



## Myeloid cells in the tumor microenvironment: role of adenosine

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### Abstract

Adenosine, deriving from ATP released by dying cancer cells and then degraded in the tumor environment by CD39/CD73 enzyme axis, is linked to the generation of an immunosuppressed niche favoring the onset of neoplasia. The effects of adenosine are mediated by four adenosine receptors, named A<sub>1</sub>, A<sub>2A</sub>, A<sub>2B</sub> and A<sub>3</sub>, that are widely expressed on several immune cell populations. A critical role of this nucleoside is emerging in the modulation of myeloid cell subsets accumulation and functions into tumor microenvironment, providing new insights that might be useful for the development of novel therapeutic approaches aimed to undermine the immune privileged sites where cancer cells grow and proliferate.

### Keywords

adenosine, adenosine receptors, myeloid cells, immune system, cancer, tumor microenvironment

## Introduction

The tumor microenvironment consists of intricate highly complex and dynamic network of cells, soluble factors, signaling molecules, extracellular matrix, and mechanical cues deputed to promote neoplastic transformation, supporting tumor growth and invasion, shielding the tumor from host immunity, fostering therapeutic resistance, and providing niches for thriving of dormant metastases.<sup>1</sup> In this context, the immune cells infiltrating the tumor milieu have gained attention since groundswell of evidences highlighted that cancer initiation and progression depend essentially on the ability of tumor to escape from host immunosurveillance.<sup>2</sup> Indeed, cancer cells put in place a variety of molecular tricks to fool the immune system, such as the shift of immune responses, with an imbalance in Th<sub>1</sub>/Th<sub>2</sub> responses, and to enhance immunosuppressive cells, such as myeloid-derived suppressor cells (MDSCs), regulatory T cells, M2 macrophages (tumor-associated macrophages), and type 2 natural killer (NK) T cells.<sup>3</sup>

This body of evidence is revitalizing the interest of the scientific community toward cancer immunotherapy as a promising strategy based on the ability to specifically target and destroy tumor cells without harming the surrounding normal cells.<sup>4</sup> In particular, a better understanding of the complex molecular mechanisms driving the interplay between immune system and cancer cells has led to the identification of several key factors governing these interactions, thus prompting the development of 'checkpoint blockades' able to counteract this suppression.<sup>5</sup> Indeed, over the last years we have witnessed the introduction of innovative immunotherapeutic approaches into the clinical practice, such as the blockade of immune checkpoints (i.e. CTL4, PD-1 or PDL-1), designed to overcome the mechanisms exploited by tumors to elude the immune destruction.<sup>5</sup> However, despite immunotherapy is emerging as a viable therapeutic approach to dam the onset and progression of different types of cancer, it has become clear that it faces serious barriers that limit its clinical efficacy.<sup>6</sup>

Several immunosuppressive pathways and factors, released in the tumor microenvironment, can blunt spontaneous or therapeutic immune responses in cancer.<sup>7</sup> In this context, a plethora of research data pointed out the importance of adenosine as a critical regulatory autocrine and paracrine factor that accumulates in the neoplastic microenvironment.<sup>8</sup>

In the cancer milieu, the dying tumor cells release ATP that participates to the recruitment of myeloid and dendritic cells as well as T lymphocytes into the tumor bed<sup>9, 10</sup>. It is well established that some areas of solid tumors often undergo transient or chronic hypoxia, which is conducive to extracellular adenosine accumulation, proposed to play a critical role in protecting cancerous tissues from antitumor immune responses<sup>11</sup>. However, clear –cut experimental evidences about it are needed. Following their release in the tumor microenvironment, ATP and ADP can be converted into AMP by CD39, and then CD73 transforms AMP into adenosine (Figure 1). In several cases, both CD39 and CD73 are expressed on tumor cells and/or tumor-infiltrating immune cells (i.e. Treg).<sup>12</sup>

Once produced in the tumor environment, adenosine participates to the generation of an immunosuppressed niche favoring the onset of neoplasia, via stimulation of four adenosine receptors, named A<sub>1</sub>, A<sub>2A</sub>, A<sub>2B</sub> and A<sub>3</sub> (Figure 1), that are widely expressed on several immune cell populations.<sup>8</sup> Adenosine suppresses T cell-mediated anti-tumor immune responses, mainly via A<sub>2A</sub> receptor activation. The inhibition of adenosine-generating enzyme CD73 or the selective blockade of A<sub>2A</sub> receptors have proved to be therapeutically effective in preclinical settings to directly improve anti-tumor T cells responses.<sup>13, 14</sup> Of note, a critical role of this nucleoside is emerging in the modulation of MDSC accumulation and functions in the tumor microenvironment<sup>15, 16</sup>, thus providing new insights useful for the development of novel therapeutic approaches, that might help to undermine the immune privileged sites where cancer cells grow and proliferate.

