Neurotoxicity Research, Vol. 3, pp. 545–556
Reprints available directly from the publisher
Photocopying permitted by license only

Adenosine A_{2A} Receptor Antagonists: Potential Therapeutic and Neuroprotective Effects in Parkinson's Disease

M. MORELLI[a] and J. WARDAS[b,*]

[a]*Department of Toxicology, University of Cagliari, Palazzo delle Scienze, Via Ospedale 72, 09124, Cagliari, Italy;* [b]*Department of Neuropsychopharmacology, Institute of Pharmacology, Polish Academy of Sciences, Krakow, Poland*

The most effective treatment of Parkinson's disease (PD) is, at present, the dopamine precursor L-3,4-dihydroxyphenyl-alanine (L-DOPA), however a number of disadvantages such as a loss of drug efficacy and severe side-effects (psychoses, dyskinesias and on–off phenomena) limit long-term, effective utilisation of this drug. Recent experimental studies in which selective antagonists of adenosine A_{2A} receptors were used, have shown an improvement in motor disabilities in animal models of PD. The A_{2A} antagonist [7-(2-phenylethyl)-5-amino-2-(2-furyl)-pyrazolo-(4,3-e)-1,2,4-triazolo(1,5--c)pyrimidine] (SCH 58261) potentiated the contralateral turning behavior induced by a threshold dose of L-DOPA or direct dopamine receptor agonists in unilaterally 6-hydroxydopamine (6-OHDA) lesioned rats, an effect accompanied by an increase in Fos-like-immunoreactivity in neurons of the lesioned striatum. Likewise, other A_{2A} receptor antagonists such as (3,7-dimethyl-1-propargylxanthine) (DMPX), [E-8-(3,4-dimethoxystyryl)-1,3-dipropyl-7-methylxanthine] (KF 17837) and [E-1,3-dietyl-8(3,4-dimethoxystyryl-7-methyl-3,7-dhydro-1H-purine-2,6-dione] (KW 6002) antagonized catalepsy induced by haloperidol or reserpine in the rat, whereas in non-human primate models of PD, KW 6002 reduced the rigidity and improved the disability score of MPTP-treated marmosets and cynomolgus monkeys. Moreover, in contrast to L-DOPA, selective A_{2A} receptor antagonists administered chronically did not produce dyskinesias and did not evoke tolerance in 6-OHDA and MPTP models of PD. An additional therapeutic potential of adenosine A_{2A} antagonists emerged from studies showing neuroprotective properties of these compounds in animal models of cerebral ischemia and excitotoxicity, as well as in the (1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine) (MPTP) model of PD. Adenosine A_{2A} receptor antagonists by reversing motor impairments in animal models of PD and by contrasting cell degeneration are some of the most promising compounds for the treatment of PD.

Keywords: Parkinson's disease; adenosine A_{2A} receptor antagonists; neuroprotection; dopamine D_1 and D_2 receptors; turning behavior; dyskinesia; striatum

Abbreviations: AC, adenylate cyclase; AD, adenosine; cAMP, cyclic adenosine monophosphate; CGS 15943, 5-amino-9-chloro-2-(2-furyl)-1,2,4-triazolo[1,5-c]quinazoline; CGS 21680, 2-[4-(2-carbonyl-ethyl)-phenethylamino]-5′-*N*-ethyl-carboxamidoadenosine; CP, caudate-putamen; CP 66713, 4-amino-1-phenyl-[1,2,4]-triazolo[4,3-a]quinoxaline; CSC, 8-(3-chlorostyryl)caffeine; DA, dopamine; DMPX, 3,7-dimethyl-1-propargylxanthine; DYN, dynorphin; ENK, enkephalin; GABA, γ-aminobutyric acid; Glu, glutamic acid; GP, globus pallidus; 2-HE-NECA, 2-hexyl-5′-*N*-ethylcarboxamidoadenosine; KF17837, (E)-1,3-dipropyl-8-(3,4-dimethoxystyryl)-7-methyl-3,7-dihydro-1H-purine-2,6-dione; KW

*Corresponding author. Tel.:+39-070-6758663. Fax: +39-070-6758612. Email: micmor@tin.it

6002, (*E*)-1,3-diethyl-8-(3,4-dimethoxystyryl)-7-methyl-3,7-dihydro-1H-purine-2,6-dione; L-DOPA, L-3,4-dihydroxyphenylalanine; MPTP, 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine; 6-OHDA, 6-hydroxydopamine; PD, Parkinson's disease; SCH 58261, 5-amino-7-(2-phenylethyl-2-(2-furyl)-pyrazolo[4,3-*e*]-1,2,4-triazolo[1,5-*c*]pyrimidine; SNr, substantia nigra pars reticulata; SP, substance P; STN, subthalamic nucleus; Th, thalamus; ZM 241385, 4-(2-[7-amino-2-(2-furyl)1,2,4-triazolo[2,3-*a*][1,3,5]triazin-5-ylamino]ethylphenol

ADENOSINE AND ITS RECEPTORS

Adenosine, which is formed within the cells from the hydrolysis of AMP by the action of ecto-5′ nucleotidase, modulates a variety of physiological processes in all tissues of mammals. Another pathway contributing to intracellular adenosine formation is from *S*-adenosylhomocysteine. In the extracellular compartment, the levels of adenosine also depend upon the rate of hydrolysis of ATP which is released from either neurons or glial cells. Extracellularly, adenosine concentrations are kept in equilibrium by a specific reuptake mechanism occurring through the action of a specialised bi-directional transporter. It is estimated that the levels of adenosine in the CNS range between 30 and 300 nM. Adenosine is then catabolyzed by the action of enzymes such as adenosine kinases and adenosine deaminase.

The action of adenosine is mediated through specific receptors located on cell membranes which belong to the family of G protein-coupled receptors. Currently, four adenosine receptors have been cloned and characterised: A_1, A_{2A}, A_{2B} and A_3 (Fredholm *et al.*, 1998). The main intracellular signaling pathways of these receptors are through the formation of cAMP, with A_1 and A_3 causing inhibition of adenylate cyclase, whereas A_{2A} and A_{2B} activate it. Other transduction mechanisms are also involved for each of the adenosine receptors, e.g. K^+ and Ca^{2+} channels. The molecular characteristics of adenosine receptors and intracellular signaling are described in detail elsewhere (Fredholm *et al.*, 1998; Olah and Stiles, 2000). Among adenosine receptors, A_{2A} receptors seem to play the most important role in the modulation of motor behavior. Their molecular, pharmacological and biochemical profiles and their distribution in the CNS are summarized in Table I.

