Chapter 1

Adenosine receptors and its ligands

1.1 Introduction to Adenosine Receptors

Purines are the most widespread chemical messengers in animal and plant kingdoms. Adenosine, a purine nucleoside is an endogenous ligand composed of adenine attached to ribose. Adenosine plays an important role in biochemical processes, such as energy transfer—as adenosine triphosphate (ATP) and adenosine diphosphate (ADP)—as well as in signal transduction as cyclic adenosine monophosphate (cAMP). It is identified as a major local regulator of tissue function especially when energy supply fails to meet cellular energy demand. Due to its ability to equalize energy intake to metabolic demand, it earned the reputation of a “retaliatory metabolite” (Sattin & Rall 1970).

Adenosine is a signaling molecule whose physiological functions are mediated by its interaction with four G-protein-coupled receptor (GPCR) subtypes, A₁, A₂A, A₂B and A₃ respectively. Extracellular adenosine acts as a local modulator with a generally cytoprotective function in the body. Its effects on tissue protection and repair fall into four categories: increasing the ratio of oxygen supply to demand; protecting against ischaemic damage by cell conditioning; triggering anti-inflammatory responses; and the promotion of angiogenesis.

The adenosine receptors (ARs) differ in their affinity for adenosine. The A₁ and A₃ adenosine receptor subtypes have high and low affinity for adenosine, respectively. They couple to Gi protein to inhibit adenylate cyclase and thus lead to decrease the cyclic AMP (cAMP). By contrast, high affinity A₂A and low affinity A₂B adenosine receptor subtypes couples to Gs and stimulates adenylyl cyclase leading to an increase of cyclic AMP (cAMP) levels (Ham & Evans 2012) (Figure-1.1). These two subtype pairs also share higher sequence identity: the human A₁ and A₃ ARs are 49% identical, and the human A₂A and A₂B ARs are 59% identical.
Figure 1.1 Adenosine receptor signaling

The four adenosine receptors have been cloned from several mammalian species, including human. There is extensive sequence similarity between species for the A_1, A_{2A} and A_{2B} receptors, whereas A_3 receptors are more variable (Londos et al. 1980). Each adenosine receptor has different but overlapping functions. Each of them is unique in pharmacological profile, tissue distribution and binding partners. The greatest challenge in developing adenosine receptor ligands for specific clinical applications is that adenosine signaling is so widespread. Adenosine itself is present ubiquitously, adenosine receptors are widely distributed throughout the body and adenosine acting at these receptors exerts a broad spectrum of physiological and pathophysiological functions (Chen et al. 2013). By designing synthesizing and screening the molecules against adenosine receptors there is hope to be able to target a disease specifically by a selective compound.

1.2 Adenosine receptors as therapeutic target

As there is great and extensive roles of adenosine receptor subtypes in both physiologic and pathophysiologic events, these receptors are becoming important drug targets in the treatment of a variety of diseases. With more and more research going on in the field of adenosine receptors it has been possible to find out a number of physiological and pathophysiological processes where one or more adenosine receptors are involved. The
list of such processes is quite wide, and since it is increasing each year it is likely that it will further lengthen.

ARs have been targets for drug development which is schematically shown in Figure 1.2 (Fredholm 2010). By far the most serious attempts have been made in the development of $A_{2A}$ antagonists for neurodegeneration where several drugs companies have candidate drugs in late phases of clinical trial and one of them has been approved (Dungo & Deeks 2013). Adenosine receptors are found on almost all the cells and so the agonists are likely to produce unwanted side effects. On contrast selective antagonists will only affect those sites where receptors are active. The fact that a majority of humans already consume an adenosine antagonist, caffeine, on a daily basis of course also makes one wonder how much can be derived by additional blockade (Fredholm 2010).

