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Short communication

Stimulation of adenosine A_1 receptors prevents the EEG arousal due to dopamine D_1 receptor activation in rabbits

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Abstract

The influence of adenosine A_1 (N^6-cyclopentyladenosine, CPA) and A_2 (2-[4-(2-carboxylethyl)phenethylamino]-5′-*N*-ethylcarboxamido-adenosine hydrochloride, CGS 21680) receptor agonists on SKF 38393-induced electroencephalographic (EEG) arousal was studied in rabbits. While CPA (0.1 mg/kg i.v.) significantly prevented the EEG effects of SKF 38393, CGS 21680 (0.2 mg/kg i.v.) did not affect them. These results demonstrate that adenosine A_1 receptors can modulate dopamine D_1 receptor-induced EEG arousal and show, for the first time, that adenosine-dopamine interactions are involved in brain functions other than motor activity.

Keywords: Dopamine D_1 receptor; Adenosine A_1 receptor; EEG (electroencephalographic) arousal; (Rabbit)

1. Introduction

Arousal is a state of activation of the central nervous system characterized by behavioural (increase in spontaneous motor activity, enhanced exploratory activity and increased response to stimuli) and electrocortical (appearance of low-amplitude, fast-frequency waves) features (Longo, 1962; Ongini and Longo, 1989; Steriade et al., 1993). The electrocortical features detected by electroencephalography (EEG) are considered a sensitive and suitable means to study arousal (Longo, 1962; Ongini and Longo, 1989; Rainnie et al., 1994). Even though EEG arousal is a very complex phenomenon, in which various ascending activating systems and several neurotransmitters are involved (Steriade et al., 1993), it has been shown that the administration of the dopamine D_1 receptor agonist SKF 38393 induces EEG arousal, thus suggesting a major role of central dopamine D_1 receptors in the induction of this activation state (Ongini, 1993; Ongini et al., 1985).

Central dopaminergic functions are known to be modulated by the adenosine system (Ferré et al., 1991b; Popoli et al., 1994), and powerful and specific interactions between adenosine and dopamine receptors have been reported. Besides the antagonistic effects exerted by adenosine A_2 receptors on dopamine D_2 receptors (Ferré et al., 1991a), a powerful inhibitory influence of adenosine A_1 receptors on striatal dopamine D_1 receptors has been recently reported (Ferré et al., 1994).

The aim of the present work was to test the possible influence of adenosine A_1 receptors on the EEG arousal induced by dopamine D_1 receptor stimulation in rabbits. The rabbit is the most suitable animal to be used for EEG arousal studies, as it always shows a synchronization-directed EEG pattern (Longo, 1962).

The following drugs were used: SKF 38393 (dopamine D_1 receptor agonist; Stoof and Kebabian, 1984); SCH 23390 (dopamine D_1 receptor antagonist; Iorio et al., 1983); N^6-cyclopentyladenosine (CPA, adenosine A_1 receptor agonist; Bruns et al., 1986); (2-[4-(2-carboxylethyl)phenethylamino]-5′-*N*-ethylcarboxamido-adenosine hydrochloride (CGS 21680, adenosine A_2 receptor agonist; Jarvis et al., 1989).

2. Materials and methods

2.1. Experimental procedure

Adult male rabbits (2.3–2.8 kg) were used. Screw cortical electrodes were implanted under anaesthesia and fixed with dental acrylic to the skull surface.

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All drugs were dissolved in saline and injected i.v. in a volume of 1 ml/kg. In the case of combined treatments, SKF 38393 was administered 10 min after the first drug. Groups of 6 rabbits each were randomly assigned to the following experimental groups: saline; SKF 38393 (10 mg/kg); SCH 23390 (0.03 mg/kg) + SKF 38393; CPA (0.02 and 0.1 mg/kg) + SKF 38393; CGS 21680 (0.2 mg/kg) + SKF 38393. In separate experiments, the effects of CPA (0.1 mg/kg) and CGS 21680 (0.2 mg/kg) administered alone were tested in 4 animals/group.

