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PURINERGIC NEUROTRANSMISSION AND NEUROMODULATION

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INTRODUCTION

The term "purinergic" was introduced by Burnstock (1) to denote nerves that utilize ATP or some related purine nucleotide as the transmitter. It has since been used in a broader sense, encompassing other purine compounds and neuromodulation as well as neurotransmission. Earlier observations on peripheral purinergic neurotransmission and related mechanisms have been comprehensively documented (2–4).

This review deals primarily with recent findings on peripheral purinergic neurotransmission and purinergic modulation of adrenergic transmission. For reviews of the role of adenosine and ATP in central synaptic transmission, see (5–7).

PURINERGIC NEUROTRANSMISSION

ATP as a Transmitter

Nonadrenergic, noncholinergic motor neurons have long been recognized to occur in various mammalian organs (8, 9) including blood vessels (10, 11). Those supplying the gastrointestinal tract are often referred to as intrinsic enteric nerves, distinct from the extrinsic sympathetic and parasympathetic, of the autonomic nervous system.

The notion that one type may use ATP or a related purine as the transmitter was, according to Burnstock (12), triggered by an electrophysiological observation in 1962. Following blockade of adrenergic and cholinergic

nerve transmission in the isolated guinea pig taenia coli, a large transitory hyperpolarization, which led to cessation of spontaneous spike discharge and relaxation, resulted upon neuronal stimulation. This finding was followed by identification of ATP as the likely transmitter substance. The lines of evidence have been recounted by the proponent (12) briefly as follows.

1. Storage: ATP and the enzyme systems that synthesize ATP occur ubiquitously in cells. Adenosine is taken up and stored as ATP in nerves. Electronmicroscopically identifiable "large opaque vesicles" and histochemically stained ATP-rich microsomal fractions and nerves appear to be associated with pharmacologically purinergic nerves. 2. Release: Efflux of ATP and its breakdown products occurs during stimulation of nonadrenergic, noncholinergic fibers in a variety of tissues. 3. Mimicry: The mechanical and electrophysiological responses to exogenously applied ATP closely mimic the responses to nonadrenergic, noncholinergic nerve stimulation in visceral and vascular tissues (see also 13). 4. Inactivation: An adenosine uptake system, as well as Mg^{2+}-activated ATPase, 5'-nucleotidase, and adenosine deaminase, have been demonstrated in various organs. 5. Drugs: The responses to adenosine nucleotides and nonadrenergic, noncholinergic nerve stimulation are both inhibited by quinidine, phentolamine and other imidazolines, 2,2'-pyridylisatogen (PIT), and/or apamin. Both responses also become tachyphylactic to exogenous ATP and they are potentiated by dipyridamole in some cases. 6. Receptors: A basis for two types of purinergic receptors specific for adenosine and adenine nucleotides has been provided.

The attempts to characterize the nonadrenergic, atropine-resistant excitatory innervation of the urinary bladder illustrate the purinergic approach. A purine nucleotide was postulated to be the transmitter in the rat and guinea pig bladder (14, 15), since ATP as well as nerve stimulation produced a rapid and brief contraction that was blocked by quinidine and depressed by tachyphylaxis to ATP. Quinacrine fluorescence histochemistry, shown to be ATP-selective, revealed nerve fibers and ganglion cells in the bladder, and a Ca^{2+}-dependent release of ATP followed nerve stimulation (16). The insensitivity of rat bladder to ATP was attributable to rapid breakdown of this nucleotide (17). In the rabbit detrusor, contractions induced by ATP and nerve stimulation were selectively depressed by theophylline, by PIT, and by desensitization with ATP (18). Prostaglandins may mediate the late sustained phase of contraction by exogenous ATP, but not the initial rapid contraction, which is more likely caused by ATP as a transmitter (19). A photoaffinity analogue of ATP, arylazido aminopropionyl ATP ($ANAPP_3$), was recently used in the guinea pig bladder. This agent antagonized the contraction by exogenous ATP without affecting that by KCl and acetylcholine. Contractile responses to transmural nerve stimulation were markedly antagonized by a combination of $ANAPP_3$ and atropine

though not by either alone. Thus, both a purine and acetylcholine seemed to be involved in neurotransmission (20). Some questions remain unresolved. For example, whereas magnesium-free media potentiated the guinea pig bladder contraction by ATP, it enhanced responses to nerve stimulation only at above 4 Hz. It was inferred that ATP is released and contributes to contractile response only at higher frequencies (21). Quinidine was reported to block the contraction of this tissue by acetylcholine and 5-hydroxytryptamine as well as ATP (22).

