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Review Article

Regulation of innate immunity by extracellular nucleotides

Stefania Gorini, Lucia Gatta, Laura Pontecorvo, Laura Vitiello, Andrea la Sala

Laboratory of Molecular and Cellular Immunology, IRCCS San Raffaele Pisana, Italy

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Abstract: Extracellular ATP (eATP) is the most abundant among extracellular nucleotides and is commonly considered as a classical danger signal, which stimulates immune responses in the presence of tissue injury. In fact, increased nucleotide concentration in the extracellular space is generally closely associated with tissue stress or damage. However non-lytic nucleotide release may also occur in many cell types under a variety of conditions. Extracellular nucleotides are sensed by a class of plasma membrane receptors called P2 purinergic receptors (P2Rs). P2 receptors are expressed by all immunological cells and their activation elicits different responses. Extracellular ATP can act as an initiator or terminator of immune responses being able to induce different effects on immune cells depending on the pattern of P2 receptors engaged, the duration of the stimulus and its concentration in the extracellular milieu. Millimolar (high) concentrations of extracellular ATP, induce predominantly proinflammatory effects, while micromolar (low) doses exert mainly tolerogenic/immunosuppressive action. Moreover small, but significant differences in the pattern of P2 receptor expression in mice and humans confer diverse capacities of ATP in regulating the immune response.

Keywords: Extracellular nucleotides, P2 purinergic receptors, extracellular ATP, innate immunity

Extracellular nucleotides

Nucleotides (ATP, ADP, UTP and UDP) are among the most ancient biologic molecules and this is consistent with their multifunctional role in living organisms. Nucleotides are the constituents of nucleic acids, represent an intracellular energy source and serve as substrate in signal transduction pathways. Intracellular nucleotides can be also massively released in the extracellular space and play a role in intercellular communication [1-3].

ATP is the most abundant among nucleotides. Intracellular concentration of ATP ranges between 1 and 10 mM while, in normal conditions, the extracellular compartment contains ATP in the low nanomolar concentration range. Because of such steep concentration gradient, ATP small size and high mobility, a dramatic increase of ATP concentration can occur in the extracellular space around damaged cells leaking their cytoplasmic content [3-7].

ATP can be also actively released by many different cell types under certain conditions. Activated platelets represent one of the most abundant source of actively released adenine nucleotides [8-10]. ATP is also released from vascular endothelial cells under mechanical or shear stress [11-13]. In addition, ATP secretion from endothelial cells as well as from leukocytes can be induced by pathogen-associated molecules [7, 14-17]. T lymphocytes secrete ATP in the early stages of activation [18]. Moreover commensal bacteria in the gut are able to secrete ATP exerting relevant modulatory effects on immune responses [19]. Finally ATP is released during the early stages of apoptosis inducing monocyte/macrophages recruitment acting as a "find me" signal to exert an efficient cell clearance [20]. Of note, different eukaryotic cells use different mechanisms to release ATP. For example, under proper stimulation, neurons and platelets secrete adenine nucleotides stored in cytoplasmic vesicles [21, 22]. In other cell types, such as T lymphocytes, PMN neutrophils and monocyte/macrophages, ATP is released in response to increased cytosolic calcium concentration through pannexin (panx)-1 hemichannels [18, 23-25]. Alternative-

Table 1. Agonists binding affinity (EC_{50}) for all P2 receptors and their main downstream signaling events

Receptor	Agonists	Agonists binding affinities EC50(uM)	Main downstream signaling events
P2X Receptors			
P2X1	ATP	0.05-1	Ca^{2+} / Na^{+} influx
P2X2	ATP	1-30	Ca^{2+} influx
P2X3	ATP	0.3-1	Cations influx
P2X4	ATP	1-10	Ca^{2+} influx
P2X5	ATP	1-10	Ion influx
P2X6	ATP	1-12	Ion influx
P2X7	ATP	>100	Cations influx and pore formation
P2Y Receptors			
P2Y1	ADP	8	PLCβ activation
P2Y2	ATP,UTP	0.1(ATP), 0.2(UTP)	PLCβ activation cAMP inhibition
P2Y4	UTP (ATP,UTP)	2.5	PLCβ activation cAMP inhibition
P2Y6	UDP,UTP	0.3 (UDP), 6 (UTP)	PLCβ activation
P2Y11	ATP	17	cAMP increase, PLCβ activation
P2Y12	ADP	0.07	cAMP inhibition
P2Y13	ADP,ATP	0.06 (ADP), 0.26 (ATP)	cAMP inhibition
P2Y14	UDP-glucose	0.1–0.5	PLCβ activation

ly, ATP can be released through the opening of volume-sensitive channels [26], purinergic X receptors (P2X)-gated channels [27, 28] or by the opening of connexin 43 channels upon mechanical stress [29]. Different secretion pathways are used by different cells of the immune system for ATP release depending on the nature of the activating stimulus and/or pathophysiological condition.

