

027P ADENOSINE A_{2A} RECEPTORS ARE RESPONSIBLE FOR NECA-INDUCED VASODILATATION IN HUMAN ISOLATED MIDDLE CEREBRAL ARTERIES

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During periods of ischaemia, hypoxia, or enhanced oxygen demand, adenosine may mediate cerebral artery vasodilatation in humans. The adenosine receptor subtype responsible is believed to be of the A₂ class (Hardebo *et al.*, 1987), although it has not been determined whether high affinity A_{2A} or low affinity A_{2B} receptors are responsible. The aim of the present study was to determine which of these subtypes is responsible for the dilatation induced by the non-selective A₁/A₂ agonist 5'-(N-ethylcarboxamido)-adenosine (NECA).

Human cerebral arteries were obtained from 5 donors (2 female, 3 male, aged between 33 and 79) at *post mortem* (PM) with the informed consent of next of kin, and with the approval of local ethics committees. Tissue was placed in PBS at 4°C and transported overnight for use the following day, the mean (\pm s.e.m.) PM delay was 5.8 ± 0.6 hours. Rings of middle cerebral artery were mounted in 10ml organ baths containing gassed Krebs solution (95% O₂ / 5% CO₂) at 37°C under isometric conditions and an initial tension of 5mN. The integrity of the endothelium in each case was not established. After 60 minutes equilibration, a cumulative concentration-effect curve to the thromboxane A₂ mimetic, U46619, was constructed. After washing, the selective A_{2A} antagonist ZM241385 (Ongini *et al.*, 1999) or the selective A_{2B} antagonist MRS1754 (Ji *et al.*, 2001) was added at 100nM and left in contact with the tissues for 30 minutes. Vessels were constricted with an approximate EC₅₀ concentration of U46619 (10-100nM) and allowed to plateau, after which NECA (100pM – 300µM) was added by cumulative application. After addition of the final concentration of NECA, prostacyclin (1µM) was administered to induce a standard relaxation. The equation $(\log_{10}(\text{concentration ratio} - 1)) - (\log_{10}[\text{antagonist}])$ was used to estimate pA₂ values. Cumulative administration of NECA to pre-constricted arteries in

the absence of antagonist resulted in a concentration-dependant vasodilatation (pEC₅₀ 7.5 ± 0.1). The A_{2B} receptor antagonist MRS1754 produced no significant shift in the NECA concentration-response curve (pEC₅₀ 7.7 ± 0.3), whereas the A_{2A} antagonist ZM241385 produced a significant rightward shift (pEC₅₀ 5.5 ± 0.3 , pA₂ 8.7 ± 0.2 , Figure 1). The estimated pA₂ value for ZM241385 compares well with its published affinity (pKi=9.1) for human recombinant A_{2A} receptors (Ongini *et al.*, 1999).

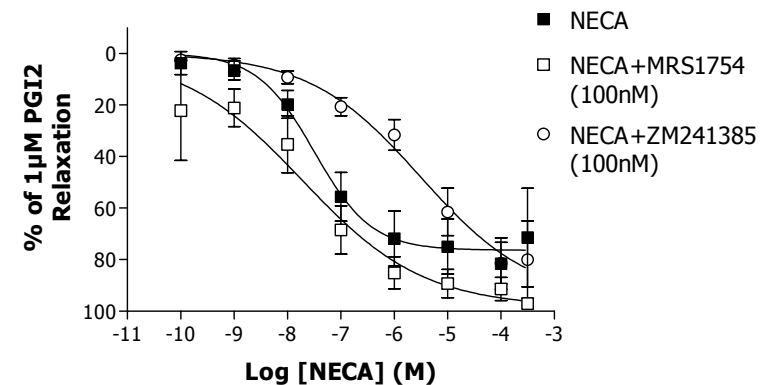


Figure 1. Concentration-response curves to NECA in U46619 pre-contracted human middle cerebral arteries in the absence and presence of A_{2R} antagonists. Data are expressed as a percentage of the relaxation caused by 1µM prostacyclin, mean \pm s.e.m. (n=4-5).

In conclusion, NECA-induced vasodilatation of human middle cerebral arteries appears to be mediated via adenosine A_{2A} receptors. Hardebo, J.E., Kahrstrom, J., Owman, C. (1987) *Eur. J. Pharmacol.*, **144**,343-52.

Ji, X., Kim, Y.C., Ahern, D.G., *et al* (2001) *Biochem. Pharmacol.*, **61**, 657-63.

Ongini, E., Dionisotti, S., Gessi, S., *et al* (1999) *Naunyn-Schmiedeberg's Archives of Pharmacology*. 359, 7-10.