Putative purinergic innervation in blood vessels is exemplified by the rabbit portal vein. This vein is known to be supplied by contractile adrenergic nerves and also by nonadrenergic, noncholinergic relaxant nerves. It effectively incorporated ^{3}H-adenosine into ^{3}H-ATP. Subsequent transmural nerve stimulation elicited an efflux of tritiated adenosine, inosine, and nucleotides. The efflux was unaffected by phenoxybenzamine but roughly halved by guanethidine and abolished by tetrodotoxin. ATP and adenosine were potent relaxants. It was proposed that an ATP-like purine is released as the transmitter from the nonadrenergic nerves and, further, as a relaxant cotransmitter from the adrenergic nerves to modulate the contractile action of the principal transmitter NE (23). Adrenergic nerve destruction by 6-hydroxydopamine also reduced the fractional release of ^{3}H-purines by 55% (24). Similar efflux results have been obtained in the portal vein of the rabbit, but not guinea pig, by other investigators (25). These authors also described fluorescent nerve plexus, nerve bundles, and ganglion cells following treatment with quinacrine, which binds ATP. The relaxant response to nonadrenergic nerve stimulation is selectively enhanced by dipyridamole and suppressed by theophylline, known as blockers respectively of adenosine uptake and of its receptor (26). These results are consistent with purinergic transmission and suggest that the transmitter, perhaps released as ATP, acts after hydrolysis into adenosine. However, the existence of nonpurinergic cotransmitter cannot be eliminated, and the ultrastructural evidence is quantitatively at variance with purinergic transmission in this vein (27).

Alternative Views to Purinergic Transmission

A number of objections to the purinergic transmission hypothesis have been raised, though they tend to be isolated observations. The effect of exogenously applied adenosine derivatives, but not stimulation of nonadrenergic, noncholinergic neurons, is enhanced by dipyridamole and inhibited by tachyphylaxis to adenosine and ATP, for example in the opossum esophageal sphincter (28). ANAPP$_3$ was recently reported to inhibit relaxation of the guinea pig taenia coli by ATP, ADP, AMP, and adenosine, but it failed to inhibit relaxant responses to transmural electrical stimulation (29).

Other discordant findings include those on the bladder, trachea, stomach, taenia coli, and sphincter and anococcygeus muscle (see 8, 30).

Substances proposed as neurotransmitters in the enteric nervous system vary from norepinephrine and acetylcholine to dopamine, GABA, vasoactive intestinal polypeptide (VIP), substance P, somatostatin, enkephalin, gastrin, cholecystokinin, bombesin, neurotensin, and ATP. VIP has increasingly become a viable alternative to ATP as an inhibitory transmitter. This polypeptide as well as several others is demonstrable by immunohistochemistry, radioimmunoassay, or bioassay in neurons throughout the gastrointestinal tract of a variety of mammals. It is released when enteric inhibitory nerves supplying the cat stomach are activated by esophageal distension or electrical stimulation. The gastric relaxation following esophageal distension is mimicked by intraarterial VIP infusion. By immunochemical techniques VIP cell bodies in the mesenteric plexus are seen to project in the anal direction and supply the circular muscle coat, as would be predicted for the descending inhibitory function of the enteric inhibitory nerves (see 9). VIP-containing nerve fibers have been immunocytochemically demonstrated around many peripheral blood vessels of the cat, and it been suggested that this peptide participates in blood flow regulation (31). Ultrastructural studies have lent additional support to the polypeptide concept. Nerves containing smaller (less than 140 nm in diameter) "large opaque vesicles," or the "small p-type," have been suggested as the nonadrenergic, noncholinergic nerves in several organs, and VIP has been electronmicroscopically identified in such nerve profiles (8). On the other hand, VIP, substance P, somatostatin, enkephalin, and neurotensin failed to mimic, as ATP did, the relaxant responses to nonadrenergic, noncholinergic nerve stimulation in the guinea pig taenia coli (32). Apamin diminished the responses to the nerve stimulation and exogenous ATP but not those to VIP. Conversely, only the responses to VIP were abolished by a proteolytic enzyme, α-chymotrypsin (33).

Whether these contradictory results and inconsistencies can be reconciled awaits further investigation. They seem to arise in part from lack of highly specific antagonists, with the welcome exception of $ANAPP_3$, and failure to distinguish between nucleoside and nucleotide(s) as the active ingredient, compounded by considerable organ- and species-differences. Further, varied nonadrenergic, noncholinergic transmitter substances probably subserve different types of enteric nerves (34).