This review is intended to discuss the role of adenosine in shaping myeloid cell biology and activity, including tumor-associated macrophages (TAM), dendritic cells (DCs), myeloid-derived

suppressor cells (MDSCs) and mast cells, that are critical orchestrators of immune responses in the tumor microenvironment, pointing out the involvement of this nucleoside in promoting cancer progression.

### *Adenosine and macrophages*

Macrophages are an integral component of innate immunity, playing a critical role in the inflammatory process and host defence.<sup>8</sup> These cells infiltrate tissues during inflammation, and form polarized populations that perform pro- or anti-inflammatory functions. Tissue resident macrophages were previously regarded as differentiated monocytes, which seed the tissues to carry out immune sentinel and homeostatic functions.<sup>17</sup> However, tissue resident macrophages do not comprise a homogenous population, but are indeed clusters of different cells with similar functions and phenotypes.<sup>17</sup> Increasing evidences revealed that macrophages differentiate predominantly in two major phenotypes, referred to as pro-inflammatory M1-type or anti-inflammatory M2-type, depending on tissue microenvironments and/or inflammatory status.<sup>18</sup>

Within the tumor microenvironment, macrophages are a major stromal component, where they are commonly termed tumor-associated macrophages (TAMs), participating both the development of malignant tumors and progression of tumor growth.<sup>19</sup> TAMs exhibit a predominantly M2-like phenotype.<sup>8, 20-22</sup> These cells contribute actively to various processes of cancer growth, such as angiogenesis, cell proliferation and metastasis, exerting a local suppression of antitumor lymphocyte-mediated immunity, and facilitating matrix deposition and remodeling.<sup>8, 20-22</sup>

The adenosine system exerts a plethora of effects on macrophage differentiation, maturation and activation.<sup>23-29</sup> In particular, this nucleoside exerts inhibitory effects, mediated mainly by A<sub>2A</sub> receptors, on M1 activation.<sup>30</sup> Moreover, adenosine, preeminently through A<sub>2B</sub> receptors and, to a lesser extent, via A<sub>2A</sub> receptors, boosts alternative macrophage activation, as shown by the increased expression of several M2 macrophage markers, which include arginase-1, tissue inhibitor of matrix metalloproteinase-1 and macrophage galactose-type C-type lectin-1.<sup>31</sup> In addition,

adenosine can support angiogenesis, stimulating vascular endothelial growth factor (VEGF) production by macrophages through the stimulation of A<sub>2A</sub> receptors (Figure 2).<sup>32-34</sup>

Recently, Bergamin et al.<sup>35</sup> evaluated the activity of a glioma-conditioned medium on macrophage differentiation, investigating the involvement of adenosine in the release of pro- and anti-inflammatory cytokines by these cells. Under these conditions, macrophages were polarized toward a M2-like phenotype and the *in vitro* treatment with exogenous adenosine elicited an increase in the release of the pro-tumoral cytokine monocyte chemoattractant protein (MCP)-1 via A<sub>2A</sub> receptors (Figure 2). In addition, recent studies displayed that A<sub>2A</sub> receptors can enhance IL-1  $\beta$  production<sup>36, 37</sup>. In this regard, it is possible to speculate a role of this receptor subtype in sustain tumor growth since it has been reported a role of this pro-inflammatory cytokine in promoting cancer development inducing several angiogenic factors from tumor and stromal cells<sup>38</sup>.