Purinergic receptors

Once in the extracellular space, nucleotides bind to specific plasma membrane receptors, named P2 receptors, widely distributed in a variety of different organisms such as mammals, plants, yeasts and bacteria, suggesting that nucleotides represent an archaic communication system [30, 31]. All eukaryotic cells express P2 receptors and nucleotides trigger intracellular signaling pathways in almost every tissue. Intracellular signaling pathways activated by P2 receptors depend on cell type, pattern of P2 receptors expressed and type/quantity of released nucleotides. Two P2 receptor subfamilies have been described so far: P2X and P2Y [32-34]. P2 receptors signaling altogether cooperate in determining the basal level of cell activation for signal transduction pathways [35]. Moreover, a wide variety of physiological functions are regulated by P2 receptors, including the regulation of cell volume, tissue blood flow and inflammation.

The P2X subfamily is composed of seven members named P2X1-P2X7. P2X receptors are ligand-gated ion channels selective for monovalent and divalent cations. The amino- and carboxyl-terminal domains of the P2X subtypes are both cytoplasmic. Upon activation, P2X subunits aggregate to form homo- or in some cases hetero-multimers and determine Ca^{2+} and Na^{+} influx and K^{+} efflux [34]. The only known physiological agonist for P2X receptors is ATP. P2X receptors were originally identified in mammalian sensory neurons, and subsequently also found in several additional cell types such as smooth muscle cells, fibroblasts, megakariocytes, platelets, lymphocytes, macrophages, granulocytes, dendritic cells [36, 37].

P2Y receptors are widely expressed, being present in platelets [38, 39] mucosal cells [40, 41], monocytes [42, 43], macrophages [43, 44], dendritic cells [45-47], NK cells [48], granulocytes [49-51], neurons [52, 53], smooth and striated muscle cells [54-58]. Eight P2Y subtypes have been cloned (P2Y1, P2Y2, P2Y4, P2Y6, P2Y11, P2Y12, P2Y13, and P2Y14) [59-62]. P2Y receptors are seven membrane-spanning, G-protein-coupled receptors whose activation exerts different effects depending on the G protein subtype involved. P2Y1, 2,4,6, and 11 are coupled to $G_{q}/_{11}$ proteins that trigger the generation of inositol 1,4,5-trisphos-