ATP as a Cotransmitter

The possible corelease of ATP and NE from adrenergic nerves was earlier noted in the guinea pig taenia coli. Following incorporation of ^{3}H-adenosine mainly into ATP, stimulation of the perivascular sympathetic nerve elicited

an efflux of tritiated material that was abolished by guanethidine, an adrenergic neuron blocking agent (35). Similar reports followed. More recently, release of tritiated compound was shown during sympathetic nerve stimulation in the guinea pig vas deferens pretreated with ^{3}H-adenosine (36). $ANAPP_3$ was earlier shown to inhibit the contractile response of the vas deferens to ATP but not to norepinephrine (NE) or acetylcholine (37). The same agent diminished the nerve stimulation-induced contraction without affecting NE release. The contraction was reduced only slightly by an α_1-antagonist, prazosin, but markedly by combined use of $ANAPP_3$ and prazosin. The initial phasic contractile phase was especially susceptible to $ANAPP_3$, and the secondary tonic phase to prazosin (38). ATP is a potent contractile agent and, furthermore, ATP and NE potentiate each other in this tissue (39). Thus, ATP was proposed to originate from adrenergic neurons and act as a transmitter concurrently with NE (36, 38). Most recently, the excitatory junction potentiates (EJPs) in this tissue were found to be resistant to prazosin but suppressed by $ANAPP_3$. These results suggest that ATP mediates the EJPs, which initiate an action potential and the twitch, whereas NE mediates the tonic contraction (40). Similarly, because botulinum neurotoxin blocked contractile response to field stimulation in the presence of atropine and guanethidine, corelease of ATP and acetylcholine as transmitters from parasympathetic nerves in the guinea pig bladder was postulated (41).

A similar proposal has been made of the myenteric plexus of the guinea pig ileum. 6-Hydroxydopamine reduced the NE content in the myenteric synaptosomes, or varicosities, from the guinea pig ileum and reduced the ATP release induced by K^+ or veratridine (42). It is noteworthy, however, that corelease of ATP with the (other) transmitter does not necessarily assure its cotransmitter role.

PURINERGIC NEUROMODULATION

Neurotransmitter release comes under the influence of various neurohumoral agents. NE release from adrenergic nerves is generally thought to be subject to feedback inhibition by NE and to alteration by numerous other agents (43). Local purine release during adrenergic nerve stimulation was demonstrated in vasculature (23, 35) and in many other organs since. Adenosine and its nucleotides inhibit NE release from adrenergic nerves, as recently reviewed by Paton (44). It was therefore postulated that a purine or purines play an endogenous feedback neuromodulator role, regulating the adrenergic transmitter release (45, 46). The mechanisms of release and action of purines are beginning to be delineated.

Inhibition of Norepinephrine Release by Purines

In a variety of organs, adenosine and adenosine nucleotides depress the effector cell response to adrenergic nerve stimulation more than that to exogenous NE. Further, following ^{3}H-NE labelling, they diminish the nerve stimulation-induced ^{3}H efflux, indicative of inhibition of adrenergic transmitter release. These organs include the adipose tissue and blood vessels of the dog; kidney, blood vessels, and heart of the rabbit; vas deferens, portal vein, and salivary gland of the rat; and the guinea pig vas deferens and heart (see 47).

The exogenously administered adenosine and ATP are generally of comparable potency, with a threshold concentration of about 0.1 μM. The inhibition is immediate in onset and not tachyphylactic. The deamination metabolites formed, inosine and hypoxanthine, are practically devoid of activity. The decrease in ^{3}H-NE efflux is not due to accelerated neuronal or extraneuronal uptake of the released NE but rather to inhibition of its release. Nor does it depend on the NE- and PG-mediated feedback pathways (46, 48–50).

Although adenosine is known as a stimulator of cyclic AMP formation, this cyclic nucleotide may contrarily facilitate NE release (51). Inhibition of NE release by adenosine is significantly blocked by theophylline at concentrations too low to inhibit phosphodiesterase, and it is resistant to two potent inhibitors of this enzyme, Ro 20–1724 and ZK 62.711 (52).