#### ***Adenosine and dendritic cells***

Dendritic cells (DCs) are the most potent antigen-presenting cells, that drive the activation and differentiation of T cells. Upon exposure to antigens, DCs migrate into proximal lymph nodes. Hence, DCs acquire high expression levels of major histocompatibility complex (MHC) class II and co-stimulatory molecules, such as CD80/86, CD40, and secrete pro-inflammatory cytokines, such as interleukin (IL)-12 and TNF.<sup>39</sup> This process is known as maturation process. Mature and activated DCs polarize T cells toward Th-1 like cells.<sup>39</sup> Notably, the differentiation and maturation process of DCs is context-dependent and it can be affected by the local milieu, generating in turn DC subsets with immune-stimulatory or immune-suppressive properties.<sup>40, 41</sup> Multiple soluble factors may influence the expression of co-stimulatory molecules on DCs and the production of cytokines, that are essential for their function as antigen-presenting cells. In the absence of co-stimulatory molecules, instead of promoting immune activation, expansion and function of T cells, DCs induce tolerance.<sup>42, 43</sup> Tolerogenic DCs produce anti-inflammatory cytokines such as IL-10;

they also express negative costimulatory molecules, including PD-1, B7-H1 and ICOS, that contribute to induce T cell anergy, T cell death or Treg expansion.<sup>43</sup>

Adenosine has been recognized as an important modulator of DC functions. A number of studies indicate that adenosine can significantly affect both the activation/maturation process of DCs as well as cytokine production. The role of adenosine in regulating DCs functions was first described *in vitro* by Panther et al.<sup>44, 45</sup> These authors showed that adenosine enhances the migration of human monocyte-derived immature DCs via A<sub>1</sub> and A<sub>3</sub> activation<sup>44</sup>, and inhibits IL-12 and TNF- $\alpha$  production in mature DCs via A<sub>2A</sub> activation<sup>44, 45</sup>, pointing out a critical role of adenosine in the down-regulation of inflammation and protection from tissue damage.<sup>46</sup> Adenosine can also enhance IL-10 and CCL17 release from mature DCs, while it inhibits CXCL-10<sup>35</sup>, which is implicated in the activation of Th1 immune response.<sup>47</sup> The concomitant presence of IL-10 producing DCs with low levels of IL-12 and CCL17 within the tumor microenvironment is related to the accumulation of Tregs.<sup>43, 48, 49</sup> Therefore, adenosine-conditioned DCs display a reduced capacity of inducing the Th1 polarization of CD4<sup>+</sup>T cells, and they fail to prime also CD8<sup>+</sup>T cells *in vitro* in a cAMP-dependent manner<sup>50</sup>. Similar results were observed from Schnurr et al.<sup>51</sup> in human plasmacytoid dendritic cells (pDCs), a subset of IFN type I-producing dendritic cells, that regulate immune responses during viral infections, autoimmunity and cancer.<sup>52</sup> In particular, this study highlighted a dual role for adenosine on human pDC function: (i) adenosine induces chemotaxis of immature pDCs to inflammatory sites via A<sub>1</sub> receptors, which are the dominant receptor subtypes expressed in these cells; (ii) during inflammation mature pDCs express high levels of A<sub>2A</sub> receptors as in mature monocyte-derived DCs<sup>44, 53</sup>; (iii) A<sub>2A</sub> receptor activation inhibits the pDC-derived cytokines release (IL-12, IL-6).<sup>51</sup>

Subsequent, later extensive studies have been performed on murine bone-marrow derived DC (BMDC) as well as on human monocyte-derived DCs, showing that adenosine-mediated effects on the expression of maturation markers and cytokine release from DCs depend on A<sub>2B</sub> receptor

activation.<sup>54, 55</sup> Notably, these studies clearly indicate that adenosine induces an aberrant differentiation of DC precursor cells into a tolerogenic DC subset that produce VEGF, IL-8, IL-10, IL-6, transforming growth factor (TGF)- $\beta$  indoleamine 2,3-dioxygenase (IDO) and arginase-2.<sup>54, 56</sup> The work by Novitskiy et al.<sup>56</sup> provided the first *in vivo* evidence on the role of A<sub>2B</sub>-stimulated DCs in promoting tumor growth when injected into mice. The pharmacological blockade of A<sub>2B</sub> receptors in tumor-bearing hosts induced a potent anti-tumor T cell-mediated immune response, by enhancing CD11b negative DC activation at the primary tumor site.<sup>34</sup> Conversely, in a mouse model of tumor metastasis, activation of A<sub>2B</sub> receptors does not appear to influence the percentage of myeloid cells, including DCs, nor their maturation/activation status.<sup>57, 58</sup> This may probably reflect a different role of A<sub>2B</sub> receptors in promoting tumor growth or metastasis.

In conclusion, adenosine, through the A<sub>2A</sub>/A<sub>2B</sub> receptor axis can induce a defective functional differentiation of DCs toward a phenotype with pro-angiogenic and tolerogenic features, which allows tumor to escape immune surveillance (Figure 2).