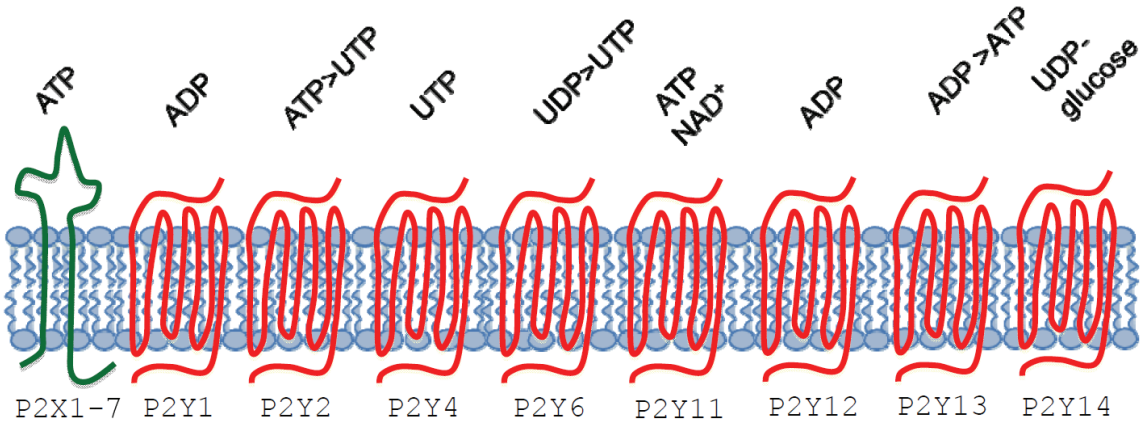

Figure 1. *Type 2 purinergic receptors and their nucleotide agonists*. Extracellular nucleotides bind to type 2 purinergic receptors exerting their effects on cells' function. Two distinct P2 receptor subfamilies were described P2X and P2Y. P2X receptors are membrane cation channels gated exclusively by extracellular ATP. Seven P2X receptors have been cloned and named P2X1-7. They are oligomers of three subunits each composed by an extracellular loop, two transmembrane domains and an amino- and a carboxyl-terminal both cytoplasmic. ATP binding induce the subunits assembly to form omo- or etero-multimerc channels permeable to monovalent and divalent cations. P2Y receptors are seven membrane-spanning, G-protein-coupled receptors. Eight P2Y subtype receptors have been cloned so far named P2Y1, P2Y2, P2Y4, P2Y6, P2Y11, P2Y12, P2Y13 and P2Y14. They can be subdivided into adenine nucleotide-preferring receptors (P2Y1, P2Y11, P2Y12 and P2Y13), uracil nucleotide-preferring receptors (P2Y4 and P2Y6), a receptor of mixed specificity (P2Y2) and a UDP-glucose-preferring receptor (P2Y14).

phate and release of Ca^{2+} from the intracellular stores. P2Y12, 13 and 14 are associated with $G_{i/0}$ proteins, which inhibits adenylate cyclases [63]. Of note, P2Y11 activation is associated with increased intracellular cAMP concentration [45, 64, 65]. As each P2Y family member display different affinity for diverse ligands, each receptor is characterized by a distinct agonist rank of potency (see **Table 1** and **Figure 1**). P2Y1, P2Y11, P2Y12 and P2Y13 are activated by ATP or ADP. P2Y2 is activated both by UTP and ATP; P2Y4 and P2Y6 have UTP and UDP as agonists, whereas UDP-glucose activates the P2Y14 subtype.

Extracellular metabolism of nucleotides

The nature and intensity of purinergic signaling depend on extracellular nucleotide/nucleoside concentrations, which are controlled by a family of ectoenzymes known as ecto-nucleoside triphosphate diphosphohydrolases (E-NTDPase 1, 2, 3 and 8). CD39/ENTPD1 ectonucleotidase (CD39) is expressed by monocytes, NK cells, T and B lymphocytes and dendritic cells [66, 67]. It can hydrolyze tri- and di-phosphate nucleosides, but is not able to hydrolyze monophosphate nucleosides [68]. Regulation of extracellular ATP concentration by ATP scavenging CD39 has been shown to regulate immune cells function and inflammation in different settings [66, 67, 69, 70].

Another important membrane-bound enzyme involved in the metabolism of extracellular nucleotides is CD73/ecto-5'-nucleotidase. It catalyzes the hydrolysis of adenosine–monophosphate (AMP) generating adenosine that is in turn recognized by P1 adenosine receptors. Interestingly CD39 and CD73 are simultaneously expressed on the same cellular population as occurs for example on murine T regulatory lymphocytes (Tregs) and on a subset of human Tregs [71] or on human monocyte-derived dendritic cells [72]. Effects due to ATP catabolites rather than to ATP itself can be distinguished by comparing the observations made using ATP with those obtained with non-hydrolyzable ATP analogues (e.g. ATP-γ-S), adenosine deaminase (ADA; that converts adenosine into inosine) or exogenous apyrase that hydrolyzes extracellular ATP.

Regulation of innate immunity by extracellular nucleotides

The innate immune system is the first line of defense against invading pathogens. Four major pattern recognition receptor (PRR) families, are involved in the recognition of a wide