DISTRIBUTION OF ADENOSINE A_{2A} RECEPTORS IN THE CNS

Adenosine A_{2A} receptors are predominantly located in basal ganglia structures (striatum, globus pallidus, substantia nigra), nucleus accumbens and tuberculum olfactorium (Jarvis and Williams, 1989; Rosin *et al.*, 1998). There are A_{2A} receptors in other brain areas, e.g. hippocampus, cerebral cortex and thalamic nuclei (Table I), with some differences found between the human brain and that of other animal species (Svenningsson *et al.*, 1997a). It remains, however, that using different methodological approaches all studies are consistent in describing high levels of A_{2A} receptors in the striatum. With regard to specific neuronal populations in the striatum, A_{2A} receptors are present in striatopallidal enkephalin-expressing neurons (Schiffmann *et al.*, 1991; Fink *et al.*, 1992). The same cells also express dopamine D_2 receptors, therefore both A_{2A} and D_2 receptors are segregated on the same neuronal pathway. In contrast, there are no A_{2A} receptors in neurons expressing D_1 receptors, substance P and dynorphin, which project from striatum to the substantia nigra (Schiffmann *et al.*, 1991; Fink *et al.*, 1992). It is worth noting that A_{2A} receptors are also present on glial cells.

ADENOSINE–DOPAMINE INTERACTION AS A BASIS FOR SEARCHING ANTIPARKINSONIAN DRUGS

Increasing number of studies suggest that adenosine A_{2A} receptors interact, either directly or indirectly, with different neurotransmitters

TABLE I Adenosine A_{2A} receptors in CNS: origin, transduction mechanism, distribution and selective agonists and antagonists

Human chromosomal location	*Amino acids*	*Major transduction mechanisms*	*Distribution in the CNS*	*Selective agonists*	*Selective antagonists*
22q11.2	412	Gs (Golf) stimulation of adenyl cyclase (AC)	*high:* striatum N. accumbens tub. olfactorium globus pallidus *low:* cerebellum hippocampus cortex thalamus substantia nigra	CGS 21680, 2-HE-NECA	CSC, KF17837, KW 6002, SCH 58261, ZM 241385, MSX-3

including dopamine (Ongini and Fredholm, 1996; Sebastiao and Ribeiro, 1996; Ferré, 1997).

Stimulation of adenosine A_{2A} receptors decreases the binding affinity of dopamine D_2 receptors (Ferré *et al.*, 1991; Dasgupta *et al.*, 1996) and elicits opposite effects to D_2 receptor activation at the level of second messenger systems (Fig. 1) and early-gene expression (Morelli *et al.*, 1995; Le Moine *et al.*, 1997; Olah and Stiles, 2000).

A_{2A} receptors are strongly involved in mediating effects related to the central control of motor activity. Stimulation of adenosine A_{2A} receptors induces sedation and catalepsy and inhibits the motor stimulating effects of dopamine receptor agonists (Durcan and Morgan, 1989; Heffner *et al.*, 1989; Barraco *et al.*, 1993; Popoli *et al.*, 1994; Kafka and Corbett, 1996; Ferré, 1997; Rimondini *et al.*, 1997). In rats with a unilateral 6-hydroxydopamine (6-OHDA) lesion of the dopaminergic nigrostriatal pathway, parenteral administration of A_{2A} receptor agonists reduces the contralateral turning behavior induced by direct dopamine receptor agonists (Vellucci *et al.*, 1993; Morelli *et al.*, 1994).

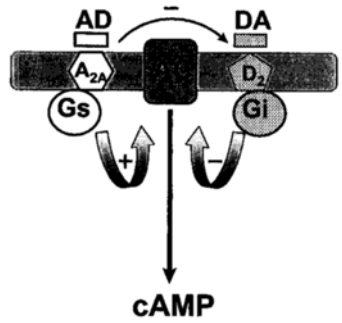

FIGURE 1 Schematic representation of the interaction between adenosine A_{2A} and dopamine D_2 receptors in the striatum. Stimulation of adenosine A_{2A} receptors decreases the affinity of dopamine D_2 receptors for dopamine and influences in the opposite manner adenylate cyclase. Activation of A_{2A} receptors enhances the AC and cAMP production through a Gs protein, whereas stimulation of D_2 receptors through a Gi protein inhibits it. AC—adenylate cyclase; AD—adenosine; DA—dopamine; A_{2A}—adenosine A_{2A} receptor; D_2—dopamine D_2 receptor; (+) stimulation; (−) inhibition.

In contrast, adenosine receptor antagonists, including caffeine and related methylxanthines, produce psychomotor stimulant effects by enhancing locomotor activity and schedule-controlled behavior (Schenk *et al.*, 1994; Garrett and Griffiths, 1997; Fredholm *et al.*, 1999). The expression of motor behaviors induced by methylxanthines is largely dependent on dopamine transmission as shown by counteraction of locomotion and turning behavior by either reserpine and α-methyl-p-thyrosine or by dopamine receptor antagonists (Herrera-Marschitz *et al.*, 1988; Josselyn and Beninger, 1991; Garrett and Holtzman, 1994a; Fenu and Morelli, 1998). The motor stimulant effects of caffeine appear to be related to an action on A_{2A} rather than A_1 receptors, since drugs blocking A_{2A} receptors induce motor stimulant effects, whereas A_1 receptor antagonists do not (Griebel *et al.*, 1991; Holtzman, 1991; Svenningsson *et al.*, 1997b; Hauber *et al.*, 1998).

Acute Adminstration of A_{2A} Antagonists in Animal Models of PD

PD, one of the most common neurodegenerative disorders, is characterized by a progressive degeneration of dopamine neurons of the substantia nigra pars compacta and a massive decrease of dopamine in the striatum.

Contralateral turning behavior in rats with unilateral 6-OHDA lesions of the dopaminergic nigrostriatal pathway is one of currently utilized animal models to test drugs active on PD (Ungerstedt, 1971). In this model, caffeine and theophylline induced contralateral turning behavior when administered alone and increased the turning behavior induced by direct dopamine agonists (Fuxe and Ungerstedt, 1974; Herrera-Marschitz *et al.*, 1988; Casas *et al.*, 1989; Jiang *et al.*, 1993; Garrett and Holtzman, 1994b).

These results have lead to the suggestion that adenosine antagonists might be beneficial to the treatment of PD. Studies in humans, however, are contradictory since some reports showed that caffeine produced no changes in the therapeutic response to antiparkinsonian drugs such as L-DOPA or bromocriptine (Shoulson and Chase, 1975; Kartzinel *et al.*, 1976), while others have reported an improvement of tremor but only after prolonged treatment (Mally and Stone, 1996). Moreover, a recent clinical survey has shown that heavy caffeine drinkers have a low risk to develop PD (Ross *et al.*, 2000).

At present L-DOPA is regarded as the most effective antiparkinsonian drug, although a number of disadvantages such as a loss of drug efficacy and severe side effects (psychoses, dyskinesias and on–off phenomena) limit long-term, effective utilization of this drug. Therefore, new, more effective drug treatments that are devoid of such side-effects are necessary.