Figure 1.2 Some of the potential uses of drugs that act as agonists (left) and antagonists (right) at the four different adenosine receptors are indicated (Fredholm 2010)
1.2.1 The $A_1$ adenosine receptor

The adenosine $A_1$AR is widely distributed in varying levels of expression about many different tissues in the human body, ranging from the colon to the brain. The highest levels found in the brain, especially at excitatory nerve endings (Daly & Padgett 1992). Activation of the $A_1$AR inhibits adenylyl cyclase activity, that activates potassium channels (including KATP channels in neurons and the myocardium), blocks transient calcium channels and increases intracellular calcium and inositol-1,4,5-trisphosphate[Ins(1,4,5)P3] levels by activating phospholipase C (PLC). $A_1$AR modulates neuronal activity by blocking neurotransmitter release and reducing the firing rate. The $A_1$AR mediates negative chronotropic and inotropic effects in the heart but they also exert effects in many other organs and cells. The physiology and pathophysiological effect are implicated in decreased renal blood flow, tubuloglomerular feedback, inhibition of renin release, inhibition of lipolysis, vasoconstriction, bronchoconstriction, inhibition of neurotransmitter release, inhibition of insulin and glucagon release, reduced heart rate, osteoclast activation and bone resorption, reduced respiration, sleep, analgesia, cardiac preconditioning (Fredholm 2010). Considerable advances have been made recently in the pharmacological and molecular characterization of $A_1$AR, which had been proposed as targets for drug design and discovery. Xanthine and xanthine derivatives, including the natural compounds theophylline and caffeine, constitute the prototypical group of antagonists at all ARs, and modifications of the xanthine structure have given various derivatives which have shown good subtype selectivity. Most of the selective $A_1$AR antagonists are xanthine-based derivatives (Baraldi, Tabrizi, Gessi, et al. 2008).

1.2.2 The $A_{2A}$ adenosine receptor

The $A_{2A}$AR has been most widely studied receptor among the all the adenosine receptors structurally as well as therapeutically. $A_{2A}$AR are highly expressed in the spleen, thymus, leukocytes, blood platelets, striatopallidal GABAergic neurons and the olfactory bulb and expressed to a lesser extent in the heart, lung, blood vessels, and other brain regions. The $A_{2A}$AR is important in mediating vasodilation, supporting the synthesis of new blood
vessels and protecting tissues from collateral inflammatory damage. In the brain, A2A AR influences the activity of the indirect pathway of the basal ganglia. Various investigations are being conducted on a number of compounds to treat inflammation, cancer, ischemia reperfusion injury. A2AAR is highly expressed in the spleen, thymus, infectious diseases, wakefulness, neurodegeneration (including Parkinson's disease and Alzheimer's disease) and other CNS disorders (de Lera Ruiz et al. 2013). To understand the insights of receptor and its ligand binding interaction, the A2AAR has been resolved by X-ray crystallographic techniques (Jaakola et al. 2008; Lebon et al. 2011; Doré et al. 2011; Xu et al. 2011; Congreve et al. 2012). Normally, adenosine receptors have a common central core consisting of seven transmembrane helices (TM1−7), each TM being mainly α-helical and composed of 20–27 amino acids. Each TM domain is linked by three intracellular (IL1, IL2, and IL3) and three extracellular (EL1, EL2, and EL3) loops. ARs differ in the length and function of their N-terminal extracellular domain, their C-terminal intracellular
domain, and their intracellular/extracellular loops. Each of these areas provides very specific properties that are critical for achieving ligand selectivity among the different receptor subtypes. Figure 1.3 shows X-ray crystallographic structure of A<sub>2A</sub>AR bound to ZM241385 (Jaakola et al. 2008). Various agonist and antagonists synthesized are based on adenosine derivatives, xanthine derivatives, tricyclic molecules and many other derivatives and they have proved to be highly active and selective to A<sub>2A</sub>AR proving to be important in many diseases specifically neurological disorders (Shook & Jackson 2011).