#### ***Adenosine and myeloid-derived suppressor cells***

Myeloid-derived suppressor cells (MDSCs) comprise a heterogeneous population of immature myeloid cells with regulatory behavior. In cancer settings, tumor-associated factors, including inflammatory mediators and growth factors, skew the differentiation of immature myeloid progenitor cells into suppressive cells (MDSCs) rather than into mature dendritic cells, macrophages and granulocytes.<sup>59, 60</sup> Once produced, MDSCs accumulate in the peripheral blood, lymphoid organs and tumor tissue in animal tumor models as well as in cancer patients.<sup>49, 61, 62</sup> In mouse, MDSCs are identified as CD11b positive Gr1 positive cells (CD11b<sup>+</sup>Gr1<sup>+</sup> cells)<sup>53, 54, 55</sup>. In humans the MDSCs are difficult to identify, although they are commonly positive to CD11b and CD33 antigens, and negative to HLA-DR and LIN molecules<sup>53, 54, 55</sup>. MDSCs, together with Tregs, represent key regulatory cells contributing to induce immunosuppression and/or angiogenesis in the tumor environment.<sup>63, 64</sup> MDSCs suppress T-cell responses by producing arginase, reactive nitrogen



and oxide species and suppressive cytokines. Within tumor lesions MDSCs produce also pro-angiogenic factors as well as proteases that endorse angiogenesis and metastases.

Emerging evidence shows that adenosine and its receptor pathways can control some aspects of MDSCs biology in the tumor microenvironment. MDSCs express high levels of CD39/CD73 in tumor lesions of some murine tumor models, including *ret* transgenic mice<sup>15, 65, 66</sup>, thereby contributing to the release of adenosine in the extracellular compartments. The expression of adenosine-generating enzymes and possibly the expression of adenosine receptors in MDSCs within tumor tissue might be regulated by hypoxia and chronic inflammatory factors, like in tumor cells, stromal cells or other immune cell populations of neoplastic lesions. For example, TGF- $\beta$  can induce the expression of CD39 and CD73 on myeloid suppressor cells.<sup>66</sup> Therefore MDSCs produce extracellular adenosine within tumor lesions as an additional mechanism to exacerbate immune-suppression (Figure 2). Notably adenosine can also influence the accumulation of MDSCs within tumors lesions (Figure 2). Experiments performed *in vitro* on bone-marrow hematopoietic cells have indeed shown that A<sub>2B</sub> receptor stimulation prevents the differentiation of hematopoietic progenitor cells into mature myeloid cells, leading to an accumulation of immature cells with immunosuppressive features.<sup>15, 56</sup> A<sub>2B</sub> receptor deficiency in Lewis lung carcinoma-bearing mice correlates with both a low number of tumor-infiltrating MDSCs and reduced levels of intratumoral VEGF.<sup>15</sup> We have recently demonstrated that the blockade of A<sub>2B</sub> receptors with a selective antagonist reduces significantly the number of tumor-infiltrating MDSCs, inhibits tumor angiogenesis and thereby improves T cell-mediated immune surveillance in a melanoma model.<sup>16, 67, 68</sup> However, despite the relevance of these observations in the mouse model, future studies are needed to provide a more detailed understanding of the impact of adenosine and their receptors on MDSCs biology in human cancer environments.

### ***Adenosine and mast cells***

Over the years, an increasing number of studies, aimed at defining the involvement of mast cells in tumor immunity, have revealed a dual role played by these cells in the tumorigenic process, displaying a stimulatory and inhibitory effect on cancer growth based on local stromal conditions.<sup>69</sup>