The synthesis of selective and potent antagonists for adenosine A_{2A} receptors (Kanda *et al.*, 1994; Zocchi *et al.*, 1996) have opened new possibilities of studying, in animal models, whether adenosine A_{2A} receptors antagonists could be useful in the treatment of PD.

Studies based on these compounds have shown that the selective adenosine A_{2A} antagonists such as SCH 58261, KF 17837, or KW 6002 decreased haloperidol-induced catalepsy and reserpine-induced akinesia in intact animals and also potentiated the anticataleptic effect of L-DOPA (Kanda *et al.*, 1994; Mandhane *et al.*, 1997; Shiozaki *et al.*, 1999; Monopoli *et al.*, 2000). Moreover, in unilaterally 6-OHDA-lesioned rats it has been shown that blockade of adenosine A_{2A} receptors with SCH 58261 markedly increased the number of contralateral rotations induced by a threshold dose of L-DOPA as well as the expression of Fos-like immunoreactivity measured in the dorsal striatum and globus pallidus on the lesioned side (Fenu *et al.*, 1997). SCH 58261, as well as other A_{2A} antagonists, also potentiated the turning behavior and striatal c-fos expression induced by stimulation of dopamine D_1 receptors (Pinna *et al.*, 1996; Pollack and Fink, 1996; Le Moine *et al.*, 1997; Stromberg

TABLE II Effects of adenosine A_{2A} antagonists in animal models of Parkinson's disease

Experimental model	*Compounds*	*Effects*	*References*
6-OHDA lesion (contralateral rotations after L-DOPA, SKF 38393 or LY 171555)	Caffeine, Theophylline, SCH 58261, KW 6002, MSX-3	Potentiation of contralateral rotations	Fenu *et al.*, 1997; Jiang *et al.*, 1993; Monopoli *et al.*, 2000; Pinna *et al.*, 1996; Stromberg *et al.*, 2000
Haloperidol or reserpine-induced catalepsy	Caffeine, theophylline, SCH 58261, KF17837, MPX, KW 6002	Reversal of catalepsy Potentiation of L-DOPA effect	Kanda *et al.*, 1994; Mandhane *et al.*, 1997; Monopoli *et al.*, 2000; Shiozaki *et al.*, 1999
MPTP-treated primates	KW 6002	Reversal of motor disability Potentiation of L-DOPA effect	Grondin *et al.*, 1999; Kanda *et al.*, 1998

et al., 2000), whereas it did not induce any turning behavior when administered alone. Turning behavior induced by stimulation of dopamine D_2 receptors was also potentiated by SCH 58261 and by others A_{2A} antagonists (Fenu *et al.*, 1997; Stromberg *et al.*, 2000), suggesting that endogenous adenosine acting through A_{2A} receptors modulates the functions mediated by both D_1 and D_2 receptors.

The antiparkinsonian action of selective adenosine A_{2A} antagonists has been confirmed in non-human primate models of PD by Kanda *et al.* (1998) and Grondin *et al.* (1999), who have shown that the xanthine derivative KW 6002 reversed the motor disabilities produced by MPTP in marmosets and cynomolgus monkeys. A summary of these results is reported in Table II.

The potentiation of dopamine D_1- and D_2-mediated motor responses by A_{2A} receptor antagonists might be related to the segregation of D_1 and D_2 receptors on different striatal neuronal populations (Fig. 2). D_1 receptors are mainly localized on the direct striatonigral pathway (Gerfen *et al.*, 1990), whereas D_2 and A_{2A} receptors are colocalized on the indirect striato-pallido-nigral pathway (Schiffmann *et al.*, 1991; Fink *et al.*, 1992). Blockade of adenosine A_{2A} receptors by SCH 58261 might, therefore, play a direct positive role on D_2-mediated turning behavior by counteracting the opposite effect played by A_{2A} receptors at both receptor and transduction mechanism level in striato-pallido-nigral neurons, whereas it would indirectly influence dopamine D_1-mediated responses through an integration, in extrastriatal areas, of the responses mediated by the striato-pallido-nigral and stiatonigral pathways (Fig. 2). Neurochemical and behavioral studies have demonstrated that a concerted stimulation of the striatonigral pathway and an inhibition of the striato-pallido-nigral pathway are required to have full motor stimulation. Therefore, facilitation of the direct striatonigral pathway by D_1 agonists, associated with inhibition of the endogenous stimulatory A_{2A}

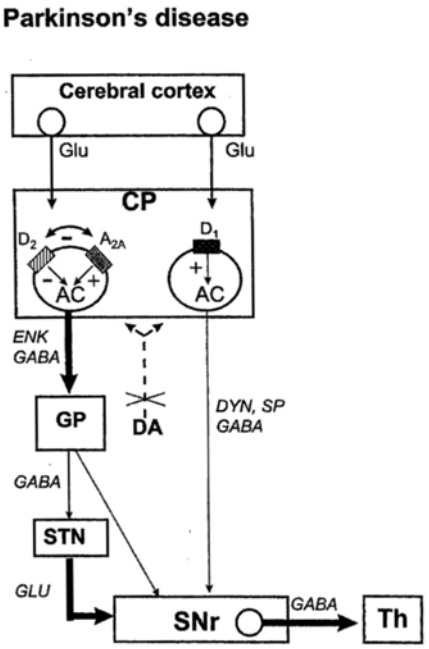

FIGURE 2 Schematic diagram illustrating changes occuring in the basal ganglia structures in Parkinson's disease and putative mechanisms by which adenosine A_{2A} antagonists might reverse these changes. In Parkinson's disease loss of DA cells in the substantia nigra pars compacta causes a cascade of events affecting activity of all components of the circuitry. The final results is an increased activity of GABAergic/ENK indirect output pathway, on which both A_{2A} and D_2 receptors are located, followed by a decreased activity of GABA pathway from GP to STN and increased activity of the glutamatergic subthalamo-nigral projection. These effects, together with inhibition of the direct GABA/SP/DYN pathway from the striatum to the SNr, lead to an increased activity of the GABA nigro-thalamic projection. Adenosine A_{2A} and dopamine D_2 receptors co-localized in the indirect striato-pallido-nigral pathway, dopamine D_1 receptors are localized on the direct striato-nigral pathway; D_1 and A_{2A} receptors stimulate cAMP formation whereas D_2 receptors inhibit cAMP production. Stimulation of the direct pathway through D_1 receptors or inhibition of the indirect pathway through D_2 receptors inhibits the activity of substantia nigra, whereas stimulation of the indirect pathway through A_{2A} receptors disinhibits SN activity. Adenosine A_{2A} antagonists can therefore potentiate dopamine-mediated inhibition of SNr activity either by a direct interaction with dopamine D_2 receptors at the level of the striato-pallido-nigral pathway or by an indirect interaction with D_1 receptors at the level of substantia nigra where the responses mediated by the striato-pallido-nigral and striatonigral pathways are integrated. Thickness of arrows indicate the degree of activation of the pathways. AC—adenylate cyclase; A_{2A}—adenosine A_{2A} receptor; CP—caudate-putamen; DA—dopamine; D_1—dopamine D_1 receptor; D_2—dopamine D_2 receptor; DYN—dynorphin; ENK—enkephalin; GABA-γ—aminobutyric acid; Glu—glutamic acid; GP—globus pallidus; SNr—substantia nigra pars reticulata; SP—substance P; STN—subthalamic nucleus; Th—thalamus

receptor tone on the indirect striato-pallido-nigral pathway, might explain the synergism between D_1 receptor stimulation and A_{2A} receptor blockade (Ongini *et al.*, 1996; Pinna *et al.*, 1996; Ferré *et al.*, 1997).