1.2.3 The A<sub>2B</sub> adenosine receptor

The A<sub>2B</sub> AR is highly expressed in the gastrointestinal tract, bladder, lung, mast cells, eye, adipose tissue, brain, kidney, liver and other tissues. The A<sub>2B</sub>AR is structurally closely related to the A<sub>2A</sub>AR and able to activate adenylate cyclase but it is functionally very different from A<sub>2A</sub>AR. It has been postulated that this subtype may utilize signal transduction systems other than adenylate cyclase because of these functional differences (Livingston et al. 2004). Among all the adenosine receptors, the A<sub>2B</sub>AR is a low affinity receptor that is thought to remain silent under physiological conditions and to be activated in consequence of increased extracellular adenosine levels. As such there is no crystal structure of A<sub>2B</sub>AR available; but the available structures of A<sub>2A</sub>AR are suitable as templates for the study of A<sub>2B</sub> AR models. Despite the similarity between the A<sub>2A</sub> and A<sub>2B</sub> ARs, there are many differences between the two receptor subtypes, e.g., the longer extracellular loop 2 of the A<sub>2B</sub>AR or the fact that the A<sub>2A</sub>AR possesses four disulfide bonds in the extracellularly oriented part of the protein whereas the A<sub>2B</sub>AR has been found to have only one disulfide bond. The adenosine A<sub>2B</sub>AR’s physiological role is very less understood, still it has been implicated to play key roles in various processes like modulation of arterial blood pressure and heart rate, glucose metabolism, angiogenesis induction, growth of some tumors, intestinal inflammation, myocardial ischemia, acute lung and kidney injury, inflammatory response and much involvement in the pathogenesis of asthma and chronic obstructive pulmonary disease (COPD) (Thimm et al. 2013). Several class of ligands found have shown good affinity towards A<sub>2B</sub>AR are adenosine
derivatives, xanthine derivative, pyrrolopyrimidines, pyrazolotriazolopyrimidines and 2-aminopyrazines (Taliani et al. 2013).

1.2.4 The A₃ adenosine receptor

The A₃AR is the last member of the adenosine receptors family to have been cloned (Jacobson & Gao 2006). Considering receptor distribution, the highest levels of human A₃AR mRNA have been found in lung and liver. However, A₃ARs have been detected in various tissues including testis, lung, kidney, placenta, heart, brain, spleen, liver, uterus, bladder, jejunum, aorta, proximal colon and eyes (Borea et al. 2009). A₃ARs, via the interaction with Gi proteins, inhibit adenylate cyclase, decreasing cyclic AMP accumulation and protein kinase A (PKA) activity. In addition, A₃ARs, by coupling with Gq proteins, stimulate phospholipase C (PLC), causing an increase of calcium levels from intracellular stores, and modulate the protein kinase C (PKC) activity. In case of adenosine A₃AR also there is no crystal structure available, but for molecular modeling studies, the homology modeled structure of adenosine A₃AR is used. The functional role of A₃AR is implicated in many disorders like joint disorder, eye disorders, cancer, respiratory disorders and cardiovascular disorders. Majority of the A₃AR receptor agonists are nucleoside derivatives. Apart from nucleoside derivatives some of xanthine and pyridine derivatives are also reported to play agonist’s role. Derivatives of pyrimidines, thiazoles, thiadiazoles, quinazolinones, quinoxalines, adenines, triazoloquinzoline, pyrazoloquinolines, pyridtriazolopyrazines have been reported to possess antagonist activity against A₃AR.

1.3 Adenosine Receptors agonists

Adenosine is an endogenous natural agonist which binds to all the ARs, so modification of adenosine is main approach for discovering AR agonists. Apart from adenosine xanthosine derivatives have been also good agonists. The structure-activity relationships of adenosine (1) at ARs have been extensively probed (Yan et al. 2003). Most of the useful analogues are modified in the N⁶-or 2-position of the adenine moiety and in the 3’, 4’- or 5’-position of the ribose moiety. Highly selective agonists of the various receptor
subtypes have been designed through both empirical approaches and a semi-rational approach based on molecular modeling (Kim et al. 2003; Tchilibon et al. 2005).

### 1.3.1 A₁ adenosine receptor agonists

To get A₁ adenosine receptor agonists, generally substitution of adenosine (1) at the $N^6$-position with a wide range of alkyl, cycloalkyl, and arylalkyl groups increases selectivity. The most successful selective agonists of the A₁AR were achieved by substituting adenosine at $N^6$ position with cycloalkyl group. $N^6$-Cyclopentyladenosine (2, CPA) and its 2-chloro analogue (3, CCPA) are among the most potent and selective A₁AR agonists and they are in wide use as pharmacological agents. $S$-(-)-ENBA (4) is an even more potent and selective agonist for both human and rat A₁ARs compared with the three other AR subtypes (Gao et al. 2003). Bayer Co. (Germany) discovered 2-amino-3, 5-dicyanopyridine derivatives e.g. capadenoson (5), as non-nucleoside-derived adenosine receptor agonists (Rosentreter et al. 2009). Other than these derivatives, several selective adenosine derivatives, including GW493838 (6) and Tecadenoson (7) have been evaluated in clinical trials for various indications (Müller & Jacobson 2011) (Figure 1.4).