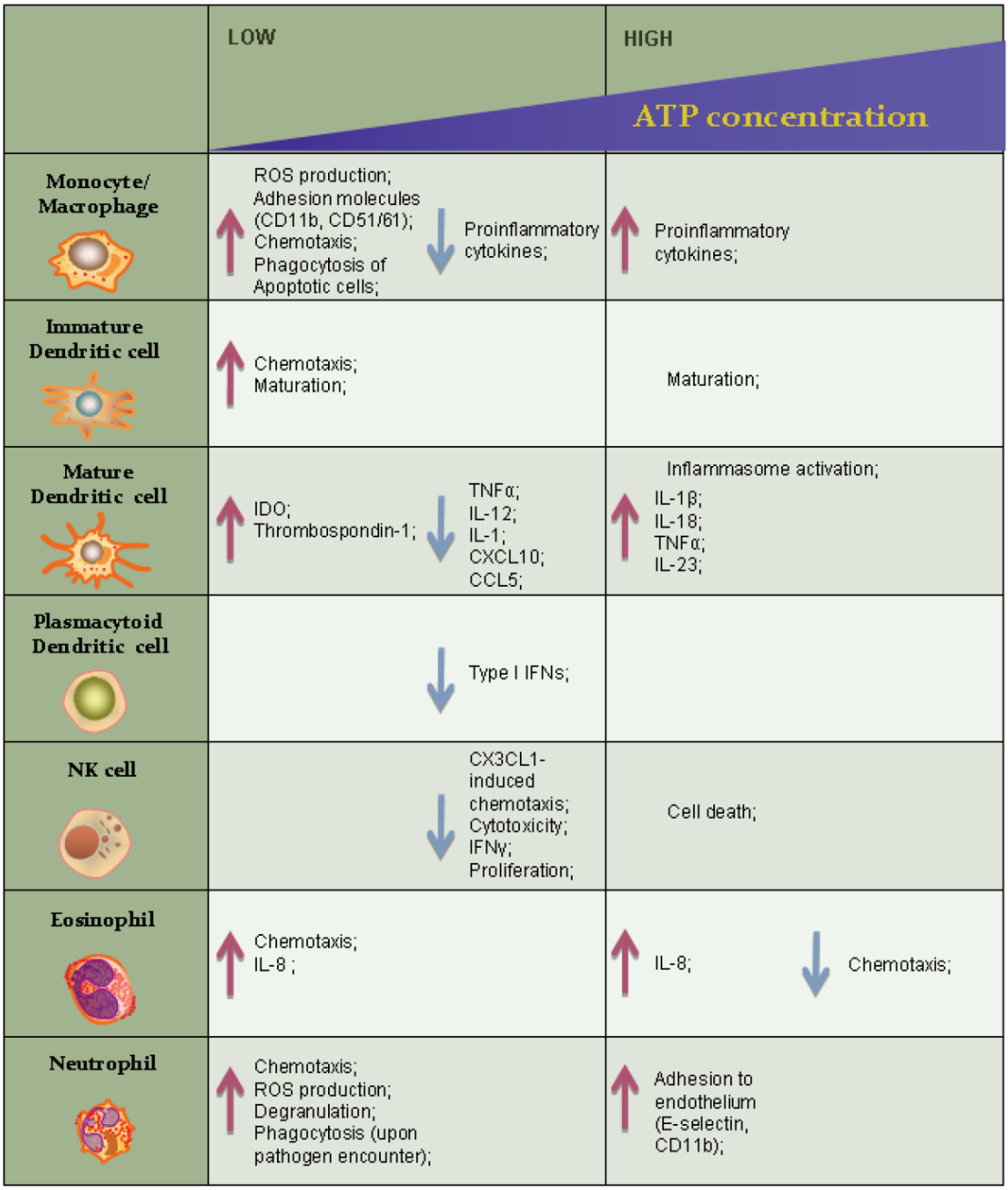

Figure 2. *Extracellular ATP exerts different effects on cells of the innate immunity depending on its concentration.* Low (1-250 μM) eATP concentrations activate high and intermediate ATP-binding affinity P2 receptors. High (1-10 mM) concentrations of eATP activate P2X7 receptor which displays low ATP-binding affinity.

range of pathogen-associated molecular patterns (PAMPs): Toll-like receptors (TLRs), cytosolic RIG-I-like receptors (RLRs), Nod-like receptors and C-type lectins. It is now clear that detection of foreign microorganisms is not sufficient to induce inflammation, but recognition of a damage signal is also necessary.

For example, DCs reside in peripheral tissues and serve as "sentinels", and they are not only activated upon encounter with foreign pathogens recognized by Toll like receptors, but they also react to the presence of environmental molecules associated with tissue stress, the so-called damage-associated molecular pat-

terns (DAMPs). Constitutively expressed endogenous molecules can function as danger signals as for example ATP, adenosine, high mobility box group 1 (HMBG1) and heat shock proteins, while other danger signal are inducible factors such as type I interferons [73]. The recognition of endogenous danger signals by cells of the immune system participate in determining the quality and the strength of the immune response and enables the immune system to distinguish between pathogenic or harmless/commensal organisms.

In order to maintain homeostasis the termination of the immune response and the resolution of inflammation is as important as its initiation and tissue damage might be caused by the intrinsic toxicity of sustained inflammation. Extracellular ATP can act as an initiator or terminator of immune responses. Relatively high concentrations of extracellular ATP (in the millimolar range) induce predominantly proinflammatory effects through the engagement of the low affinity receptor P2X7. On the other hand low (micromolar) doses exert mainly tolerogenic/immunosuppressive action (**Figure 2**) through the activation of the high affinity P2Y11 receptor [74, 75].

Monocytes/macrophages

Macrophages continuously differentiate from monocytes that leave blood flow to reach the tissues throughout the body. When a potentially pathogenic microorganism crosses the epithelial barrier is immediately recognized by macrophages that reside in the host tissues and that are able to pahgocyte and kill it.