Besides the well-known antagonistic interaction between adenosine A_{2A} and dopamine D_2 receptors, A_{2A} adenosine receptors can operate independently of dopamine D_2 receptors (Kirk and Richardson, 1994; Kurokawa *et al.*, 1994; Mori *et al.*, 1996; Richardson *et al.*, 1997). These results were recently confirmed by studies in dopamine D_2 or A_{2A} receptor knock-out mice (Aoyama *et al.*, 2000; Zahniser *et al.*, 2000; Chen *et al.*, 2001) which showed both dependent and independent mechanisms in the A_{2A}/ D_2 receptor interaction.

All these results strongly suggest that despite an intramembrane A_{2A}/D_2 receptor interaction, other mechanisms involving GABA or acetylcholine release from presynaptic sites in the striatum are also of importance for A_{2A}/D_2 receptor interaction.

Chronic Administration of Adenosine A_{2A} Antagonists

It is well known that the major problems related to L-DOPA therapy in PD is the progressive loss of L-DOPA efficacy, accompanied by motor fluctuations and dyskinesias which limit long-term effective application of this drug.

It has been shown that the motor stimulant effects of the non-selective adenosine A_1/A_{2A} receptor antagonist caffeine which, together with theophylline, was first proposed as a potential candidate for the treatment of PD, exhibit tolerance that develops over a few days of treatment in experimental animals (Holtzman, 1991; Fredholm *et al.*, 1999).

Interestingly, the repeated administration of the adenosine A_{2A} receptor antagonist, SCH 58261, did not induce tolerance to contralateral turning behavior in unilaterally 6-OHDA-

TABLE III Neuroprotective effects of adenosine A_{2A} receptor antagonists

Experimental model	*Compound*	*Effect*	*References*
Global or focal forebrain ischemia in gerbil or rat, neonatal hypoxia-ischemia, hyperglycaemic cerebral ischemia	CGS 15943, CSC, CP 66713, SCH 58261, ZM 241385	Neuroprotection	Bona *et al.*, 1997; Gao and Phillis, 1994; Higashi *et al.*, 2000; Monopoli *et al.*, 1998; Phillis, 1995; Von Lubitz *et al.*, 1995
Focal ischemia in A_{2A} knock-out mice	A_{2A} KO mice	Neuroprotection	Chen *et al.*, 1999
Kainic or quisqualic acid-induced excitoxicity (peripheral or intra-hippocampal injection)	ZM 241385, SCH 58261, CSC	Neuroprotection in CA1, CA2, CA3	Jones *et al.*, 1998 a,b; Stone and Behan, 2000
MPTP model of PD	A_{2A} KO mice CSC	Neuroprotection	Chen *et al.*, 2001 a,b

lesioned rats (Pinna *et al.*, 2001). SCH 58261, in fact, induced a similar enhancement of L-DOPA turning behavior following single or seven-day administration regimens. When this compound was injected for 14 days, the effect was even stronger than that observed after a single administration (Pinna *et al.*, 2001). These data are consistent with studies performed in marmosets and cynomologus monkeys showing that another A_{2A} antagonist, KW 6002, retained its activity over a 21-days treatment schedule (Kanda *et al.*, 1998). Moreover, Pinna *et al.* (2001) observed no sensitization to the contralateral turning behavior in the repeated treatment (19 days) with SCH 58261 and L-DOPA, in contrast to the intermittent chronic L-DOPA. A similar type of contralateral rotation without sensitization was obtained after chronic administration of the dopamine agonist bromocriptine which has a low dyskinetic potential (Henry *et al.*, 1998). Again, these data, in rat models of dyskinesia, are consistent with studies showing that KW 6002 did not produce any dyskinesia in primates previously primed with L-DOPA to exhibit involuntary movements (Kanda *et al.*, 1998; Grondin *et al.*, 1999).

NEUROPROTECTIVE ROLE OF ADENOSINE A_{2A} ANTAGONISTS

It is well established that adenosine plays a pivotal role in neurodegeneration (Rudolphi *et al.*, 1992; Ongini and Schubert, 1998). Low concentrations of adenosine, normally present in the CNS extracellular fluid, increase dramatically following hypoxia or ischemia. In these pathological conditions, adenosine-potentiating agents which elevate endogenous adenosine levels, either by inhibiting its degradation (adenosine deaminase and kinase inhibitors) or by inhibiting adenosine transport, offer protection against ischemic or excitotoxic neuronal damage. Both A_1 and A_{2A} adenosine receptors play a role in neuroprotective mechanisms, although the same

net results can be achieved by either stimulating A_1 receptors or blocking A_{2A} receptors (Ongini and Schubert, 1998).

Adenosine A_1 agonists consistently attenuate ischemic or excitotoxic neuronal damage (Phillis, 1995; de Mendonça *et al.*, 2000). Much less is known about the neuroprotective role of adenosine A_{2A} receptors. Stimulation of adenosine A_{2A} receptors by a selective agonist, CGS21680, reduces ischemic or excitotoxic hippocampal damage (Sheardown and Knutsen, 1996; Jones *et al.*, 1998b); however, these neuroprotective properties can be due to actions occurring in the periphery rather than at neuronal sites. The main mechanisms which may account for the A_{2A}-mediated protection include: vasodilation, inhibition of platelet aggregation and supression of neutrophil superoxide generation (Ongini and Schubert, 1998).

The neuroprotective properties of adenosine A_{2A} antagonists in different models of neurodegeneration have been described by several studies. The selective adenosine A_{2A} receptor antagonists CSC and ZM 241385 as well as the less selective CGS 15943 and CP 66,713 were able to ameliorate the hippocampal cell injury following global forebrain ischemia in gerbils or rats (Gao and Phillis, 1994; Phillis, 1995; von Lubitz *et al.*, 1995; Higashi *et al.*, 2000). Similarly, the selective A_{2A} receptor antagonist, SCH 58261, could reduce cortical infarct volume in a focal cerebral ischemia model of permanent middle cerebral artery occlusion (Monopoli *et al.*, 1998). This compound also decreased brain damage in neonatal rats in which the unilateral carotid artery was severed (Bona *et al.*, 1997). Administration of selective adenosine A_{2A} receptor antagonists (SCH 58261, ZM 241385 or CSC) also decreased the neuronal cell death observed in hippocampal regions following kainic, kynurenic and quinolinic acid administration in rodents (Jones *et al.*, 1998a,b; Stone and Behan, 2000). In summary, fairly consistently, selective adenosine A_{2A} antagonists were able to exert neuroprotective effects.

