HYPOPHAGIC AND HYPOLOCOMOTIVE EFFECTS OF METACHLORO PHENYL PIPERAZINE IN RATS TREATED WITH THEOPHYLLINE AND CAFFEINE

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ABSTRACT

Long term intake of coffee is known to produce anxiety and suppression of appetite. 5- hydroxytryptamine (5-HT) acting via 5-HT-2C receptors elicits anorexia and anxiety. The present study is design to monitor metachloro phenyl piperazine (m-CPP) at a dose of 3mg/ml/kg, induces hypophagia and hypolocomotion in rats taking a solution of caffeine (a component of coffee and tea) or theophylline (a component of tea) as a sole source of water. We found that hypophagic and hypolocomotive effects of m-CPP were attenuated in theophylline but not in caffeine treated animals suggesting that long term intake of theophylline may attenuate anorexiogenic and anxiogenic effects of 5-HT. A possible role of 5-HT-2C receptors in the modulation of anxiety and appetite in people drinking coffee or tea discussed.

Keywords: Anxiety, anorexia, m-CPP, caffeine, theophylline.

INTRODUCTION

Caffeine and theophylline are the central nervous system stimulants, which are not categorized as the drug of abuse. (Gasior *et al*, 2002) These are most commonly used psychoactive drugs present in coffee and tea that is available as social drinks. (Uysal *et al*., 2009) The intake is known to increase alertness, work performance and delay the onset of fatigue (Heatherley *et al*., 2005).

This study is designed to investigate the effects of caffeine and theophylline on food intake and activity of rats to compare the stimulation and appetite suppressant effects of the two stimulants. Anxiety and anorexia are the two adverse effects reported in excessive consumer of coffee (White *et al*., 1980). Serotonergic system is involved in the regulation of appetite and energy balance (Curzon, 1990; Haleem, 1996; Leibowitz, 1986; Tecott, 2007; Bickerdike 2003). Anxious and obsessional behavior and impulse control (Barr,Goodman, Price, McDoule, and Charney 1992; Higley and Linnoila, 1997; Lucki, 1998; Mann 1999)

In view of role of 5-HT-2C receptor in anxiety and anorexia it was assumed that long term intake of caffeine and theophylline may increase serotonin function by 5-HT-2C receptors to produce adverse effect on appetite and mood. The present study was designed to monitor the responsiveness of 5-HT2C receptor after long-term administration of caffeine and theophylline in rats.

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MATERIALS AND METHODS

Animals

Locally bred albino Wister rats having weight 180-220 gm purchased from HEJ Research Institute of Chemistry, University of Karachi, Pakistan were used in the study. These rats were housed with free access to the standard rodent diet cubes and tap water.

Drugs

Caffeine and theophylline were used in the treatment. The groups of rat treated with caffeine and theophylline were given 5 mg/100 ml caffeine and theophylline dissolved in water respectively. m-CPP was injected intraperitoneally at a dose of 3 mg/ kg body weight.

Experimental protocol

36 rats were divided into three groups:

1) Caffeine treated, (caffeine dissolved in drinking water)
2) Theophylline treated (theophylline dissolved in drinking water) and
3) Water treated rats (tap water).

The animal received respective treatment for 5 weeks. For monitoring the effects of m-CPP water, caffeine and theophylline treated rats were further divided into two groups, saline and m-CPP injected groups. Activity was monitored in open field after 30 min of m-CPP and saline injection. Hypophagic effects of m-CPP were monitored as 2-h and 4-h food intake starting after the injection of saline and m-CPP.

Behavioral study

Open field activity

Open field apparatus was used to monitor activity in a novel environment. The open field apparatus used in this study consisted of a square area 76 × 76 with walls 42 cm high. The floor of apparatus was divided by lines into 25 equal squares. To determine open field activity a rat was placed in the center square on the floor of the apparatus, numbers of the squares crossed with all four paws were scored for five minutes as described earlier (Haleem, 1996).

RESULT

Fig. 1 depicts the effects of m-CPP on open field activity in water treated, caffeine treated and theophylline treated rats. In fig I(b) data analyzed by Mann-Whitney U test show that m-CPP significantly (P<0.01) decrease number of square crossing in water treated, caffeine treated as well as water treated rats. In fig.1a data analyses by Mann-Whitney U test show that m-CPP significantly increase latency in caffeine treated (P<0.05) and in theophylline treated (P<0.01) rats. Increases in water treated rats were not significant. Theophylline treated m-CPP injected rat exhibit greater latency to move than water treated m-CPP injected rats.