![Figure 1.4 A₁ adenosine receptor agonists](image-url)
1.3.2 A\textsubscript{2A} adenosine receptor agonists

Substitution of adenosine at the 2-position, especially with (thio) ethers, secondary amines, and alkynes, has resulted in many synthetic analogues selective for the A\textsubscript{2A}AR. NECA (8) is a potent nonselective agonist has a 5’-N-alkyluronamide modification, and a 5’-N-ethyluronamide modification has maintained or enhanced the selectivity for the A\textsubscript{2A}AR which is present in case of CGS21680 (12). 2-(2-phenylethyl) amino is also important modification in case of CGS21680 for enhancing the affinity at A\textsubscript{2A}AR. UK-432097 (11) is \(N^6\)-(2, 2- diphenylethyl) adenosine analogues, so some \(N^6\)-position substitutions have also been found to increase the affinity at the A\textsubscript{2A}AR. Regadenoson (9, Lexiscan\textsuperscript{TM}) and Apadenoson (10) has been developed for vasodialation (Müller & Jacobson 2011) (Figure 1.5).

![Chemical structures of NECA, Regadenoson, Apadenoson, UK432097, and CGS21680](attachment:chemical_structures.png)

Figure 1.5 A\textsubscript{2A} adenosine receptor agonists

1.3.3 A\textsubscript{2B} adenosine receptor agonists

The adenosine A\textsubscript{2B}AR is least characterized subtype in the AR family. Combinations of changes at adenosine have resulted in compounds that activated the A\textsubscript{2B}AR with good selectivity. \(N^6\)-substituted adenosines, \(N^6\)-substituted-5’-N-alkyl-carboxamido adenosines, \(C^2\)-substituted adenosines and \(C^2\)-substituted-5’-N-alkyl-carboxamido adenosines have
been reported to show good affinity and selectivity towards A$_2$BAR (13, 14). Apart from adenosine derivatives, BAY 60-6583 (15) is one of the 2-aminopyridine-3, 5-dicarbonitriledervatives which is found to activate the A$_2$BAR (Baraldi, Tabrizi, Fruttarolo, et al. 2008; Müller & Jacobson 2011) (Figure 1.6).

**1.3.4 A$_3$ adenosine receptor agonists**

The vast majority of A$_3$AR agonists reported to date reflects the nucleoside structure of the endogenous ligand, adenosine. The most successful structural modification of the adenosine skeleton in enhancing A$_3$AR potency and selectivity involve N$_6$-, C$_2$-, and 5′-substitutions or combination of these. Substitution with an N$_6$-benzyl group or substituted benzyl group increases selectivity for the A$_3$AR. IB-MECA (CF101, 16) and the more selective agonist CI-IB-MECA(CF102, 17) have been widely used as pharmacological probes. The 4′-thioadenosine derivative LJ-529 (18) also acts as a highly potent and selective A$_3$AR agonist with a subnanomolar affinity.

The new design which includes [3.1.0]bicyclohexane ring system in place of the ribose 5-membered ring was utilized to get more potent and selective analogues such as
MRS3558 (19), which displays nanomolar affinity at the A₃AR (Müller & Jacobson 2011; Baraldi et al. 2012) (Figure 1.7).

1.4 Adenosine receptors antagonists

Traditionally, the adenosine receptor antagonists have been xanthine derivatives. The natural products like caffeine and theophylline behaves as weak and nonselective AR antagonists. The structure activity relationship (SAR) of xanthine derivatives as AR antagonists has been the core research area to get selective AR antagonists. The effects of receptor subtype selectivity of substitution at the 1-, 3-, 7-, and 8-positions have been explored in detail. However, many new nonxanthine and non-purine derivatives have been developed as highly selective AR antagonists.