In macrophages, millimolar extracellular ATP engages P2X7 and trigger the activation of the inflammasome [16, 76]. K^+ efflux occurring through the opened P2X7 channel is a key event leading to the assembly of the Natch Domain-, Leucine-Rich Repeat-, PYD-Containing Protein 3 (NLRP3) inflammasome [77]. Two distinct triggering signals are necessary for macrophages to secrete IL-1 β and IL-18: the activation of Toll-like receptor pathway that determines the expression and accumulation of pro-IL-1β and pro-IL-18, and the engagement of P2X7 receptor that activate the inflammasome, composed by NLRP3, the ASC adaptor and procaspase1. Once activated, NLRP3 promotes the oligomerization of procaspase 1 and its subsequent proteolytic activation into active Caspase 1 that in turn cleaves pro-IL-1β and pro-IL-18 into active cytokines [78-81]. In keeping, macrophages from P2X7 KO mice display impaired NLRP3 inflammasome activation and reduced secretion of IL-1β and IL-18 after LPS stimulation [82]. As a consequence, in a monoclonal anti-collagen induced arthritis model, P2X7 KO mice develop less severe synovial inflammation as well as reduced cartilage destruction [83]. Moreover high levels (mM) of extracellular ATP increase macrophage secretion of inflammatory cytokines such as IL-1α [84], IL-1β [85-87], IL-6 [82], IL-18 [88, 89], TNF-α [90, 91], whereas low micromolar ATP concentrations sufficient to trigger the P2Y11 but not the P2X7 receptor, inhibit TNF-α and CCL-2 production while increasing the production of the immunoregulatory cytokine IL-10 [92]. Extracellular nucleotides have been shown to regulate several other cell functions in a P2X7 receptor-independent manner. For example macrophages exposed to micromolar levels of extracellular nucleotides, display increased ROS production, [44, 93, 94]. Such event in turn activates different signalling pathways leading to the production of macrophage inflammatory protein-2 (MIP-2), that promote migration of neutrophils toward inflamed tissues [95]. In addition micromolar levels of both extracellular ATP and ADP also induce chemotaxis of monocyte/macrophages [92, 96-98]. Phagocytic activity of macrophages is also influenced by extracellular nucleotides. Clearance of apoptotic cells is a crucial task performed by macrophages. Removal of apoptotic cells normally does not lead to upregulation of co-stimulatory molecules or cytokine production by macrophages and therefore does not contribute to or stimulate immune responses [99]. On the contrary upon encounter with necrotic cells macrophage proinflammatory activity is stimulated while phagocytosis is not. As dying cells release nucleotides and macrophages express most of purinergic receptors, Marques-da-Silva and colleagues recently investigated whether extracellular nucleotides could influence phagocytosis of murine macrophages through the activation of purinergic receptors [100]. Pretreatment of macrophages with low concentrations of several extracellular nucleotides, induced increased expression of adhesion molecules such as CD11b/CD18 (Mac-1) and CD51/61 and consequent