Studies in genetically manipulated mice confirmed a role for adenosine A_{2A} receptors in mediating hypoxic/ischemic damage. Cerebral infarction and neurological deficits were attenuated in adenosine A_{2A} receptor knock-out mice (A_{2A} KO) subject to a temporary middle cerebral artery occlusion in comparison with the wild type littermates (Chen *et al.*, 1999). By using the genetic approach it was also shown that in the mice MPTP model of PD, which resembles the biochemical and neuropathological features of the disease well, dopamine depletion and dopamine transporter decrease were significantly attenuated in the striatum of A_{2A} KO mice and in mice pretreated with the selective A_{2A} antagonist, CSC (Chen *et al.*, 2001). A summary of these results is reported in Table III.

It is well known that A_1 and A_{2A} receptors affect in an opposite direction the release of glutamate, which determines the risk of excitotoxic nerve cell damage, where stimulation of A_1 receptor inhibits and A_{2A} stimulates glutamate release. Stimulation of adenosine A_{2A} receptors enhances the release of glutamate under both ischemic and non-ischemic conditions (O'Regan *et al.*, 1992; Simpson *et al.*, 1992; Popoli *et al.*, 1995), therefore blockade of A_{2A} receptors might afford neuroprotection after ischemia because of reduced glutamate release-induced excitotoxicity.

Functional evidences exist showing that both A_1 and A_{2A} subtypes of adenosine receptors can coexist in the same nerve terminal. It has been shown that the stimulation of A_{2A} receptors decreased the binding of adenosine A_1 receptors in hippocampal and striatal synaptosomes and attenuated the ability of A_1 agonists to inhibit excitability and synaptic transmission in the hippocampus (Dixon *et al.*, 1997; de Mendonça *et al.*, 2000; Sebastiao and Ribeiro, 2000). Thus, activation of A_{2A} receptors leads to a decrease in the effects mediated by A_1 receptors. This functional interaction between both subtypes of adenosine receptors seems to suggest that the action of endogenous adenosine, mediated by A_1

receptors, might be attenuated if there is a concomitant activation of A_{2A} receptors. Therefore, on the basis of above-mentioned studies it has been suggested that the beneficial effect of selective adenosine A_{2A} antagonists might be, at least in part, due to the relief of tonic inhibition upon adenosine A_1 receptors (de Mendonça *et al.*, 2000). Apart from the reduction of glutamate release, there are several other mechanisms by which adenosine A_{2A} receptor antagonists can achieve neuroprotection, e.g. by diminishing the state of activation of microglia and diminishing cytokine release (TNF-alfa, IL-1beta) (Ongini and Schubert, 1998). However, the mechanisms underlying these neuroprotective properties have not been explored yet.

Despite a great interest, up to now there were no clear-cut data showing specific changes of adenosine levels or receptors in discrete brain areas in patients suffering from PD (Martinez-Mir *et al.*, 1991). Recently Hurley *et al.* (2000) showed significant changes in the level of A_{2A} mRNA in caudate-putamen (a decrease) and substantia nigra pars reticulata (an increase). Since those patients were receiving dopaminergic medication at the time of death it is conceivable that this would have caused the alterations in A_{2A} mRNA expression and that such changes may not be present in brains from untreated patients. Therefore, further studies are needed to clarify whether dopamine deficiency might alter adenosine A_{2A} transmission.

CONCLUSIONS

In animal models of PD reversal of motor dysfunctions as well as neuroprotective properties of selective adenosine A_{2A} receptor antagonists have been shown. Moreover, in these models, long-term administration of A_{2A} receptor antagonists is devoid of the motor complications which accompany long-term L-DOPA therapy. At the moment adenosine A_{2A} receptor antagonists appear to be some of the most promising compounds for the treatment of PD.

References

Aoyama, S., Kase, H. and Borelli, E. (2000) "Rescue of locomotor impairment in dopamine D_2 receptor-deficient mice by an adenosine A_{2A} receptor antagonist", *J. Neurosci.* **20**, 5848–5852.

Barraco, R.A., Martens, K.A., Parizon, M. and Normile, H.J. (1993) "Adenosine A_{2a} receptors in the nucleus accumbens mediate locomotor depression", *Brain Res. Bull.* **31**, 397–404.

Bona, E., Aden, U., Gilland, E., Fredholm, B.B. and Hagberg, H. (1997) "Neonatal cerebral hypoxia ischemia: the effect of adenosine receptor antagonists", *Neuropharmacology* **36**, 1327–1338.

Casas, M., Ferre, S., Cobos, A., Grau, J.N. and Jané, F. (1989) "Relationship between rotational behaviour induced by apomorphine and caffeine in rats with unilateral lesion of the nigrostriatal pathway", *Neuropharmacology* **28**, 407–409.

Chen, J.F., Huang, Z., Ma, J., Zhu, J.M., Moratalla, R., Standaert, D., Moskowitz, M.A., Fink, J.S. and Schwarzschild, M.A. (1999) "A_{2A} adenosine receptors deficiency attenuate brain injury induced by transient focal ischemia in mice", *J. Neurosci.* **19**, 9192–9200.

Chen, J.F., Moratalla, R., Standaert, D., Impagnatiello, F., Grandy, D.K., Cuellar, B., Rubistein, M., Beilstein, M., Hackett, E., Fink, J.S., Low, M.J., Ongini, E. and Schwarzschild, M.A. (2001a) "The role of the D_2 dopamine receptor (D_2R) in A_{2A} adenosine receptor ($A_{2A}R$)-mediated behavioral and cellular responses as revealed by A_{2A} and D_2 receptor knockout mice", *Proc. Natl. Acad. Sci.* **98**, 1970–1975.

Chen, J.F., Xu, K., Petzer, J.P., Staal, R., Xu, Y-H., Beilstein, M.A., Sonsalla, P.K., Castagnoli, K., Castagnoli, Jr, N. and Schwarzschild, Jr, M.A. (2001b) "Neuroprotection by caffeine and A2A adenosine receptor inactivation in a model of Parkinson's Disease", *J. Neurosci.* **21**, 143.

Dasgupta, S., Ferré, S., Kull, B., Hedlund, P.B., Finnman, U.B., Ahlberg, S., Arenas, E., Fredholm, B.B. and Fuxe, K. (1996) "Adenosine A_{2A} receptors modulate the binding characteristics of dopamine D_2 receptors in stably cotransfected cells", *Eur. J. Pharmacol.* **316**, 325–331.