Adenosine inhibits ^{3}H-NE efflux evoked by adrenergic nerve stimulation or elevated K^+ concentration, but not that induced by tyramine in isolated blood vessels (46, 53) or heart (49). This suggests that adenosine only interferes with the Ca^{2+}-dependent transmitter release. It was suggested that adenosine reduces the influx of extracellular Ca^{2+} (54), and reported that it decreases ^{45}Ca uptake by synaptosomes stimulated by high K^+ (55). Other authors find it more likely that adenosine interferes with the coupling role of the activator Ca^{2+} or accelerates its extrusion from the neuroplasm (56).

An apparent dichotomy exists as adenosine inhibits NE release but potentiates the postsynaptic NE effects in the rabbit kidney (45) and guinea pig vas deferens (39). Thus, postsynaptic inhibitory or facilitatory modulation of transmitter effect may occur where purine concentration reaches a sufficient level. Interestingly, NE release from the feline left auricle, spleen, and nictitating membrane is unaffected by adenosine (54, 57, 58).

Presynaptic Purinoceptor

Classification of purinoceptors into P_1 and P_2 types was suggested by Burnstock in 1978. P_1 is preferentially activated by adenosine and AMP and competitively blocked by methylxanthines, and appears to be related to the

adenylate cyclase system in some cases; P_2 is most sensitive to ATP and ADP, is blocked by PIT and high concentrations of quinidine and 2-substituted imidazoline, and may be related to induction of prostaglandin synthesis (see 59).

The presynaptic inhibitory purine action on adrenergic transmitter release is antagonized by theophylline but not PIT in the rat vas deferens (60) and vascular preparations (61). Nor is it affected by $ANAPP_3$ in the guinea pig vas deferens (38). The selectivity of adenosine and adenine nucleotides is complicated by the hydrolysis of the latter, the tissue uptake and deamination of adenosine, and potential adenosine release by nucleotides (62). Unlike adenosine or ATP, β,γ-methylene-ATP (APPCP) failed to depress the contractile response of the canine saphenous vein to adrenergic nerve stimulation. This suggested that ATP had to be hydrolyzed to adenosine, which exerted the inhibition (62). The fact that the presynaptic inhibitory action of ATP, ADP, and AMP, as well as adenosine, in the rat vas deferens was antagonized by theophylline and potentiated by an adenosine uptake inhibitor was explained on the same basis (60). Further support was given by demonstration of formation of adenosine from AMP, ADP, and ATP applied to the rat vas deferens, and reduction of the effect of nucleotides as well as adenosine by addition of adenosine deaminase (63). These findings are compatible with the P_1 receptor as the mediator of the presynaptic inhibition. The latter, however, does not appear to be causally related to the adenylate cyclase system (see 64). For the same reason, the relationship between P_1 and the other adenosine receptor nomenclature R_a/R_i or A_1/A_2, pertaining to modulation of adenylate cyclase activity, remains uncertain. The structure-activity relationship for presynaptic inhibition by adenosine has been reviewed by Paton (65). The P_2 receptor is also present at the adrenergic nerve terminals in view of the phentolamine-sensitive release of NE by ATP in high concentrations (61).

Purine Release by Adrenergic Nerve Stimulation

Like the guinea pig taenia coli noted above, the rabbit portal vein and aortic adventitia that contain adrenergic nerves incorporate ^{3}H-adenosine mainly into ATP. Transmural nerve stimulation elicited a ^{3}H efflux that was blocked by tetrodotoxin or bretylium (in adventitia) but not by phenoxybenzamine. This occurred in the absence of muscle contraction in the adventitial preparation. Adrenergic neurons, therefore, were suggested as the main source of purine release, possibly by exocytosis (23).

Purine release by sympathetic nerve stimulation has since been observed in the canine and feline adipose tissue, rabbit kidney and heart, and cat nictitating membrane by Fredholm and his associates by use of ^{3}H-adenosine or assay of endogenous adenosine. These authors maintain that purines are predominantly formed by the postsynaptic structures (47, 66), recently

referred to as effector tissues not necessarily at subsynaptic location (67). This view is derived from their findings in the canine subcutaneous adipose tissue, rabbit kidney and heart, rat vas deferens, and cat nictitating membrane, that adrenergic α-receptor blocking agents diminish purine release induced by sympathetic nerve stimulation and that exogenous NE and angiotensin II cause purine release. Finally, in the cat nictitating membrane, purine release was induced by NE, tyramine, and acetylcholine as well as sympathetic nerve stimulation in amounts correlated with the contractile responses to these stimuli (68). Thus, smooth muscle contraction or membrane depolarization and/or local energy imbalance is deemed responsible for purine release during nerve activity (47). These authors envisage "transsynaptic modulation" of transmitter release, in an analogy to the "retrograde inhibition" of acetylcholine release by ATP in the Torpedo electric organ (69). Fredholm et al (70) found that phentolamine increased ^{3}H-NE release and abolished ^{14}C-purine release, whereas adenosine decreased release of ^{3}H-NE but not ^{14}C-purine, and that the ATP-NE ratio was very low in the small NE storage vesicles, in the rat vas deferens. They therefore believe exocytotic purine release is unlikely in this organ.