2.2. EEG analysis

The EEG was recorded by an OTE Biomedical apparatus (model E 10b poligraph) and simultaneously registered by a computer (IBM PS/2, model 70 386). Sequential power spectra of 20-s EEG epochs (1 epoch every min) were produced by Fast Fourier Transform (FFT) with frequency resolution of 0.35 Hz (software developed by Enrico Staderini, MD). Each power spectrum for a 20-s epoch was the mean spectrum resulting from the single 2-s power spectra, overlapped by 50%, of the epoch. All the power spectra relevant to an EEG tracing were recorded on an optical disk (940 MB, RPS) and then analyzed to calculate the relevant power of each frequency band. The EEG activity was recorded at the level of the frontal cortex (A = +3 to 4 mm, L = ±1.5 to 2 mm from bregma and sagittal suture, respectively), and the following frequency bands were considered: 0–4 Hz (band 1); 4–8 Hz (band 2); 8–12 Hz (band 3); 12–16 Hz (band 4); 16–30 Hz

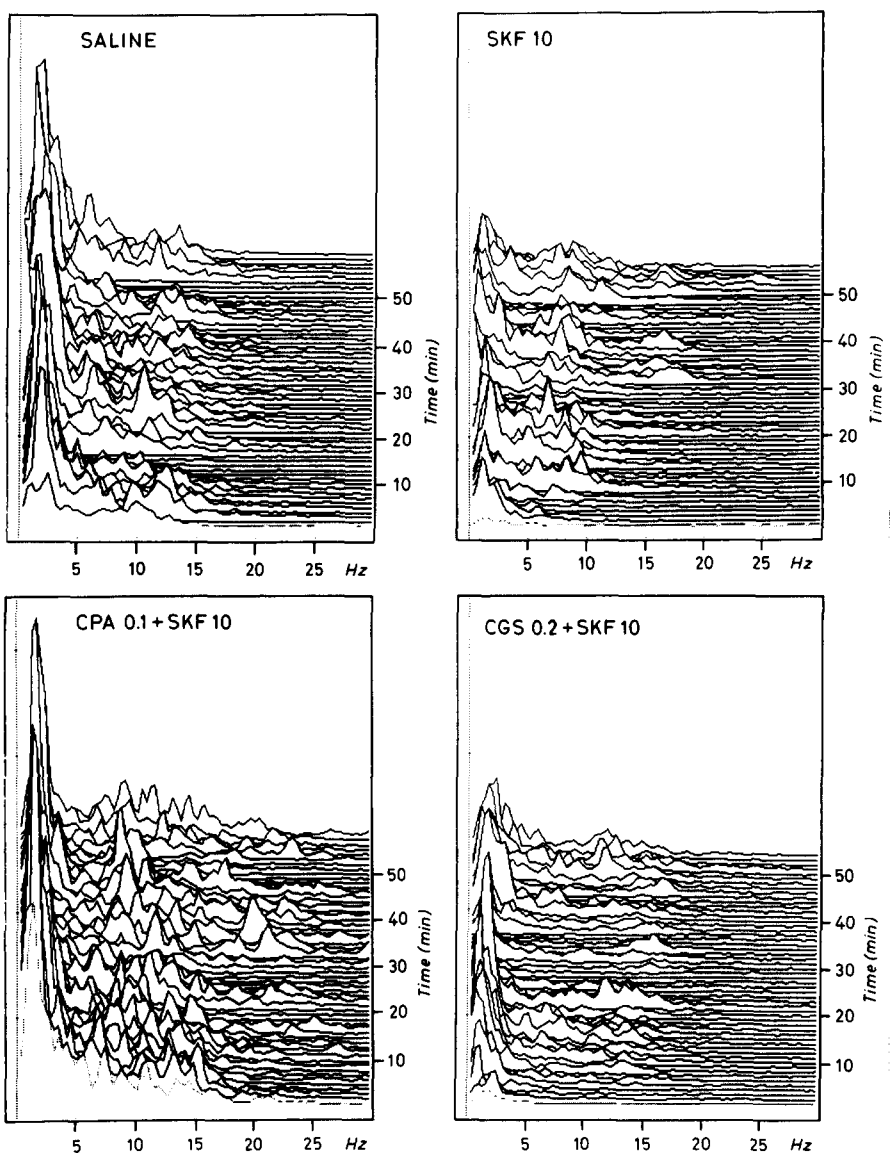

Fig. 1. Examples of sequential EEG power spectra recorded at the level of rabbit frontal cortex. The figure shows some representative sequential EEG power spectra from rabbits treated with saline, SKF 38393 10 mg/kg, CPA 0.1 mg/kg or CGS 21680 0.2 mg/kg + SKF 38393. While CPA counteracted the EEG effects of SKF (i.e. marked reduction of power in the slow-frequency bands), CGS 21680 was unable to affect them.

(band 5). The total power, in μV^2, was expressed as the sum of the power of each of the above frequency bands.

