (following metabolic breakdown of ATP) are released with noradrenaline from sympathetic nerves in a large number of tissues, in variable proportions depending on the tissue and neuronal activity (see Burnstock and Ralevic, 1996). In rat atria, augmentation of [^{3}H]noradrenaline release in the presence of P2 receptor antagonists was dependent on the frequency of stimulation and this, together with other data, was taken as an indication of presynaptic P2 autoreceptors activated by ATP coreleased from sympathetic nerves (Von Kügelgen et al., 1995, 1996). In the hamster mesenteric arterial bed, however, sympathetic contractile responses to electrical field stimulation are blocked by the α_1-adrenoceptor antagonist prazosin, suggesting that ATP is not a major component of sympathetic neurotransmission in this tissue. Although ATP is a major component of sympathetic neurotransmission in rabbit mesenteric arteries (Ramme et al., 1987), inhibition by endogenous adenosine of sympathetic excitatory junction potentials was only weakly related to activity, and it was suggested that the adenosine originates principally from the vascular smooth muscle cells rather than from the nerve terminals (Illes et al., 1988). DPCPX alone had no effect on sympathetic neurotransmission in the hamster mesenteric arterial beds, which indicates that there is no endogenous release of adenosine under the conditions of the present study: adenosine A_1 receptors on the sympathetic nerves may, however, assume a significant role in vivo or in pathophysiological conditions associated with release of adenosine to high concentrations extracellularly.

In conclusion, the results of this study show that adenosine A_1 receptors, but not P2 receptors, mediate prejunctional inhibition of sympathetic neurotransmission in hamster mesenteric arteries. This is in contrast to the reported expression of both P1 and P2 receptors on sympathetic nerve terminals in the vas deferens and atria of the rat and mouse, and tail artery, kidney and iris of the rat.

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