1.4.1 A₁ adenosine receptor antagonists

Potent and selective antagonists for A₁AR have been developed by modification of the xanthines at 8-position with aryl or cycloalkyl groups and these modification has led to high affinity and selectivity (Moro et al. 2006). For example, the 8-cyclopentyl derivative DPCPX or CPX (8-cyclopentyl-1, 3-dipropylxanthine, 20) is highly selective at the human A₁AR.

Figure 1.8 A₁ adenosine receptor antagonists
Rolofylline (KW-3902, 21) has a bicycloalkyl group present at 8\textsuperscript{th} position of xanthine ring and has a good selectivity. Bamifylline (22) is another selective derivative with benzyl group at 8\textsuperscript{th} position on xanthine ring. Apart from the xanthine derivatives, non-xanthine structures are also reported to show high affinity and selectivity towards A\textsubscript{1}AR (Kiesman et al. 2009). FK-453 (23) is a pyrazole derivative and SLV-320 (24) is a pyrrolopyrimidine derivative, both are highly selective. New 2-aminothiazole derivatives which are PERD-MCD (25) derivatives have shown high A\textsubscript{1}AR affinity and selectivity (Scheiff et al. 2010) (Figure 1.8).

### 1.4.2 A\textsubscript{2A} adenosine receptor antagonists

Historically, A\textsubscript{2A} AR antagonists have been divided into xanthine-based and non-xanthine-based derivatives. Modification of xanithines at the 8-position with alkenes has led to selectivity for the A\textsubscript{2A}AR. Istradefylline (KW-6002, 26) is a xanthine based, selective A\textsubscript{2A}AR antagonist that was approved recently for Parkinson’s Disease (Dungo & Deeks 2013). MSX-2 (27) is selective A\textsubscript{2A}AR inhibitor which was further converted into two different prodrugs phosphate prodrug (MSX-3, 28) and L-valine ester prodrug (MSX-4, 29) to make them water soluble (Sauer et al. 2000; Vollmann et al. 2008).

![Figure 1.9 A\textsubscript{2A} adenosine receptor antagonists](image)
Both are now broadly used as pharmacological tools in particular for in vivo studies. Xanthine derivatives however, have several limitations as pharmacologic tools because of poor pharmacokinetic profile. So number of monocyclic, fused bicyclic and tricyclic derivatives other than xanthine derivatives has been developed as adenosine A\(_2\)AR antagonists (de Lera Ruiz et al. 2013). ZM-241385 (30) is highly selective triazolopyrimidine derivative which may become useful for neurodegenerative diseases. Triazolopyrazolopyrimidine derivatives like Preladenant (31) and benzothiazole derivatives like Tozadenant (33) have reached late stage clinical trials for Parkinson’s Disease (Figure 1.9).

### 1.4.3 A\(_2\)B adenosine receptor antagonists

One of the first compounds as A\(_2\)BAR antagonist was the xanthine derivative MRS1754 (34) which is potent and selective in humans. Furthermore potent and A\(_2\)BAR selective xanthine derivatives include MRE-2029-F20 (35) and PSB-1115 (36) which have been used as radioligands. Besides xanthines derivatives, nonxanthine A\(_2\)BAR antagonists have recently been developed is LAS38096 (37), a pyrimidine derivative. Other non-xanthine derivative includes thiazole in QAF 805 (38) which is Novartis compound. All of these A\(_2\)BAR antagonists (Figure 1.10) have been in the development for various inflammatory diseases and asthma. (Baraldi, Tabrizi, Fruttarolo, et al. 2008)
1.4.4 A<sub>3</sub> adenosine receptor antagonists

Unlike other three adenosine receptor subtypes (A<sub>1</sub>, A<sub>2A</sub> and A<sub>2B</sub>), the A<sub>3</sub>AR has low binding affinity towards xanthine derivatives. Still, some of the cyclized xanthine derivatives like KF-26777 (39) showed good affinity and selectivity towards A<sub>3</sub>AR. Because of low affinity towards xanthine derivatives, various classes of non-xanthine monocyclic, bicyclic and tricyclic heterocyclic derivatives were developed as A<sub>3</sub>AR antagonists. Pyridine derivative like MRS1523 (40) and 2-amino thiazole derivative like CGH2466 (41) had shown nanomolar affinity towards A<sub>3</sub>AR. VUF5574 (42) is a quinazoline derivative and LJ1251 (43) is thioadenosine derivatives which had shown antagonist activity towards A<sub>3</sub>AR. Triazoloimidazolepyrimidine derivative OT-7999 (44) is developed for the treatment of glaucoma. The A<sub>3</sub>AR antagonists are under consideration for treatment of cancer, stroke, asthma, COPD and inflammation (Baraldi et al. 2012) (Figure 1.11).