One-way analysis of variance (ANOVA) followed by Dunnett's test was used for the statistical analysis of the results.

3. Results

As previously reported by Ongini et al. (1985), the i.v. administration of SKF 38393 in rabbits induced an EEG picture characterized by low-amplitude, fast-frequency waves ('desynchronization'). In terms of quantitative EEG analysis, this effect is reflected both in a marked reduction of the total power, and in a different distribution of the power in the various frequency bands (Figs. 1 and 2). In particular, SKF 38393 induced a significant reduction of power in the 0–4 Hz band and a significant increase of power in the range of fast frequencies (Fig. 2).

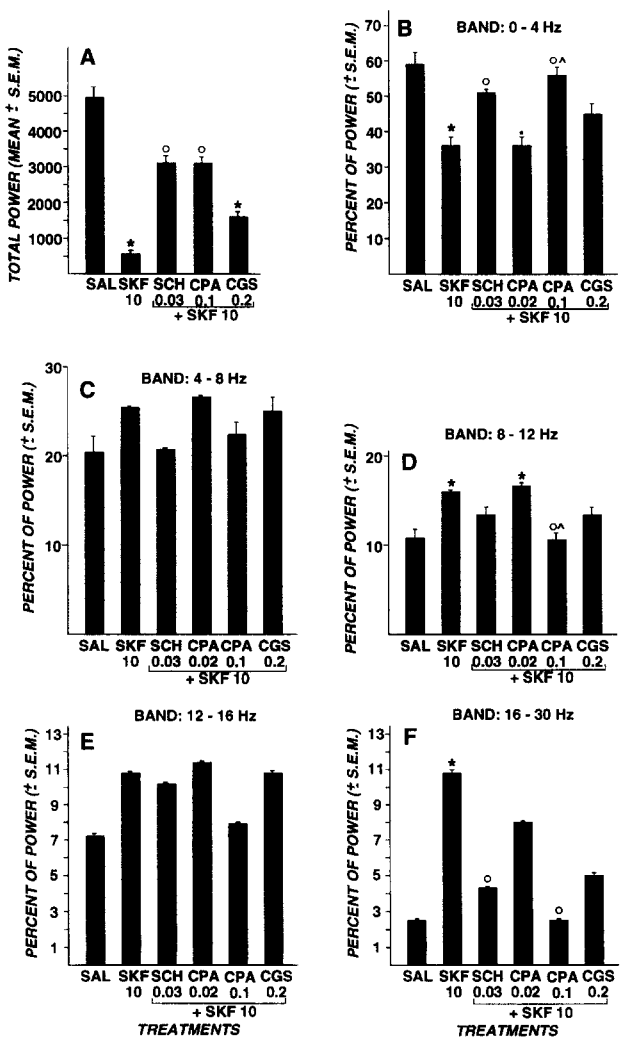

Fig. 2. Influence of SCH 23390, CPA and CGS 21680 on the effects induced by SKF 38393 on rabbit EEG (total power and relative power distribution). SKF 38393 induced a significant reduction of mean total EEG power (A) and of the percentage of power in the 0–4 Hz band (B). Conversely, a significantly increased power was seen in bands 8–12 (D) and 16–30 (F) Hz. The effects of SKF were significantly prevented by both SCH 23390 and CPA, but not by CGS 21680. Doses are expressed as mg/kg i.v. Each group was composed of 6 animals. Significantly different ($P < 0.01$): * vs. saline; ° vs. SKF 38393; ^vs. CPA 0.02 mg/kg (one-way ANOVA followed by Dunnett's test).