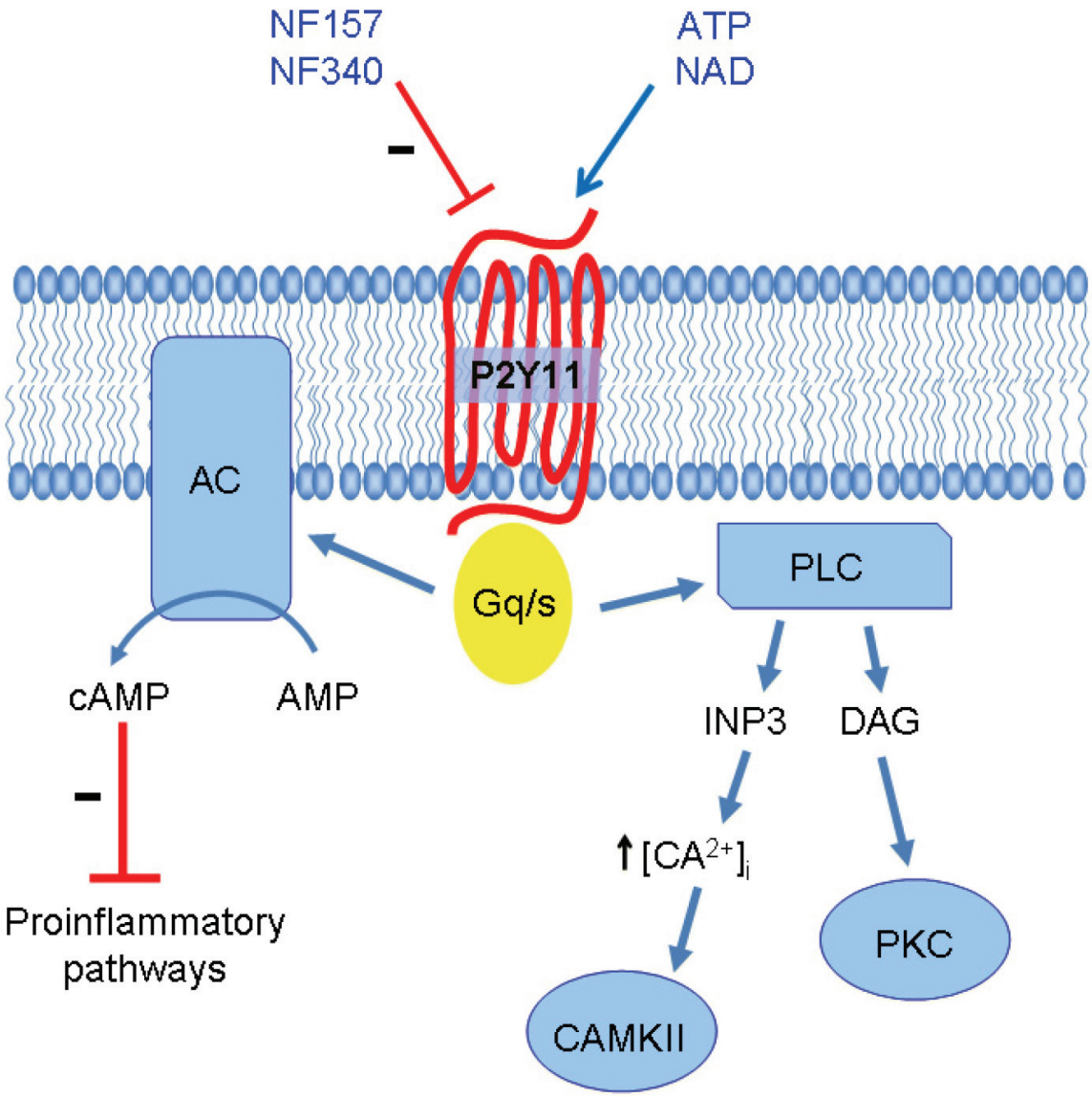

Figure 3. *Peculiarities of the P2Y11 receptor.* P2Y11 is expressed on human cells but has no ortholog in rodents. ATP is the P2Y11 preferred physiological ligand, it can also be activated by extracellular NAD^+ and NAADP. The P2Y11 is the only P2Y receptor coupled to a $G_{q/s}$ protein and upon activation stimulates adenylate cyclase (AC) as well as phospholypase C (PLC) activities. Consequent increased concentration of cAMP mediates several inhibitory effects of eATP on human cells of the innate immunity.

enhanced phagocytosis, possibly through the engagement of P2X1, P2X3 ,P2Y1 and/or P2Y6. This scenario is consistent with an homeostatic environment where low levels of nucleotides, released by apoptotic cells, stimulate macrophages to clear apoptotic bodies enhancing pahgocytosis. Higher concentrations of extracellular nucleotides, consistent with a necrotic environment, do not stimulate the upregulation of adhesion molecules, nor the clearance of necrotic cells, determinig the amplification of inflammatory effects exerted by necrotic debris.

Dendritic cells

Dendritic cells (DCs) are professional antigen presenting cells. They reside in tissues where they uptake the antigen and then migrate to lymph nodes toward cytokines gradients, to stimulate T cells. Extracellular ATP is able to induce immature (but not mature) DCs migration [101]. P2X7 activation on DCs is able to induce inflammasome activation as well as secretion of proinflammatory cytokines such as IL-1β, IL-18, TNF-α and IL-23. On the other hand,