![Chemical structures of A<sub>3</sub> adenosine receptor antagonists](image)

**Figure 1.11** A<sub>3</sub> adenosine receptor antagonists

1.5 Allosteric modulation of adenosine receptors

Allosteric modulators of adenosine receptors are the alternative to direct acting AR agonist and antagonists. Allosteric modulators bind at a distinct site other than the natural
ligand binding site. They exert their effect only in the presence of the orthosteric ligand. A positive allosteric modulator (PAM) induces an enhancement of effects of the orthosteric ligand, while a negative allosteric modulator (NAM) attenuates those effects (Göblyös & Ijzerman 2009).

**A<sub>1</sub> AR**: Bruns and colleagues introduced the first allosteric modulators of A<sub>1</sub>AR in 1990s (Bruns & Fergus 1990; Bruns et al. 1990). They described various 2-amino thiophene derivatives such as PD 71605 (45), PD 81723 (46) and PD 117975 (47) as allosteric modulators of A<sub>1</sub>AR. Other thiophene derivative T62 (48) is clinically evaluated for treatment of neuropathic pain. Allosteric enhancers at the A<sub>1</sub>AR have received attention as anti-arrhythmic cardiac agents, and, more recently, as anti-lipolytic agents. In addition, this class of compounds has therapeutic potential as analgesics and neuroprotective agents (Figure 1.12).

**A<sub>2A</sub> AR**: Allosteric modulation for the A<sub>2A</sub>AR has not been much developed; still amiloride (49) and analogues developed by Gao and Ijerman were demonstrated to be allosteric inhibitors for the A<sub>2A</sub>AR. However, these compounds are not selective for this subtype and they also allosterically modulate action at both A<sub>1</sub> and A<sub>3</sub> ARs (Göblyös & Ijzerman 2009).

**A<sub>2B</sub> AR**: Allosteric modulators for A<sub>2B</sub>AR have also been not much reported. Despite that, a recent report has shown the allosteric modulation of adenosine A<sub>2B</sub>AR by indole derivatives (Trincavelli et al. 2014).
**A3AR**: Allosteric modulation of the A3AR was first observed by Gao et al with VUF5455 (50) and other 2-pyridinyl isoquinoline derivatives which were previously reported as A3AR antagonists. Imidazoquinolinamines are another structural class of A3AR modulators, which were originally A1AR antagonists. Imidazoquinolinamine derivative DU124183 (51) is an allosteric enhancer of radioligand binding at the A3AR (Baraldi et al. 2012).

### 1.6 Selective disease targets for Adenosine receptors

The various disorders targeted by drugs that are in pre or advanced clinical trials modulating adenosine receptors include CNS-disorders, cardiovascular disorders, antinflammatory, autoimmune disorders and cancer.

**CNS disorders**: Several pharmacological studies suggest that the A2AAR is involved in motor activity. In particular, adenosine A2AAR antagonists have been demonstrated to restore the deficits caused by degeneration of the striatonigral dopamine system, and therefore offer a possible treatment for Parkinson’s Disease.