The dopamine D_1 receptor antagonist SCH 23390 and the adenosine A_1 receptor agonist CPA exerted comparable antagonistic effects on SKF 38393-induced EEG activation (Figs. 1 and 2). Conversely, the adenosine A_2 receptor agonist CGS 21680 did not significantly modify the EEG effects of SKF 38393 (Figs. 1 and 2).

Administered alone at the same doses used in the experiments with SKF 38393, neither CPA nor CGS 21680 induced significant EEG effects with respect to control animals (relative power distribution = band 1: CPA = 60.4 ± 3.2, CGS 21680 = 54.3 ± 4.2; band 2: CPA = 17.4 ± 1.4, CGS 21680 = 20.7 ± 2.5; band 3: CPA = 9.1 ± 1.1, CGS 21680 = 10.5 ± 1.6; band 4: CPA = 10.1 ± 1.5, CGS 21680 = 10.2 ± 0.25; band 5: CPA = 2.5 ± 0.3, CGS 21680 = 3.9 ± 0.5, NS vs. saline; mean total power: CPA = 5300 ± 285 μV^2; CGS 21680 = 5020 ± 260 μV^2, NS vs. saline).

4. Discussion

The involvement of dopamine D_1 receptors in the induction of EEG arousal is confirmed by the findings that (i) the dopamine D_1 receptor agonist SKF 38393 induced a desynchronized EEG pattern characterized by significant changes in both total EEG power and relative power distribution. Even though SKF 38393 is a partial agonist at dopamine D_1 receptors, it has been reported to induce typical D_1-dependent EEG and behavioural effects as compared with the full agonist A68930 (Trampus et al., 1993); (ii) the effects of SKF 38393 were prevented by the dopamine D_1 receptor antagonist SCH 23390.

Dopamine D_1 receptor-induced EEG activation may therefore be considered a suitable functional model to test the existence of A_1–D_1 interactions.

The present data show that the EEG arousal induced by dopamine D_1 receptor activation is significantly prevented by the stimulation of adenosine A_1, but not A_{2a}, receptors. Adenosine A_1 receptor agonists by themselves induced a marked slowing of the rabbit EEG at high doses (Popoli et al., 1988). Nevertheless, in the present experiments CPA significantly counteracted the EEG effects of SKF 38393 at doses which did not influence the EEG tracing per se. Thus, this action cannot be ascribed to the sedative effects of the drug.

The present finding of an inhibitory influence of CPA on SKF 38393-induced EEG arousal agrees with a previous report from our group showing that A_1 receptor stimulation decreased D_1-induced motor activation in mice and rabbits and produced an uncoupling of D_1 receptors from

the G-protein in rat striatal membranes (Ferré et al., 1994). The present results thus confirm the existence of an interaction between adenosine A_1 and dopamine D_1 receptors and show, for the first time, that adenosine-dopamine interactions are involved in brain functions other than motor activity.

Several reports indicate that adenosine A_1 receptors mediate the sedative and hypnotic effects of adenosine, while their blockade is likely to mediate the psychostimulant effects of adenosine antagonists, like caffeine (Chagoya de Sànchez et al., 1993; Durcan and Morgan, 1990; Radulovacki et al., 1984; Yanik and Radulovacki, 1987). Recently, adenosine A_1 receptors have been reported to mediate the inhibitory influence of adenosine on mesopontine cholinergic neurons and, as a consequence, on EEG arousal (Rainnie et al., 1994). Our findings suggest an additional or integrative mechanism by which adenosine A_1 receptors affect EEG arousal, namely by an antagonistic A_1/D_1 receptor interaction.

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References

Bruns, R.F., G.H. Lu and T.A. Pugsley, 1986, Characterization of the A_2 adenosine receptor labeled by [H^3]NECA in rat striatal membranes, Mol. Pharmacol. 29, 331.