dendritic cells maturing in the presence of micromolar concentrations of extracellular ATP display impaired production of TNF-α, IL-1β and IL-12 as well as reduced secretion of inflammatory chemokines such as CXCL-10, CCL-5, CCL-2 and CCL-3, while the expression of IL-10, IL-1 receptor antagonists or of CCL-17 and CCL-22 are either unaffected or upregulated [102-104]. In the same experimental setting, pharmacological inhibition of P2Y11 receptor restores the production of TNFα and IL-12 by DCs (la Sala et al., unpublished). Moreover extracellular ATP has been shown to induce the expression of two important immunosuppressive proteins: indoleamine 2,3-dioxygenase (IDO) and thrombospondin-1 via P2Y11 activation [105]. Of note the P2Y11 receptor is the only P2YR coupled to a Gs protein that in turn is able to activate adenylate cyclase determining an increase in intracellular cAMP concentration as depicted in **Figure 3**. Interestingly the treatment of DCs with different cyclic AMP elevating agents or cell-permeable cAMP analogs produce similar modifications of DC maturation process resulting in impaired capacity of DC to promote type 1 T cell responses [102, 106-108]. Depending on the microenvironment, extracellular ATP can promote immunogenic or tolerogenic activity of DCs. For example micromolar concentrations of eATP that block IL-12 expression elicited by LPS, synergize with TNF for the induction of IL-12p70 [45, 108]. Moreover, eATP has been shown to inhibit IL-27 secretion via P2Y11 activation and upregulate IL-23 mRNA expression through a P2Y11-independent mechanism [109].

The "dualism" between P2X7 as low affinity ATP receptor exerting mainly proinflammatory effects and P2Y11 an high affinity receptor triggering cyclic AMP-mediated immune suppression, determines a complex regulation of immune functions also in other leukocyte subpopulations. Importantly as no orthologue gene of the human P2Y11 receptor have been identified in rodents, murine cells converge in delineating a marked proinflammatory role for ATP, while in the human system both pro and anti-inflammatory effect have been documented [110]. Adding to such complex scenario, the duration of the stimulus must be taken into consideration as well. While P2X7 opening for a short time leads to the activation of the proinflammatory pathway to sustain inflammation, prolonged P2X7 receptor stimulation causes the enlargement of the pore that leads to cell death.

NK cells

Natural killer cells are bone marrow-derived circulating lymphocytes that contribute to the innate immune response by exerting cytolytic activity against virally infected and neoplastic cells and by secreting cytokines, especially IFN-γ. Extracellular ATP is a modulator of the activity of NK cells as well. It inhibits NK cells proliferation and IFN-γ production [111]. In addition NK cells cytotoxic activity and chemotaxis elicited by CX3CL1 is blocked by eATP, an effect that is mediated by the P2Y11 receptor [48]. CX3CL1 might have a role in the crosstalk between leukocytes and endothelial cells. Soluble CX3CL1 is released by activated ECs during early stages of inflammation, and is able to induce the recruitment of leukocytes expressing its cognate receptor CX3CR1. In addition, CX3CL1 triggers interferon-γ production by NK cells that reinforces CX3CL1 expression by ECs [112]. Moreover, activated ECs can express both the soluble and the membrane-bound form of CX3CL1, the latter acting as an adhesion molecule thus reinforcing the strength of leukocyte-endothelial cell interaction [113]. In addition CX3CL1 can stimulate the cytolytic activity of NK cells toward ECs [114]. Noteworthy ECs represent a major source of actively secreted ATP [115], pointing it out as an important player in the regulation of NK-EC interaction. It has been proposed that activated NK cells may mediate vascular injury in different pathological conditions such as vascular leak syndrome, allograft rejection, and cytomegalovirus infection [113, 114, 116]. In the presence of ATP, CX3CL1 failed to enhance NK cell–mediated cytolysis of endothelial cells. Most importantly increased degradation of extracellular ATP by exogenous apyrase significantly increased NK cells capacity to kill endothelial cell. In addition ATP influences NK chemotaxis by inhibiting CX3CL1-induced cell migration. Such effect is not due to a general inhibition of the capacity of NK cell to migrate because in the same experimental settings extracellular ATP has proven able to increase chemotaxis toward CXCL12 and enhance chemokinesis [48].

Polymorphonuclear cells (PMN)

Eosinophils are bone marrow-derived granulocityc leukocytes. Only few of these cells are

normally present in the circulation the majority of them residing in connective tissues, just under epithelium. Eosinophils exert two effector functions: upon activation they release highly toxic granule proteins and free radicals, and secrete several cytokines and chemical mediators to attract and activate other immunological cells. Extracellular ATP in low (micromolar) concentration enhances eosinophil migration toward inflamed tissues [117, 118].

In eosinophils, extracellular ATP increases intracellular calcium concentration through the opening of ion channels allowing Ca2+ influx from the extracellular space and by triggering Ca^{2+} release from the intracellular stores as well [119]. As actin reorganization is preceded by increased intracellular Ca^{2+} concentration, extracellular nucleotides, especially ATP, UTP and ADP, are able to induce a rapid and transient actin polymerization, in a concentration dependent manner [119, 120]. In addition, extracellular nucleotides trigger the secretion of eosinophil cationic protein, and IL-8, two potent chemoattractants recruiting other eosinophils and neutrophils [121, 122].