<sup>70</sup> Indeed, mast cells, once recruited into the cancer milieu via tumor-derived chemoattractants [i.e. MCP-1, RANTES and stem cell factor (SCF)], are able to selectively secrete pro-tumorigenic factors, such as growth factors, histamine, pro-angiogenic factors (i.e. heparin, VEGF and IL-8), as well as proteases that endorse angiogenesis and metastases.<sup>69</sup> By contrast, these cells can counteract the onset of neoplasia, secreting molecules that promote apoptosis (IL-4 and TNF), as well as the pro-inflammatory cytokines IL-1 and IL-6, or tryptase, that is able to stimulate protease-activated receptor-induced inflammation, thus stemming the metastatic processes.<sup>69</sup> Of note, it has been recognized that tumor-infiltrating mast cells participate actively to the remodeling of tumor microenvironment as well as to the promotion of tumor growth.<sup>71-73</sup> Mast cell infiltration and activation in tumors were found to be mediated by the tumor-derived stem cell factor (SCF) and its receptor c-Kit on mast cells. In these settings, tumor-infiltrating mast cells, upon stimulation by high concentrations of SCF, express multiple proinflammatory factors and increase IL-17 expression in cancers.<sup>71</sup> In particular, the presence of mast cells promote the infiltration of tumor microenvironment by MDSCs and their production of IL-17. MDSC-derived IL-17 then indirectly attract Treg cells, increasing their suppressor function and inducing IL-9 production by Treg cells.<sup>72</sup> <sup>73</sup> In turn, the release of IL-9 strengthen the survival and pro-tumorigenic action of mast cells within the tumor microenvironment.<sup>72</sup> In this context, SCF-activated mast cells exacerbated also tumor immunosuppression by releasing adenosine and increasing Treg cells expansion and activity, with a consequent suppression of T cell and NK cells in tumors.<sup>71</sup>

*In vivo* studies have shown an adenosine-induced mast cell activation.<sup>74-78</sup> In particular, set of investigations revealed an involvement of A<sub>1</sub>, A<sub>2B</sub>, and A<sub>3</sub> receptors in the control of these proinflammatory actions on mast cells.<sup>75, 79- 84</sup>

Nowadays, the involvement of adenosine in the regulation of mast cell maturation and differentiation within the neoplastic microenvironment remains scarcely examined, and deserves more focused investigations. However, available data rise the concept that mast cells comprise a highly heterogeneous cell population. Indeed, these cells originate in bone marrow, but mature in peripheral organs where they acquire different phenotypic properties after differentiation in the local milieu, which may differ among tissues. It is possible that the microenvironment conditions where the mast cell reside conditionates the adenosine receptor expression and function, accounting for the differences in the receptor actions observed among the studies. In this regard, it has been observed that the incubation of human mast cells with the pro-inflammatory cytokines IL-4 or IL-13 for 6 h induced the expression of  $A_{2A}$  and  $A_{2B}$  adenosine receptors. By contrast, a prolonged stimulation with IL-4 or IL-13 reduced the  $A_{2A}$ , but not  $A_{2B}$ , receptor expression along with an increase in histamine levels.<sup>85</sup>

Despite the increasing understanding of the contribution of the adenosine system in orchestrating the activity of immune cells in cancer microenvironments, the picture is far from being complete and several outstanding questions remain unanswered. For example, given the critical role of adenosine in conditioning the mast cell activity, what is the differential role of adenosine receptors in regulating the release of pro- and anti-tumorigenic mediators released from mast cells? What is the role of adenosine in regulating the interplay between mast cells tumor stroma?

### **Conclusions and future directions**

Several lines of evidence suggests that adenosine, a soluble factor released in highly expressing-CD73 tumors, induces pro-tumor effects, including immunosuppression and angiogenesis. The adenosine-mediated effects result from the activation of adenosine receptor subtypes ( $A_1$ ,  $A_{2A}$ ,  $A_{2B}$  and  $A_3$ ) on a variety of cell components within tumor microenvironment. In the myeloid compartment adenosine enhances the immunosuppressive activity of TAM and skews the differentiation of DCs into tolerogenic and immunosuppressive DCs. Recent findings support also a

critical role of adenosine in the accumulation of MDSCs within tumor environment. These properties create and sustain a condition of immunosuppression within the tumor microenvironment, with significant suppressive consequences on CD4<sup>+</sup> and CD8<sup>+</sup> T-cell effector functions. It is well appreciated that, in the tumor microenvironment, adenosine inhibits directly T cell-mediated anti-tumor responses and induces Treg mainly via A<sub>2A</sub> activation.<sup>14, 86</sup> Compelling preclinical evidence has shown that targeting the signaling, with selective adenosine receptor antagonists (i.e. A<sub>2A</sub>/A<sub>2B</sub> receptors) or inhibitors of adenosine-generating enzymes (i.e. CD73) might have a therapeutic potential to boost anti-tumor immune responses, thus limiting tumor growth and invasion (reviewed in <sup>87</sup> and in <sup>13</sup>). Accordingly, therapies targeting the adenosine inhibitory pathway could have a great impact on tumor microenvironment. On one hand, the blockade of the A<sub>2</sub> receptor axis can counteract the adenosine-mediated inhibitory effects on cytotoxic lymphocyte responses. On the other hand, it appears that these strategies could ameliorate the myeloid cell responses that initiate, coordinate and support anti-tumor immune responses. In this regards, studies using a tumor model in mice with a myeloid-selective deletion of *Adora2a* have shown that the expression of IL-12 and MHC class II on TAM is increased, while IL-10 production by TAM, DCs and MDSCs is reduced.<sup>88</sup> This effect was associated with improved NK and CD8<sup>+</sup>T cell activities.<sup>88</sup>