On the other hand, other investigators have presented new evidence for neurogenic purine release. Westfall and colleagues (36) used hypertonic solution to prevent the electrical stimulation-induced contraction of the guinea pig vas deferens, but it failed to prevent the tetrodotoxin-sensitive ^{3}H efflux from tissues preincubated with ^{3}H-NE or ^{3}H-adenosine. Thus, the purine release seemed unlikely to be secondary to muscle contraction. They further used 6-hydroxydopamine for degeneration of adrenergic nerves. The residual contractile response to transmural electrical stimulation, presumably resulting from cholinergic fibers, was unaffected by ANAPP$_3$, which in the intact vas deferens effectively blocked the contraction. This was taken as evidence for normal adrenergic neuronal release of purines along with NE as a cotransmitter (38). Whether the disparity with the conclusions of Fredholm et al (70) can be attributed to species difference is uncertain. The neuronal purine release was analyzed in the isolated rabbit portal vein pretreated with ^{3}H-NE or ^{3}H-adenosine (24). The α_1-adrenoceptor blocker, prazosin, did not affect the ^{3}H-NE efflux by transmural electrical stimulation. It largely blocked the contraction and reduced the ^{3}H-purine efflux by 20%, suggesting that 80% of the release was of neuronal origin. These authors reasoned that if NE and ATP were coreleased by exocytosis, alterations in neuronal purine release should parallel those in NE release. As expected, yohimbine, which preferentially blocks the presynaptic α_2 receptors, promoted, and clonidine, an α_2 agonist, reduced the fractional releases of ^{3}H-NE and ^{3}H-purines to a similar extent. Similarly, in the dog basilar artery, phentolamine, which blocks both α_1 and α_2 receptors, augmented the transmural stimulation-induced ^{3}H-NE and

^{3}H-purine effluxes, and exogenously applied NE inhibited the latter efflux (71).

In another approach, muscle-free varicosities were separated from the guinea pig ileum myenteric plexus. Depolarizing agents, K^+ and veratridine, induced a Ca^{2+}-dependent release of undegraded ATP, perhaps along with NE or acetylcholine (72). ATP release from varicosities was also evoked by acetylcholine and nicotine (73), which activates the nonadrenergic inhibitory neurons.

The origin of purines in the ^{3}H-adenosine labeled pulmonary artery of the rabbit has been reexamined by Katsuragi & Su (74). KCl was used in the place of electrical nerve stimulation. KCl (30–70 mM) markedly enhanced the ^{3}H efflux, which was prevented by removal of Ca^{2+} and denervation with 6-hydroxydopamine or cold storage, but not significantly affected by phentolamine. Epinephrine (5 μM) also elicited a ^{3}H efflux, but this action was inhibited by phentolamine and not by cold storage or 6-hydroxydopamine. In the nerve-free, smooth muscle-containing medial strip of the thoracic aorta, KCl evoked a very small ^{3}H efflux that was not Ca^{2+}-dependent. Thus, the KCl-induced efflux was largely attributable to adrenergic nerves. Postsynaptically, the transmitter NE as well as epinephrine may additionally release purines mediated by α-receptors (74). The KCl-induced ^{3}H-purine efflux from the pulmonary artery was enhanced by clonidine, reported to be a P_1-receptor blocker (75), and by theophylline. It was suppressed by adenosine or ATP. The epinephrine-induced ^{3}H efflux was not increased by theophylline or clonidine. These results are in keeping with presynaptic autoinhibition of purine release and, hence, presynaptic purine release from vascular adrenergic nerves. Moreover, since KCl and clonidine contracted the denervated and normal arterial preparations, respectively, with little or no ^{3}H-purine efflux, the efflux can be dissociated from smooth muscle depolarization or contraction (76, 77).

Clearly, discrepancies exist among investigators. However, the evidence for adrenergic neuronal and exocytotic purine release seems strong enough to warrant its further investigation. It may be noted that electrical stimulation evokes purine release from nerve fibers of the nonmyelinated autonomic nerve trunk. However, mostly inosine and hypoxanthine are released with little or no adenosine and nucleotides in a Ca^{2+}-independent manner (78).