The current treatment for Parkinson’s Disease is primarily based on dopamine replacement therapy. Levodopa (L-DOPA), a metabolic precursor of dopamine, has been used for the treatment of Parkinson’s Disease for decades. The A2AAR is present in good concentration in striatum, which interacts with the D2 receptor. Preclinical studies of A2AAR antagonist is demonstrating motor benefit in rodent and non-human primate models of Parkinson’s disease (Richardson et al. 1997; Schwarzschild et al. 2006; Shook & Jackson 2011). So A2AAR antagonists have emerged as leading non-dopaminergic drugs for the treatment of Parkinson’s Disease. Over the past 8 years, a total of 25 clinical trials have been conducted. Six double-blind, placebo-controlled clinical Phase IIb and Phase III trials of Istradefylline (KW-6002, 26), involving a total of >2,000 patients with advanced Parkinson’s Disease, and one Phase IIb trial with Preladenant (SCH420814, 31), involving 253 patients with advanced Parkinson’s Disease, have been reported (Hauser et al. 2011). These clinical Phase IIb and Phase III trials have shown a modest but significant reduction in the average ‘off-time’ by about 1.7 hours compared to the optimal
L-DOPA (levodopa) dose regimen. However, Preladenant did not prove to be effective in phase-III clinical trials so it was discontinued in May 2013, whereas Kyowa Hakko Kirin got the first global approval of drug Istradefylline (26) for Parkinson’s Disease.

Adenosine as a contributing element in the pathophysiology of schizophrenia embraces several neurotransmitter systems and brain regions due to its multiple and widespread modulatory actions (Lara et al. 2006).

Cardiovascular disorders: The A1AR is potential therapeutic target for a number of disorders including atrioventricular (AV) node block and supraventricular tachyarrhythmia (AR agonist); AV block of cardiac arrest (AR antagonist); bradyarrhythmias in transplanted hearts (AR antagonists); diuresis (AR antagonists). In patients with documented paroxysmal supraventricular tachycardias involving the AV node, 99% are successfully terminated with standard doses of adenosine (Strickberger et al. 1997).

Ellenbogen et al. in 2005 found that Tecadenoson (7) is a potent selective A1AR agonist with a dose-dependent negative dromotropic effect on the AV node. They evaluated tecadenoson, a selective A1AR agonist, for the acute termination of paroxysmal supraventricular tachycardia (PSVT). In the atrial-paced guinea pig heart model, tecadenoson caused an A1AR receptor mediated negative dromotropic effect on the AV node and lengthening of the AV nodal refractory period, leading to termination of reentrant PSVT at doses that did not affect blood pressure (BP), sinus cycle length, or the His-ventricular interval. Side effects mediated by the A2A, A2B and A3 ARs such as flushing, chest pressure, hypotension, and bronchospasm were infrequent, consistent with the A1AR selectivity of the drug (Ellenbogen et al. 2005).

Impaired renal function is common in patients with acute heart failure; it directly contributes to deterioration of the heart and is associated with an adverse outcome, including increased mortality. Local adenosine production in the kidney is increased in patients with heart failure as a result of hypoxia caused by reduced renal perfusion and by stimulation with diuretics. Based on our understanding of the mechanisms associated with
renal dysfunction and the demonstrated control of renal function via A₁AR, A₁AR antagonists were developed. These antagonists reduced the risk of persistent worsening renal failure by >50% in a Phase IIb study involving 301 patients with acute heart failure, and improved renal plasma flow in 63 ambulatory patients with chronic heart failure. Based on these promising results, a placebo-controlled, randomized Phase III trial involving 2,033 patients with acute heart failure (the PROTECT study) was carried out with the A₁AR antagonist rolodelfine; this was the largest study to date involving the use of A₁AR antagonists to target renal function (Cotter et al. 2008). Unfortunately, the results were disappointing and rolodelfine did not prevent persistent worsening renal function. The reason for this absence of renoprotective effects is likely to be due to an enhanced diuretic effect in the rolodelfine group, which may have offset the effects of rolodelfine on the preservation of renal function. Moreover, pharmacological and genetic studies have clearly demonstrated that A₁AR mediate protective effects against ischaemic kidney injury and brain injury, which is consistent with the increased frequency of stroke and seizure activity in clinical trials of A₁AR antagonists. Thus, the development of A₁AR antagonists for the treatment of disorders associated with impaired fluid retention, such as congestive heart failure, should proceed with caution.