Chagoya de Sànchez, V., R. Hernàndez Mùñoz, J. Suàrez, S. Vidrio, L. Yàñez and M. Diàz Mùñoz, 1993, Day-night variations of adenosine and its metabolizing enzymes in the brain cortex of the rat – possible physiological significance ofor the energetic homeostasis and the sleep-wake cycle, Brain Res. 612, 115.

Durcan, M.J. and P.F. Morgan, 1990, Prospective role for adenosine and adenosinergic systems in psychiatric disorders, Psychol. Med. 20, 475.

Ferré, S., G. Von Euler, B. Johansson, B.B. Fredholm and K. Fuxe, 1991a, 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.

Ferré, S., K. Fuxe, G. Von Euler, B. Johansson and B.B. Fredholm, 1991b, Adenosine-dopamine interactions in the brain, Neuroscience 51, 501.

Ferré, S., P. Popoli, L. Giménez-Llort, U.-B. Finnman, E. Martinez, A. Scotti de Carolis and K. Fuxe, 1994, Postsynaptic antagonistic interaction between adenosine A_1 and dopamine D_1 receptors, NeuroReport 6, 73.

Iorio, L.C., A. Barnett, F.H. Leitz, V.P. Houser and C.A. Korduba, 1983, SCH 23390, a potential benzazepine antipsychotic with unique interactions on dopaminergic systems, J. Pharmacol. Exp. Ther. 226, 462.

Jarvis, M.F., R. Schulz, A.J. Hutchison, U.H. Do, M.A. Sills and M. Williams, 1989, [^{3}H]CGS 21680, a selective A_2 adenosine receptor agonist direclty labels A_2 receptors in rat brain, J. Pharmacol. Exp. Ther. 251, 888.

Longo, V.G., 1962, Electroencephalographic Atlas for Pharmacological Research, Effects of Drugs on the Electrical Activity of Rabbit Brain (Elsevier, Amsterdam).

Ongini, E., 1993, Role of D_1 versus D_2 receptors in the modulation of states of arousal and sleep, in: Dopamine Receptor Interactions, ed. J. Waddington (Academic Press, London) p. 175.

Ongini, E. and V.G. Longo, 1989, Dopamine receptor subtypes and arousal, Int. Rev. Neurobiol. 31, 239.

Ongini, E., M.G. Caporali and M. Massotti, 1985, Stimulation of dopamine D_1 receptors by SKF 38393 induces EEG desynchronization and behavioural arousal, Life Sci. 37, 2327.

Popoli, P., M. Benedetti and A. Scotti de Carolis, 1988, Anticonvulsant activity of carbamazepine and N^6-L-phenylisopropyladenosine in rabbits. Relationship to adenosine receptors in central nervous system, Pharmacol. Biochem. Behav. 29, 533.

Popoli, P., A. Pèzzola and A. Scotti de Carolis, 1994, Modulation of striatal adenosine A_1 and A_2 receptors induces rotational behaviour in response to dopaminergic stimulation in intact rats, Eur. J. Pharmacol. 257, 21.

Radulovacki, M., R.M. Virus, M. Djuricic-Nedelson and R.D. Green, 1984, Adenosine analogs and sleep in rats, J. Pharmacol. Exp. Ther. 228, 268.

Rainnie, D.G., H.C.R. Grunze, R.W. McCarley and R.W. Greene, 1994, Adenosine inhibition of mesopontine cholinergic neurons: implications for EEG arousal, Science 263, 689.

Steriade, M., D.A. McCormick and T.J. Sejnowski, 1993, Thalamocortical oscillations in the sleeping and aroused brain, Science 262, 679.

Stoof, J.C. and J.M. Kebabian, 1984, Two dopamine receptors: biochemistry, physiology and pharmacology, Life Sci. 335, 2281.

Trampus, M., N. Ferri, M. Adami and E. Ongini, 1993, The dopamine D_1 receptor agonists, A68930 and SKF 38393, induce arousal and suppress REM sleep in the rat, Eur. J. Pharmacol. 235, 83.

Yanik, G. and M. Radulovacki, 1987, REM sleep deprivation up-regulates adenosine A_1 receptors, Brain Res. 402, 362.