Although currently available knowledge indicate that adenosine plays a critical role in regulating several aspects of myeloid cells within tumor microenvironment, it would be of interest to investigate thoroughly the mechanisms by which the pathway of adenosine and its receptors could regulate immune cell functions at the tumor site. For example, the effects of adenosine on mast cells in the tumor microenvironment are not well described and deserve more focused studies. An important issue to address is how adenosine cooperates with other soluble factors to affect different cells within the tumor microenvironment. In addition, it is crucial to evaluate the role of adenosine in regulating the interplay between stroma cells and tumor-infiltrating immune cells.

Since the effects of adenosine appear to be dependent on the cell types well as on the adenosine receptor subtypes distribution and expression, differences in adenosine signaling among the species (i.e. rodent versus human species) can occur. Therefore, understanding the relative role of adenosine pathway in the context of cancer patients is of critical importance. Future research will thus be essential to address the therapeutic potential of the pharmacological tools targeting adenosine system for clinical development, both alone and in combination with immune checkpoint inhibitors.

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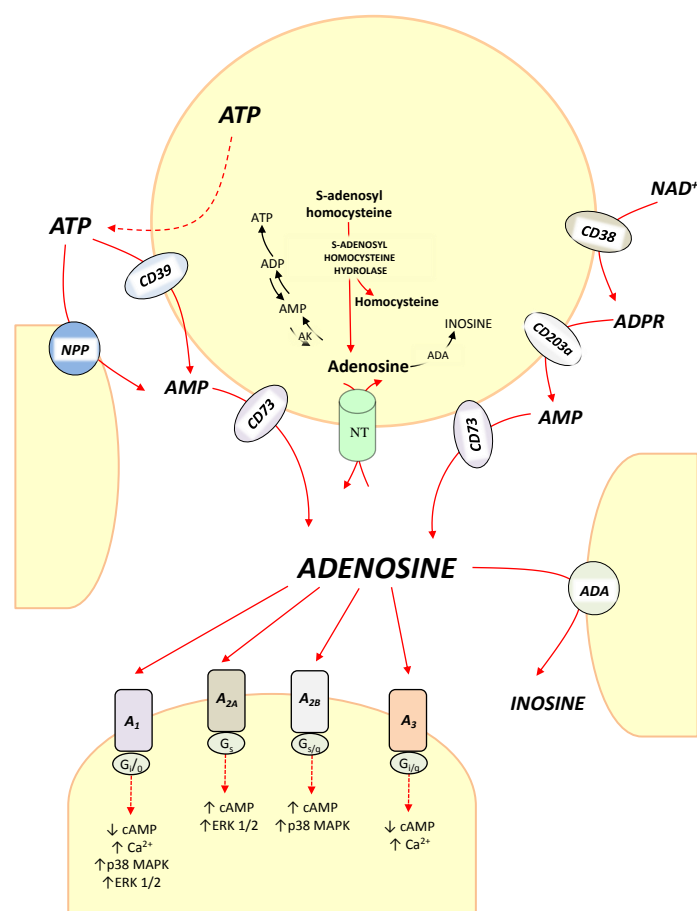