Dixon, A.K., Widdwson, P.J. and Richardson, P.J. (1997) "Desensitisation of the adenosine A_1 receptor by the A_{2A} receptor in the striatum", *J.Neurochem.* **69**, 315–321.

Durcan, M.J. and Morgan, P.F. (1989) "Evidence for adenosine A_2 receptor involvement in the hypomobility effects of adenosine analogues in mice", *Eur. J. Pharmacol.* **168**, 285–290.

Fenu, S. and Morelli, M. (1998) "Motor stimulant effects of caffeine in 6-hydroxydopamine-lesioned rats are dependent on previous stimulation of dopamine receptors: a different role of D_1 and D_2 receptors", *Eur. J. Neurosci.* **10**, 1878–1884.

Fenu, S., Pinna, A., Ongini, E. and Morelli, M. (1997) "Adenosine A_{2A} receptor antagonism potentiates L-DOPA-induced turning behaviour and c-fos expression in 6-hydroxydopamine-lesioned rats", *Eur. J. Pharmacol.* **321**, 143–147.

Ferré, S. (1997) "Adenosine-dopamine interactions in the ventral striatum. Implications for the treatment of schizophrenia.", *Psychopharmacology* **133**, 107–120.

Ferré, S., Von Euler, G., Johansson, B., Fredholm, B.B. and Fuxe, K. (1991) "Stimulation of high affinity adenosine A-2 receptors decreases the affinity of dopamine D-2 receptors in rat striatal membranes", *Proc. Natl. Acad. Sci. USA.* **88**, 7238–7241.

Ferré, S., Fredholm, B.B., Morelli, M., Popoli, P. and Fuxe, K. (1997) "Adenosine-dopamine receptor-receptor interactions as an integrative mechanism in the basal ganglia", *Trend Neurosci.* **20**, 482–486.

Fink, J.S., Weaver, D.R., Rivkees, S.A., Peterfreund, R.A., Pollack, A.E., Adler, E.M. and Reppert, S.M. (1992) "Molecular cloning of the rat A_2 adenosine receptor: selective co-expression with D_2 dopamine receptors in rat striatum", *Mol. Brain Res.* **14**, 186–190.

Fredholm, B.B., Ijzerman, A.P., Jacobson, K.A., Linden, J. and Stiles, G.L. (1998) "Adenosine receptors", IUPHAR Compendium of Receptor Characterization and Classication (IUPHAR Media, London), pp. 48–57.

Fredholm, B.B., Battig, K., Holmen, J., Nehlig, A. and Zvartau, E.E. (1999) "Actions of caffeine in the brain with special reference to factors that contribute to its widespread use", *Pharmacol. Rev.* **51**, 83–133.

Fuxe, K. and Ungerstedt, U. (1974) "Action of caffeine and theophyllamine on supersensitive dopamine receptors: considerable enhancement of receptor response to treatment with DOPA and dopamine receptor agonists", *Med. Biol.* **52**, 48–54.

Gao, Y. and Phillis, J.W. (1994) "CGS 15943, an adenosine A2 receptor antagonist, reduces cerebral ischaemic injury in the Mongolian gerbil", *Life Sci.* **55**, 61–65.

Garrett, B.E. and Griffiths, R.R. (1997) "The role of dopamine in the behavioral effects of caffeine in animals and humans", *Pharmacol. Biochem. Behav.* **57**, 533–541.

Garrett, B.E. and Holtzman, S.G. (1994a) "D_1 and D_2 dopamine receptor antagonists block caffeine-induced stimulation of locomotor activity in rats", *Pharmacol. Biochem. Behav.* **47**, 89–94.

Garrett, B.E. and Holtzman, S.G. (1994b) "Caffeine cross-tolerance to selective dopamine D_1 and D_2 receptor agonists but not to their synergistic interaction", *Eur. J. Pharmacol.* **262**, 65–75.

Gerfen, C.R., Engber, T.M., Mahan, L.C., Susel, Z., Chase, T.N., Monsma, Jr, F.J. and Sibley, Jr, D.R. (1990) "D1 and D2 dopamine receptor-regulated gene expression of striatonigral and striatopallidal neurons", *Science* **250**, 1429–1432.

Griebel, G., Saffroy-Spittler, M., Misslin, R., Remmy, D., Vogel, E. and Bourguignon, J.J. (1991) "Comparison of the behavioral effects of an adenosine A1/A2-receptor antagonist, CGS 5943A, and an A1-selective antagonist, DPCPX", *Psychopharmacology* **103**, 541–544.

Grondin, R., Bédard, P.J., Hadj Tahar, A., Grégoire, L., Mori, A. and Kase, H. (1999) "Antiparkinsonian effect of a new selective adenosine A_{2A} receptor antagonist in MPTP-treated monkeys", *Neurology* **52**, 1673–1677.

Hauber, W., Nagel, J., Sauer, R. and Muller, C.E. (1998) "Motor effects induced by a blockade of adenosine A_{2A} receptors in the caudate-putamen", *Neroreport* **9**, 1803–1806.

Heffner, T.G., Wiley, J.N., Williams, A.E., Bruns, R.F., Coughenour, L.L. and Downs, D.A. (1989) "Comparison of the behavioral effects of adenosine agonists and dopamine antagonists in mice", *Psychopharmacology* **98**, 31–37.

Henry, B., Crossman, A.R. and Brotchie, J.M. (1998) "Characterisation of enhanced behavioral responses to L-DOPA following repeated administration in the 6-hydroxydopamine-lesioned rat model of Parkinson's disease", *Exp. Neurol.* **151**, 334–342.

Herrera-Marschitz, M., Casas, M. and Ungerstedt, U. (1988) "Caffeine produces contralateral rotation in rats with unilateral dopamine denervation: comparisons with apomorphine-induced responses", *Psychopharmacology* **94**, 38–45.

Higashi, H., Marwaha, A.S., Memo, J.R. and Winn, H.R. (2000) "Correlation between hippocampal injury and neurobehavior following normo- and hyperglycemic cerebral ischemia, effect of theophylline and ZM241385", *Drug Dev. Res.* **50**, 71–092.

Holtzman, S. and G, . (1991) "CGS 15943, a non xanthine adenosine receptor antagonist: effects on locomotor activity of non tolerant and caffeine-tolerant rats", *Life Sci.* **49**, 1563–1570.

Hurley, M.J., Mash, D.C. and Jenner, P. (2000) "Adenosine A_{2A} receptor mRNA expression in Parkinson's disease", *Neurosci. Lett.* **291**, 54–58.

Jarvis, M.F. and Williams, M. (1989) "Direct autoradiographic localization of adenosine A_2 receptors in the rat brain using the A_2-selective agonist, [3H]CGS 21680", *Eur. J. Pharmacol.* **168**, 243–246.