Polymorphonuclear neutrophilic leukocytes are short living cells very abundant in blood, but normally not present in healthy tissues. They share with macrophages a key role in innate immunity because they are able to recognise, ingest and kill many pathogens without an aid of adaptive immune response. Extracellular ATP enhances chemotactic response of neutrophils [24, 123].

Noteworthy, neutrophils express P2 receptors [50, 124, 125], and they are able to actively secrete ATP [126]. Neutrophils can transiently but rapidly secrete ATP through panx1 and connexin 43 channels, from the protruding edge of the cell during migration. ATP activate P2Y2 receptors through an autocrine pathway and at the same time ATP is hydrolyzed by ectonucleotidases to adenosine that engages the Gi-coupled A3 receptor. These two concomitant mechanisms determine an amplification of chemotaxis [123]. Neutrophils also express a maxi-anion channel known as human tweety homolog 3 (hTTYH3), which upon cell activation by N-formil-Met-Leu-Phe bacterial peptide receptors (FPRs), is able to secrete ATP. Noteworthy panx1 hemichannels colocalize with FPRs and hTTYH3 at the leading edge of migrating neutrophils, delimiting an area for active ATP release [24]. ATP secretion by neutrophils is induced not only upon FPRs stimulation, but also after activation by IL-8, leukotriene B4 (LTB4), the complement component C5α and FcγR receptor, pointing out the importance of this autocrine purinergic pathway [127].

Neutrophil adhesion to endothelium [128-132], the production of reactive oxygen species (ROS) [50, 133-138] and degranulation are also increased by extracellular ATP [134, 136, 139, 140]. Interestingly extracellular nucleotides regulate neutrophils phagocytosis in a complex manner. It has been previously shown that both ATP and ADP at micromolar concentrations stimulate phagocytosis via activation of Mac-1 [141, 142], but recently Kudo and colleagues have shown that low micromolar concentration of the same nucleotides can inhibit neutrophil phagocytosis until pathogen stimulation [143]. It is possible that ATP and ADP can enable phagocytic cup formation thus inihibiting the binding/uptake of antigens [144]. The inhibition of phagocytic activity by neutrophils should might be important for limiting excessive phagocytosis that occurs in pathological conditions such as hemophagocytic syndrome [145]. However neutrophil bactericidal activity is unaffected by this regulatory mechanism as the inhibition of phagocytosis by ATP and ADP is abrogated by stimulation with fMLP or LPS [143].

Plasmacytoid dendritic cells

The regulation of plasmacytoid dendritic cells (pDCs) function by extracellular nucleotides has been only partially investigated. Plasmacytoid DCs are a subpopulation of dendritic cells playing a crucial role in antiviral immunity. These cells are specialized in the rapid and abundant production type I interferons (IFN-α, -β, -ω) in response to viral infection [146, 147].

The presence of nucleotides such as ATP, ADP, UTP, UDP and UDP-glucose in the extracellular milieu inhibits type I IFN production by pDCs in response to influenza virus or the TLR9 agonist CpG. Nucleotides that exert the most potent inhibitory effect include UDP, UTP and UDP-glucose. This finding suggests the involvement of P2Y4, P2Y6 and P2Y14 receptors [148]. Because type I IFNs enhance cytotoxic activity

and IFN-γ production by NK and CD8+ T lymphocytes, their inhibition may reduce immune surveillance against virally infected and neoplastic cells.

Conclusions

Extracellular nucleotides can modulate the function of cells of the innate immune system as well as of T lymphocytes. The role of extracellular ATP in the regulation of immune responses and inflammation appears to be different in humans as compared to that established in mice. While several observations point out ATP as a signal that induces the innate immune system to trigger and sustain inflammation, other evidences suggest that ATP might represent a negative feedback signal to limit detrimental inflammation in the surrounding of stressed or damaged cells. Several of such regulatory effects of extracellular ATP are mediated by the P2Y11 receptor expressed in humans but not in rodents and linked to increased intracellular cAMP levels that play a major role as immunosuppressive signal.

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Conflicts of interest

The authors declare no conflicts of interest.

Address correspondence to: Dr. Andrea la Sala, Laboratory of Molecular and Cellular Immunology, IRCCS San Raffaele Pisana, Via di Val Cannuta, 247 00166 Rome – Italy. Phone: +39-06-66130427; Fax: +39-06-52255561; E-mail: andrea.lasala@sanraffaele.it

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