The A₂AAR is involved in vasodilation in the aorta and coronary artery. It was suggested that the tachycardic effect of A₂AAR activation is mediated by centrally located receptors, whereas its hypotensive effect is mediated by the peripheral A₂AAR (Schindler et al. 2005). In the late 1960s and 1970s, metabolically stable AR agonists were tested clinically as antihypertensives, and this was an intended use of the A₂AAR agonist CGS21680 (12); however its clinical path was stopped due to non selectivity towards adenosine receptors. In platelets, an A₂AAR agonist was shown to inhibit aggregation by increasing intracellular cAMP levels, suggesting that adenosine agonists might have utility as antithrombotic agents. Then after various efforts has been carried out to further improve subtype-selectivity of A₂AAR agonists for novel therapeutic applications, including imaging. Adenosine (1), under the name Adenoscan (Astellas Pharma), is used in myocardial stress imaging to evaluate coronary artery disease by achieving vasodilation in patients unable to exercise adequately. Regadenoson (CVT-3146, 9), a
potent and selective $A_{2a}$AR agonist, is approved drug for myocardial perfusion imaging (Hendel et al. 2005).

**Inflammatory diseases, autoimmune disorders and cancer:** The adenosine receptor subtypes are highly expressed in all the cells of immune system, the adenosine $A_1$, $A_{2a}$, $A_{2b}$ and $A_3$ receptors are being actively engaged as therapeutic targets for autoimmune diseases, chronic inflammatory disorders and cancer (Voors et al. 2011).

The potent $A_3$AR antagonists have been developed for therapeutic treatment of inflammatory diseases such as asthma and glaucoma. Activation of $A_3$AR has been shown to stimulate phospholipase C and to inhibit adenylate cyclase. $A_3$AR agonists also cause stimulation of phospholipase D and the release of inflammatory mediators, such as histamine from mast cells, which are responsible for inflammation and hypotension. For these reasons, the clinical use of $A_3$AR antagonists for the treatment of asthma and inflammatory disease has been suggested. Another suggestion says that this effect is mediated by the $A_{2b}$AR in human and canine mast cells. A bioavailable thiazole derivative that acts as a mixed $A_{2b}/A_3$ AR antagonist, has failed to attenuate bronchial hyper responsiveness to inhaled AMP in a phase Ib clinical trial in asthmatics, but has also been investigated for other indications.

Based on preclinical pharmacology and encouraging safety data in Phase I studies, the $A_3$AR agonists CF101 (16) and CF102 (17) have been tested in several Phase II trials for rheumatoid arthritis (Gessi et al. 2011). Based on anecdotal findings from this trial indicating that CF101 also improved indicators of dry eye syndrome, a follow-up Phase II trial was carried out, which determined that CF101 improved the clearance of corneal staining, tear break-up time and tear meniscus height with no side effects. In addition, active Phase II clinical trials are underway to test the efficacy of $A_3$AR agonists for the treatment of hepatocellular carcinoma and hepatitis (Fishman et al. 2012). Furthermore, experimental studies in mice suggest a possible use of $A_3$AR agonists in suppressing melanoma growth by inducing T cell-mediated adoptive immunity and in the control of chronic neuropathological pain (Fishman et al. 2012; Chen et al. 2012). These therapeutic
effects of CF101 are believed to be mediated by its inhibition (via cAMP and calcium signaling) of the oxidative burst and its anti-inflammatory activity (Gessi et al. 2002).

**Conclusion:**

It has been proved by different studies that extracellular adenosine is an important modulator of various physiological and pathological processes. With all these studies, it has emerged that adenosine receptors can be safely targeted by various ligands and various highly specific agonists and antagonists of adenosine receptors can be generated. As a result, increasing numbers of clinical trials testing of novel adenosine-based drugs in various indications have been initiated during the past decade.

Activation of adenosine receptors is beneficial in the treatment of various inflammatory and autoimmune disorders, pain, arrhythmia as well as sleep disorders and some metabolic disorders. Because of adenosine receptors are widely distributed in the body adenosine receptor agonists produces effects in almost all the tissues which make them difficult to use.

Selective antagonists of the adenosine receptors are more important. So, inhibition of adenosine receptors is having larger effects in treatment of the diseases like asthma, neuroprotection, diabetes, pain and cancer. The fact that a majority of humans already consume an adenosine antagonist, caffeine, on a daily basis of course also makes one wonder how much benefit can be derived by additional blockade.