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Research Report

Effect of caffeine on the expression of cytochrome P450 1A2, adenosine A_{2A} receptor and dopamine transporter in control and 1-methyl 4-phenyl 1, 2, 3, 6-tetrahydropyridine treated mouse striatum

Seema Singh[a,1], *Kavita Singh*[a,1], *Satya Prakash Gupta*[a], *Devendra Kumar Patel*[a], *Vinod Kumar Singh*[b], *Raj Kumar Singh*[b], *Mahendra Pratap Singh*[a,*]

[a]*Indian Institute of Toxicology Research, (Council of Scientific and Industrial Research), Mahatma Gandhi Marg, Post Box-80, Lucknow-226 001, India*

[b]*Chhatrapati Shahuji Maharaj Medical University, Lucknow-226 003, India*

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ABSTRACT

Parkinson's disease (PD) is a progressive neurodegenerative disorder, characterized by the selective loss of dopaminergic neurons of the nigrostriatal pathway. Epidemiological studies have shown an inverse relationship between coffee consumption and susceptibility to PD. Cytochrome P450 1A2 (CYP1A2) is involved in caffeine metabolism and its clearance. Caffeine, on the other hand, antagonizes adenosine A_{2A} receptor and regulates dopamine signaling through dopamine transporter (DAT). The present study was undertaken to investigate the expression of CYP1A2, adenosine A_{2A} receptor and DAT in mouse striatum and to assess their levels in 1-methyl 4-phenyl 1, 2, 3, 6-tetrahydropryridine (MPTP) treated mouse striatum with and without caffeine treatment. The animals were treated intraperitoneally daily with caffeine (20 mg/kg) for 8 weeks, followed by MPTP (20 mg/kg) +caffeine (20 mg/kg) for 4 weeks or vice versa, along with respective controls. Tyrosine hydroxylase immunoreactivity, levels of dopamine and 1-methyl 4-phenylpyridinium ion (MPP^+), expressions of CYP1A2, adenosine A_{2A} receptor and DAT and CYP1A2 catalytic activity were measured in control and treated mouse brain. Caffeine partially protected MPTP-induced neurodegenerative changes and modulated MPTP-mediated alterations in the expression and catalytic activity of CYP1A2, expression of adenosine A_{2A} receptor and DAT. The results demonstrate that caffeine alters the striatal CYP1A2, adenosine A_{2A} receptor and DAT expressions in mice exposed to MPTP.

1. Introduction

The neurodegeneration in Parkinson's disease (PD) targets dopaminergic neurons of the nigrostriatal pathway. Dopaminergic neurodegeneration causes dopamine depletion in the striatum leading to motor disturbances and onset of resting tremor, rigidity, postural instability and bradykinesia, the major hallmarks of PD. Aging, genetic factors and environ-

* *Corresponding author*. Fax: +91 522 2628227.
E-mail address: singhmahendrapratap@rediffmail.com (M.P. Singh).
[1] Equal contribution.

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mental exposure to pesticides and heavy metals are the major contributors to PD (Tanner et al., 1999; Vieregge et al., 1999; Wirdefeldt et al., 2004). 1-methyl 4-phenyl 1, 2, 3, 6 tetrahydropyridine (MPTP) causes selective degeneration of dopaminergic neurons and is regarded as one of the best rodent models to understand the biochemical and molecular events leading to PD and also to assess the efficacy of anti-PD drugs. MPTP produces several biochemical, molecular and phenotypic symptoms that mimic the sporadic PD (Przedborski et al., 2000; Gerlach and Riederer, 1996). Current therapies alleviate the symptoms of PD and offer only symptomatic relief to the patients. Despite delaying neurodegeneration, the available therapies exert several adverse effects and therefore attempts are consistently being made to develop better therapies with minimal side effects.