Jiang, H., Jackson-Lewis, V., Muthane, U., Dollison, A., Ferreira, M., Espinosa, A., Parsons, B. and Przedborski, S. (1993) "Adenosine receptor antagonists potentiate dopamine receptor agonist-induced rotational behavior in 6-hydroxydopamine-lesioned rats", *Brain Res.* **613**, 347–351.

Jones, P.A., Smith, R.A. and Stone, T.W. (1998a) "Protection against hippocampal kainate excitotoxicity by intracerebral administration of an adenosine A_{2A} receptor antagonist", *Brain Res.* **800**, 328–335.

Jones, P.A., Smith, R.A. and Stone, T.W. (1998b) "Protection against kainate-induced excitotoxicity by adenosine A_{2A} receptor agonists and antagonists", *Neuroscience* **85**, 229–237.

Josselyn, S.A. and Beninger, R.J. (1991) "Behavioral effects of intrastriatal caffeine mediated by adenosinergic modulation of dopamine", *Pharmacol. Biochem. Behav.* **39**, 97–103.

Kafka, S.H. and Corbett, R. (1996) "Selective adenosine A_{2A} receptor/dopamine D_2 receptor interactions in animal models of schizophrenia", *Eur. J. Pharmacol.* **295**, 147–154.

Kanda, T., Shiozaki, S., Shimada, J., Suzuki, F. and Nakamura, J. (1994) "KF17837: a novel selective adenosine A_{2A} antagonist with anticataleptic activity", *Eur. J. Pharmacol.* **256**, 263–268.

Kanda, T., Jackson, M.J., Smith, L.A., Pearce, R.K.B., Nakamura, J., Kase, H., Kuwana, Y. and Jenner, P. (1998) "Adenosine A_{2A} antagonist: a novel antiparkinsonian agent that does not provoke dyskinesia in parkinsonian monkeys", *Ann. Neurol.* **43**, 507–513.

Kartzinel, R., Shoulson, I. and Calne, D.B. (1976) "Studies with bromocriptine III. Concomitant administration of caffeine to patients with idopathic parkinsonism", *Neurology* **26**, 741–743.

Kirk, I.P. and Richardson, P.J. (1994) "Adenosine A_{2a} receptor-mediated modulation of striatal [3H]GABA and [3H]Acetylcholine release", *J. Neurochem* **62**, 960–966.

Kurokawa, M., Kirk, I.P., Kirkpatric, K.A., Kase, H. and Richardson, P.J. (1994) "Inhibition by KF 17837 of adenosine receptor-mediated modulation of striatal GABA and ACh release", *Br. J. Pharmacol.* **113**, 43–48.

von Lubitz, D.J.K.E., Lin, R.C.S. and Jacobson, K.A. (1995) "Cerebral ischemia in gerbils: effects of acute and chronic treatment with adenosine A_{2A} receptor agonist and antagonist", *Eur. J. Pharmacol.* **287**, 295–302.

Mally, J. and Stone, T.W. (1996) "Potential role of adenosine antagonist therapy in pathological tremor disorders", *Pharmacol. Ther.* **72**, 243–250.

Mandhane, S.N., Chopde, C.T. and Ghosh, A.K. (1997) "Adenosine A2 receptors modulate haloperidol-induced catalepsy in rats", *Eur. J. Pharmacol.* **328**, 135–141.

Martinez-Mir, M.I., Probst, A. and Palacios, J.M. (1991) "Adenosine A_2 receptors: selective localization in the human basal ganglia and alterations with disease", *Neuroscience* **42**, 697–706.

de Mendonça, A., Sebastião, A.M. and Ribeiro, J.A. (2000) "Adenosine: does it have a neuroprotective role after all?", *Brain Res. Rev.* **33**, 258–274.

Le Moine, C., Svenningsson, P., Fredholm, B.B. and Bloch, B. (1997) "Dopamine–adenosine interactions in the striatum and globus pallidum: inhibition of striatopallidal neurons through either D_2 or A_{2A} receptors enhances D_1 receptor-mediated effects on c-foc expression", *J. Neurosci.* **17**, 8038–8048.

Monopoli, A., Lozza, G., Forlani, A., Mattavelli, A. and Ongini, E. (1998) "Blockade of A_{2A} adenosine receptors by SCH 58261 results in neuroprotective effect in cerebral ischemia in mice", *NeuroReport* **9**, 3955–3959.

Monopoli, A., Impagnatiello, F., Bastia, E., Fredduzzi, S. and Ongini, E. (2000) "Anti-parkinsonian effects of selective A_{2A} adenosine receptor antagonists in relevant rodent models", *Drug Dev. Res.* **50**, 70–87.

Morelli, M., Fenu, S., Pinna, A. and Di Chiara, G. (1994) "Adenosine A_2 receptors interact negatively with dopamine D_1 and D_2 receptors in unilaterally 6-hydroxydopamine-lesioned rats", *Eur. J. Pharmacol.* **251**, 21–25.

Morelli, M., Pinna, A., Wardas, J. and Di Chiara, G. (1995) "Adenosine A_2 receptors stimulate c-fos expression in striatal neurons of 6-hydroxydopamine-lesioned rats", *Neuroscience* **67**, 49–55.

Mori, A., Shindou, T., Ichimura, M., Nonaka, H. and Kase, H. (1996) "The role of adenosine A_{2A} receptors in regulating GABAergic synaptic transmission in striatal medium spiny neurons", *J. Neurosci.* **16**, 605–611.

Olah, M. and Stiles, G.L. (2000) "The role of receptor structure in determining adenosine receptor activity", *Pharmacol. & Therap.* **85**, 55–75.

Ongini, E. and Fredholm, B.B. (1996) "Pharmacology of adenosine A_{2A} receptors", *Trends Pharmacol. Sci.* **17**, 364–372.

Ongini, E. and Schubert, P. (1998) "Neuroprotection induced by stimulating A1 or blocking A_{2A} adenosine receptors: an apparent paradox", *Drug Dev. Res.* **45**, 387–393.

Ongini, E., Dionisotti, S., Morelli, M., Ferré, S., Svenningsson, P., Fuxe, K. and Fredholm, B.B. (1996) "Neuropharmacology of the adenosine A_{2A} receptors", *Drug Dev. Res.* **39**, 450–460.

O'Regan, M.H., Simpson, R.E., Perkins, L.M. and Phillis, J.W. (1992) "The selective A2 agonist CGS 21680 enhances excitatory transmitter amino acid release from the ischemic rat cerebral cortex", *Neurosci. Lett.* **138**, 169–172.

Phillis, J.W. (1995) "The effects of selective A_1 and A_{2a} adenosine receptor antagonists on cerebral ischemic injury in the gerbil", *Brain Res.* **705**, 79–84.