Functional Significance of Endogenous Purines

The neuromodulator role of locally released purines is beginning to be delineated. Dilazep in combination with erythro-9-(2-hydroxy-3-nonyl) adenine, inhibitors of adenosine uptake and deamination, respectively, significantly reduced the ^{3}H-NE overflow and contraction induced by trans-

mural stimulation in the rat portal vein (79). Another adenosine uptake inhibitor, dipyridamole, inhibited cardioaccelerator effect of sympathetic nerve stimulation in the anesthetized dog (80). Theophylline (10–100 μM) slightly enhanced the stimulated ^{3}H-NE efflux in the rabbit kidney and rat portal vein (52, 81). These results are consistent with the view that endogenous adenosine plays a modulator role in adrenergic neurotransmission.

On the other hand, theophylline in concentrations sufficient to antagonize exogenously applied adenosine did not affect contraction of the rat vas deferens or ^{3}H-NE efflux in the dog basilar artery in response to sympathetic nerve stimulation (60, 71). Diazepam was shown to potentiate the inhibitory action of adenosine, possibly by inhibition of its uptake, in the rat vas deferens. However, this agent per se had no effect on its contractile responses (82).

Analyses with these agents are compounded by their multiple actions, and the purine concentrations achieved at the presynaptic receptors remain difficult to ascertain. Where purinergic negative feedback has been detected, it appears to be modest in extent compared to the adrenergic feedback. The pathological consequences can only be speculated. The mesenteric vasculature of the spontaneously hypertensive rats was found to have diminished purinergic inhibition of adrenergic nervous vasoconstriction. This alteration can potentially contribute to the hypertensive state (83, 84). Many other pathophysiological purine roles are possible (3). The significance of circulating or "metabolically" released purines and the purinergic modulation of cholinergic neurotransmission are beyond the scope of this discussion.

SUMMARY AND CONCLUSIONS

Conflicting views abound on the peripheral neurotransmitter and neuromodulator roles of purine compounds. Substantial organ- and species-related variations have become apparent. There is, however, a body of compelling evidence for such roles, if not so broad and ubiquitous as those envisioned (7) for the central nervous system.

The variations may in part be attributable to the neuroeffector synaptic geometry. The transmitter concentration found at the postsynaptic membrane drops precipitously with increase in the synaptic cleft (85). Where the cleft is narrow, a purine may serve as the primary or sole transmitter (purinergic nerve) and presynaptic modulator. Alternatively, from nonpurinergic (e.g. adrenergic) nerves a purine may be released, possibly ATP by exocytosis, to act as a cotransmitter. It may also serve as pre- and postsynaptic modulator, potentially with a contribution from postsynaptic release (Figure 1). The purine could conceivably diffuse and affect other varicosities. Where the cleft is wide, the postsynaptic concentration of the

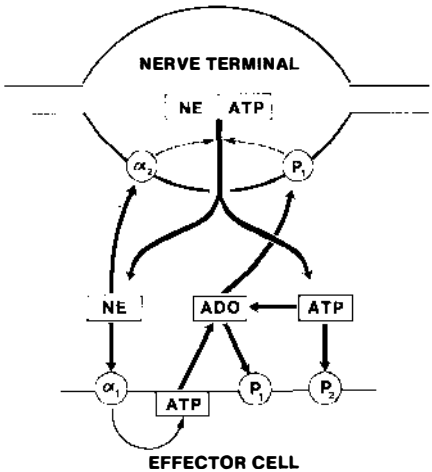

Figure 1 Purinergic cotransmission and modulation at adrenergic neuroeffector synapse. NE and ADO represent norepinephrine and adenosine. α_2 and P_1 receptors mediate presynaptic inhibition. Postsynaptic adreno- and purinoceptors vary in population and effect with effector cells. Modulation of NE actions and release of purines by purines are not shown.

neurogenic purine may be too low to permit a transmitter or postsynaptic modulator function. The concentration of the nonpurine transmitter may also be insufficient to elicit a significant postsynaptic purine release. The neuronally released purine may, however, presynaptically exert inhibition of transmitter release much as in a narrow cleft.

It seems, therefore, that the origin of synaptic purines and their function, be it transmitter, cotransmitter, or modulator, are dictated at least in part by the characteristics of the purine pools, purinergic receptors, and synaptic configuration, which await further assessment.

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