Caffeine is one of the most widely consumed psychostimulants and dietary components, with an average consumption of about 200–250 mg/day/person, as a standard cup of coffee contains 100 mg of caffeine (Fredholm, 2004). Both retrospective and prospective epidemiological studies have linked caffeine consumption to reduced PD risk and postulated that the neuroprotective effect of caffeine could be due to its ability to antagonize adenosine A_{2A} receptor (Ascherio et al., 2001). Caffeine (5–30 mg/kg) exposure to animals corresponding to the typical human exposure also resists MPTP-induced dopamine depletion in a dose dependent manner (Chen et al., 2001). Although caffeine non-selectively antagonizes adenosine A_1, A_{2A}, A_{2B} and A_3 receptors, only A_{2A} receptor is abundant in the striatum (Svenningsson and Fredholm, 2003). Adenosine A_{2A} receptor modulates γ-amino butyric acid, acetylcholine and glutamate-mediated neurotransmissions (Kurokawa et al., 1996; Mori et al., 1996; Richardson et al., 1997; Ochi et al., 2000; Fuxe et al., 1998; Canals et al., 2003; Ciruela et al., 2004). Caffeine-mediated neuroprotection could be partially contributed by its ability to inhibit the blood brain barrier dysfunction in MPTP-treated mouse (Chen et al., 2008). Caffeine also prevents apoptotic cell death by the activation of phosphoinositide 3-kinase or serine-threonine protein kinase and Akt pathways (Nakaso et al., 2008). As caffeine alters dopamine signaling and receptor affinity in the striatum by stimulating dopaminergic responses, therefore, blockade of adenosine A_{2A} receptors probably direct anti-PD effects (Ferre et al., 1992). Caffeine stimulates potassium channel opening, which prevents neurons from depolarization and neurotransmitter release and inhibits excitotoxicity by altering neuronal metabolism (Mao et al., 2007; Avshalumov et al., 2005; Jones, 2008). Caffeine is metabolized mainly by CYP1A2 in the liver and increased CYP1A2 activity is associated with habitual caffeine consumption (Carrillo and Benitez, 1996). Dopamine transporter (DAT) is expressed in the dopaminergic neurons and clears the dopamine released into the extra-cellular spaces, thereby regulating the dopamine signaling (Kurosaki et al., 2003). Although CYP1A2, adenosine A_{2A} receptor and dopamine transporter are critical in PD, their roles in MPTP-mediated dopaminergic neurodegeneration and caffeine-mediated neuroprotection are not established. The present study was performed to investigate the expression of CYP1A2, adenosine A_{2A} receptor and dopamine transporter in mouse striatum and to assess their roles therein.

2. Results

2.1. Dopamine content

MPTP treatment for 2–4 weeks produced a significant reduction in the striatal dopamine content (Fig. 1a). The decrease in dopamine level was significantly less both in caffeine pre-and post-treated animals. Treatment with ciprofloxacin, an inhibitor of CYP1A2, enhanced MPTP-mediated depletion of dopamine contents in the pre-treatment group (Fig. 1b).

2.2. Dopaminergic neurodegeneration

Caffeine treatment did not produce any change in tyrosine hydroxylase (TH) immunoreactivity; however, MPTP

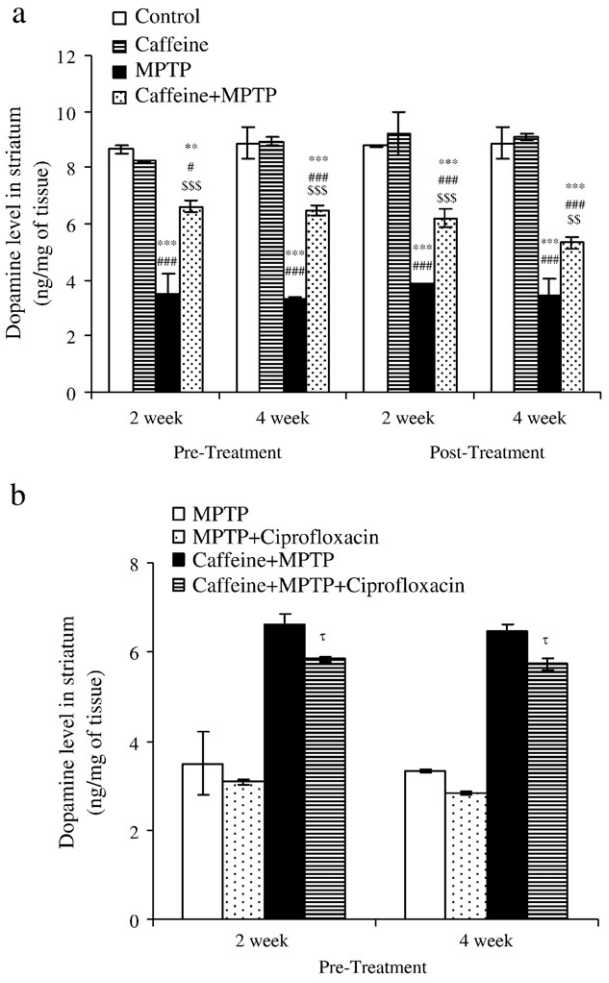

Fig. 1 – Dopamine content in the striatum (ng/mg of tissue) of caffeine treated animals (a) and effect of CYP1A2 inhibitor ciprofloxacin on dopamine content in the pre-treatment group (b). The data are expressed as means ± S.E.M. (n = 3–5 separate experiments). Significant changes $^{**}(P<0.01)$, $^{***}(P<0.001)$ are expressed in comparison with control, #($P<0.05$), ###($P<0.001$) with caffeine, $$($P<0.01$), $$$($P<0.001$) with MPTP and τ($P<0.05$) with caffeine + MPTP.