Pinna, A., Di Chiara, G., Wardas, J. and Morelli, M. (1996) "Blockade of A_{2a} adenosine receptors positively modulates turning behaviour and c-Fos expression induced by D_1 agonists in dopamine-denervated rats", *Eur. J. Neurosci.* **8**, 1176–1181.

Pinna, A., Fenu, S. and Morelli, M. (2001) "Motor stimulant effects of the adenosine A_{2A} receptor antagonist SCH 58261 do not develop tolerance after repeated treatments in 6-hydroxydopamine-lesioned rats", *Synapse* **39**, 233–238.

Pollack, A.E. and Fink, J.S. (1996) "Synergistic interaction between an adenosine antagonist and a D_1 dopamine agonist on rotational behavior and striatal c-Fos induction in 6-hydroxydopamine-lesioned rats", *Brain Res.* **743**, 124–130.

Popoli, P., Pezzola, A., Reggio, R. and Scotti de Carolis, A. (1994) "Modulation of striatal adenosine A1 and A2 receptors induces rotational behavior in response to dopaminergic stimulation in intact rats", *Eur. J. Pharmacol.* **257**, 21.

Popoli, P., Betto, P., Reggio, R. and Ricciarello, G. (1995) "Adenosine A_{2A} receptor stimulation enhances striatal extracellular glutamate levels in rats", *Eur. J. Pharmacol.* **287**, 215–217.

Richardson, P.J., Kase, H. and Jenner, P.G. (1997) "Adenosine A_{2A} receptor antagonists as new agents for the treatment of Parkinson's disease", *Trends Pharmacol. Sci.* **18**, 338–344.

Rimondini, R., Ferre, S., Ögren, S.O. and Fuxe, K. (1997) "Adenosine A_{2A} agonists: a potential new type of atypical antipsychotics", *Neuropsychopharmacology* **17**, 81–91.

Rosin, D.L., Robeva, A., Woodard, R.L., Guyenet, P.G. and Linden, J. (1998) "Immunohistochemical localization of adenosine A_{2A} receptors in the rat central nervous system", *J. Comp. Neurol.* **401**, 163–186.

Ross, G.W., Abbott, R.D., Petrovitch, H., Morens, D.M., Grandinetti, A., Tung, K.-H., Tanner, C.M., Masaki, K.H., Blanchette, P.L., Curb, J.D., Popper, J.S. and White, L.R. (2000) "Association of coffee and caffeine intake with the risk of Parkinson disease", *J. Med. American Ass.* **283**, 2674–2679.

Rudolphi, K., Schubert, P., Parkinson, F.E. and Fredholm, B.B. (1992) "Neuroprotective role of adenosine in cerebral ischaemia", *Trends Pharmacol. Sci.* **13**, 439–445.

Schenk, S., Valadez, A., Horger, B.A., Snow, S. and Wellman, P.J. (1994) "Interaction between caffeine and cocaine in tests of self-administration", *Behav. Pharmacol.* **5**, 153–158.

Schiffmann, S.N., Jacobs, O. and Vanderhaeghen, J.J. (1991) "The striatal restricted adenosine A2 receptor (RDC8) is expressed by enkephalin but not by substance P neurons. An in situ hybridization histochemistry study", *J. Neurochem.* **57**, 1062–1067.

Sebastiao, A.M. and Ribeiro, J.A. (1996) "Adenosine A_{2A} receptor-mediated excitatory actions on the nervous system", *Prog. Neurobiol.* **48**, 167–189.

Sebastiao, A.M. and Ribeiro, J.A. (2000) "Fine-tuning neuromodulation by adenosine", *Trends Pharmacol Sci.* **21**, 341–346.

Sheardown, M.J. and Knutsen, L.J.S. (1996) "Unexpected neuroprotection observed with the adenosine A_{2A} receptor agonist CGS 21680", *Drug Dev. Res.* **39**, 108–114.

Shiozaki, S., Ichikawa, S., Nakamura, J., Kitamura, S., Yamada, K. and Kuwana, Y. (1999) "Actions of adenosine A_{2A} receptor antagonist KW-6002 on drug-induced catalepsy and hypokinesia caused by reserpine or MPTP", *Psychopharmacology* **147**, 90–95.

Shoulson, I. and Chase, T. (1975) "Caffeine and the antiparkinsonian response to levodopa or pirebidil", *Neurology* **25**, 722–724.

Simpson, R.E., O'Regan, M.H., Perkins, L.M. and Phillis, J.W. (1992) "Excitatory transmitter amino acid release from the ischemic rat cerebral cortex: effects of adenosine receptor agonists and antagonists", *J. Neurochem.* **58**, 1683–1690.

Stromberg, I., Popoli, P., Muller, C., Feree, S. and Fuxe, J. (2000) "Electrophysiological and behavioural evidence for an antagonistic modulatory role of adenosine A_{2A} receptor regulation in the rat dopamine-denervated striatum", *Eur. J. Neurosci.* **12**, 4033–4037.

Stone, T.W. and Behan, W.M.H. (2000) "Adenosine receptors, kynurenines and neuroprotection", *Drug Dev. Res.* **50**, 15.

Svenningsson, P., Hall, H., Sedvall, G. and Fredholm, B.B. (1997a) "Distribution of adenosine receptors in the post-mortem human brain: an extended autoradiographic study", *Synapse* **27**, 322–335.

Svenningsson, P., Nomikos, G.G., Ongini, E. and Fredholm, B.B. (1997b) "Antagonism of adenosine A_{2A} receptors underlies the behavioural activating effect of caffeine and is associated with reduced expression of messenger RNA for NGFI-A and NGFI-B in caudate-putamen and nucleus accumbens", *Neuroscience* **79**, 753–764.

Ungerstedt, U. (1971) "Postsynaptic supersensitivity after 6-hydroxydopamine induced degeneration of the nigros-triatal dopamine system", *Acta. Physiol. Scand.* **367**, 69–74.

Vellucci, S.V., Sirinathsinghji, D.J.S. and Richardson, P.J. (1993) "Adenosine A_2 receptor regulation of apomorphine-induced turning in rats with unilateral striatal dopamine denervation", *Psychopharmacology* **111**, 383–388.

Zahniser, N.R., Simosky, J.K., Mayfield, R.D., Negri, C.A., Hanania, T., Larson, G.A., Kelly, M.A., Grandy, D.K., Rubinstein, M., Low, M.J. and Fredholm, B.B. (2000) "Functional uncoupling of adenosine A_{2A} receptors and reduced response to caffeine in mice lacking dopamine D_2 receptors", *J. Neurosci.* **20**, 5949–5957.

Zocchi, C., Ongini, E., Conti, A., Monopoli, A., Negretti, A., Baraldi, P.G. and Dionisotti, S. (1996) "The non-xanthine heterocyclic compound SCH 58261 is a new potent and selective A_{2A} adenosine receptor antagonist", *J. Pharmacol. Exp. Ther.* **276**, 389–404.