Figure 1

**Figure 1. Schematic diagram illustrating adenosinergic pathways.** ATP is released into the extracellular environment, which is followed by its sequential degradation to AMP by the cell surface enzyme CD39 or alternatively via Nucleotide Pyrophosphatase/Phosphodiesterase (NPP), and to adenosine by CD73. Adenosine can also be generated intracellularly from AMP or S-adenosylhomocysteine, and then released into the extracellular space via nucleoside transporters (NTs). In addition, another source of adenosine is the extracellular NAD<sup>+</sup> that once converted by the CD38 ectoenzyme into ADP-ribose (ADPR), can be further split by CD203a to form AMP and subsequently converted into adenosine by CD73. Extracellular adenosine can bind to 4 different G-protein-coupled receptors that either stimulate (A<sub>2A</sub> and A<sub>2B</sub>) or inhibit (A<sub>1</sub> and A<sub>3</sub>) adenylyl cyclase activity and cAMP production in the cell. Furthermore, all adenosine receptors couple to mitogen-activated protein kinase (MAPK) pathways, including extracellular



signal-regulated kinase 1/2 and p38 MAPK. In the extracellular space, adenosine concentrations are controlled by the adenosine deaminase (ADA), which catalyzes its conversion into inosine as well as by the activity of nucleoside transporters (NT). Nucleoside transporters, also mediate uptake of extracellular adenosine. After intracellular uptake, adenosine undergoes rapid phosphorylation to AMP by adenosine kinase, or deamination to inosine by adenosine deaminase (ADA). ADA: adenosine deaminase; AK: adenosine kinase; AMP: adenosine monophosphate; ATP: adenosine triphosphate; NT: nucleoside transporter.

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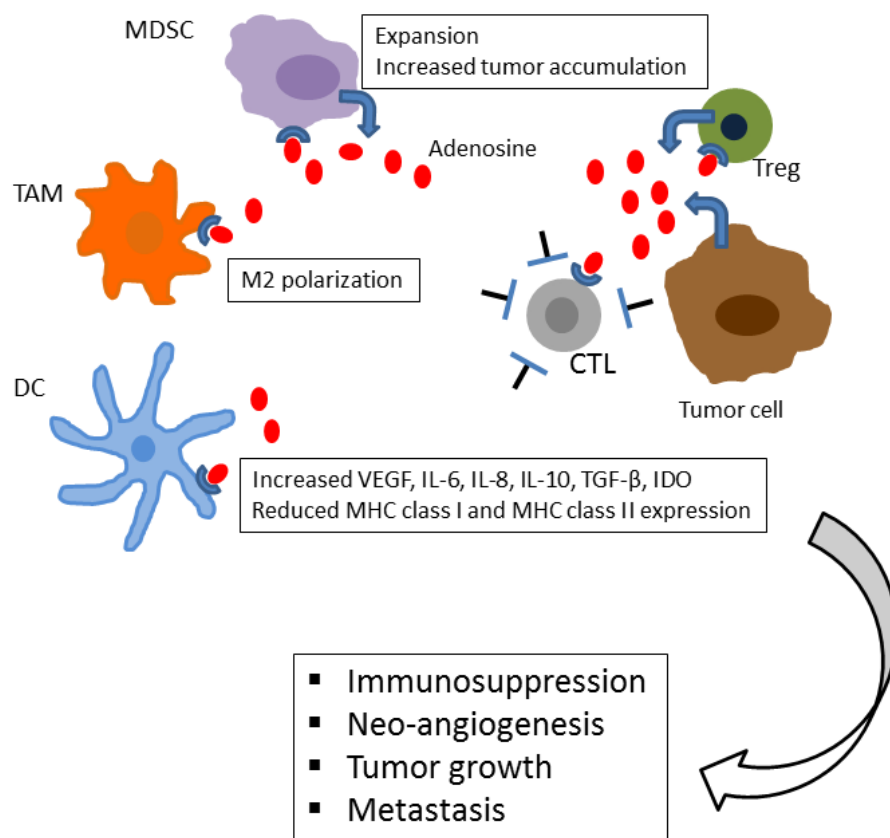


Figure 2

**Figure 2. Tumor-promoting effects of adenosine.** Adenosine is either produced by tumor cells and/or by tumor-infiltrating cells, including MDSC and Treg. Adenosine promotes polarization of myeloid cells toward immunosuppressive and pro-angiogenic phenotypes, that prevent T cell activation and function. In addition, adenosine inhibits anti-tumor immune response by acting directly on effector T cells. These effects contribute to establish a tolerogenic environment, that facilitates tumor growth and invasion. DC: dendritic cell; TAM: tumor-associated macrophage; MDSC: myeloid-derived suppressor cell; Treg: regulatory T cell; CTL: cytotoxic T lymphocyte; VEGF: vascular endothelial growth factor; IL: interleukin; TGF- $\beta$ : transforming growth factor- $\beta$ ; IDO: indolamine 2, 3-dioxygenase; MHC: major histocompatibility complex.