Adenosine A_{2A} receptor signalling in the rat dentate gyrus and in the oligodendrocyte progenitor cells.

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Adenosine A_{2A} receptors ($A_{2A}R$) are G_s-coupled P1 purinergic receptors which are largely localized in the rat hippocampus. This brain area is comprised of two distinct subfields that show different responses to hypoxic/ischemic brain injury: the CA1 region is particularly susceptible whereas the dentate gyrus (DG) is quite resistant. Our first aim was to determine the synaptic and proliferative response of the DG to severe oxygen and glucose deprivation (OGD) in acute rat hippocampal slices and to investigate the contribution of $A_{2A}R$ to recovery of synaptic activity after OGD. Field excitatory post-synaptic potentials (fEPSPs) in granule cells of the DG in brain slices prepared from male Wistar rats were recorded. A 9-min OGD is needed in the DG to always induce the appearance of anoxic depolarization (AD) and the irreversible block of synaptic activity, as recorded up to 24 h from the end of the insult, whereas only 7-min OGD is required in the CA1 region. Selective antagonism of $A_{2A}R$ by ZM241385 (100 nM, n=21) significantly prevents or delays the appearance of AD and protects from the irreversible block of neurotransmission induced by 9-min OGD in the DG. The effects of 9-min OGD on proliferation and maturation of cells localized in the subgranular zone of DG in slices prepared from 5-bromo-2'-deoxyuridine- (BrdU) treated rats was investigated. Slices were also incubated with an immature neuronal marker, doublecortin (DCX). The number of $BrdU^+$ cells was significantly decreased 6 h after 9-min OGD and this effect was antagonized by ZM241385 (n=8). After 24 h from the end of 9-min OGD, the number of $BrdU^+$ cells returned similar to that found before OGD and increased arborization of tertiary dendrites of DCX^+ cells was observed, indicating cell maturation toward neuronal phenotype.

Since $A_{2A}R$ antagonism is known to provide protection from ischemic damage and demyelination in *in vivo* model of cerebral ischemia (Melani *et al*., 2009), further aim of the work was to investigate the role of adenosine $A_{2A}R$ on oligodendrocyte maturation. Oligodendrocyte progenitor cells (OPCs) are a population of cycling cells which persist in the adult central nervous system, where, under opportune stimuli, they differentiate into mature myelinating oligodendrocytes. Oligodendrocytes express distinct voltage-gated ion channels depending on their maturation and express $A_{2A}R$. Experiments were run on purified primary OPCs prepared from rat cortex by patch-clamp whole cell recordings coupled with immunocytochemical labelling. Second aim was to study the role of $A_{2A}R$ on membrane currents and differentiation of OPCs. The selective $A_{2A}R$ agonist, CGS21680, inhibited sustained, delayed rectifier, K^+ currents (I_K) without modifying transient (IA) conductance. The effect was observed in all cells tested, independently from time in culture. CGS21680 inhibition of I_K current was concentration-dependent (10-200 nM) and blocked by the selective $A_{2A}R$ antagonist SCH58261 (100 nM). It is known that I_K currents play an important role during OPC development, since their block decreases cell proliferation and differentiation. In light of these data, our further aim was to investigate whether $A_{2A}R$ modulate these processes. CGS21680, applied at 100 nM in the culture medium of oligodendrocyte cultures, inhibited OPC differentiation (an effect prevented by SCH58261), without affecting cell proliferation.

Results demonstrate that cultured OPCs express functional $A_{2A}R$ whose activation negatively modulate I_K currents. We propose that, by this mechanism, $A_{2A}R$ inhibit OPC differentiation.

On the all, data suggest that the antagonism of $A_{2A}R$ is protective at early stages after ischemia by preventing synaptic failure and the decrease of cell proliferation and by directly stimulating OPC maturation.

Melani et al., (2009). *Brain*132:1480-95.