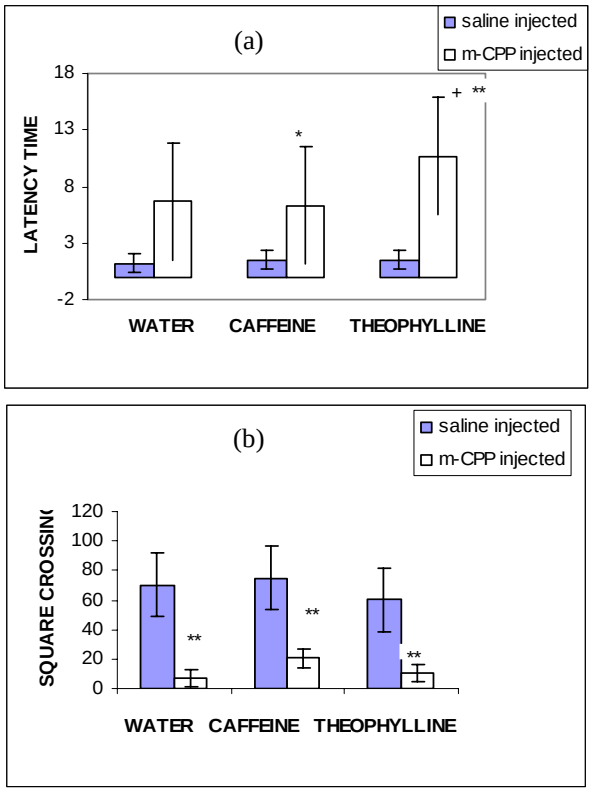

Fig. 1: Effect of m-CPP 3mg/kg on (a) latency time and (b) square crossing of water treated, caffeine treated and theophylline treated rats values are means± S.D, n=6.
Significant differences by Mann-Whitney U-test.
*P<0.05,**P<0.01 from similarly treated saline injected rats.
+P<0.05 from similarly injected water treated rats.

Fig. 2 shows effect of m-CPP on food intake measured 2 hr and 4 hr post injections in water, caffeine and theophylline treated rats. Mann-Whitney U test that administration of m-CPP decreased 2 hrs and 4 hrs food intake in water caffeine and theophylline treated animals. Caffeine and theophylline treated animals injected with saline also exhibit smaller 2 hrs and 4 hrs food intake than water treated saline injected animals. m-CPP induced decreases in 4 hrs but not 2 hrs food intakes were smaller in theophylline then water or caffeine treated animals.

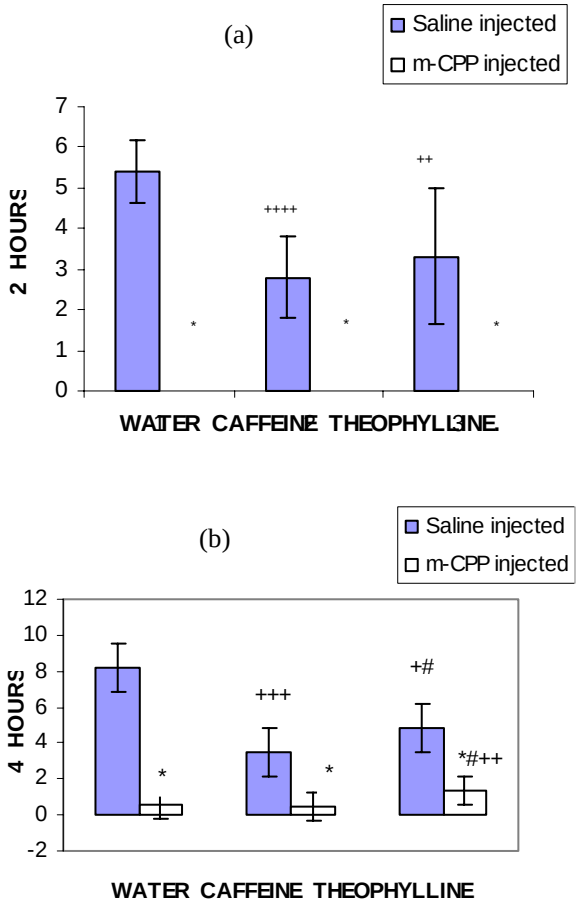

Fig. 2: Effect of m-CPP on food intake measured 2 hr. & 4 hr post injection in water caffeine and theophylline treated rats. Values are means ± S.D, n=6
Significant differences by Mann-Whitney U-test
*P,0.001 from similarly treated saline injected rats.
+P<0.05,++P<0.025,+++P<0.01 & ++++P<0.001 from similarly injected water treated rats.
#P<0.025 from caffeine treated similarly injected rats.

DISCUSSION

The present results demonstrate that m-CPP which is a selective 5-HT 2 C agonist (Murrphy *et al*., 1993; Kennet *et al*., 1993) and produces hypophagia (Samanin *et al*., 1979; Schuhler *et al*., 2005) and hypolocomotion (Lucki *et al*. 1989; Gleason and Shannon, 1998) via 5-HT2C

Table 1: Open-field activity

Parameter		Water treated rats		Caffeine treated rats		Theophylline treated rats	
		Saline	m-CPP	Saline	m-CPP	Saline	m-CPP
Latency Time	Mean	1.2	6.66	1.5	6.34*	1.5	10.7^{+**}
	S.D	±0.4	±3.75	±0.84	±4.8	±0.84	±5.2
Square Crossing	Mean	70.3	6.84**	75	20.6**	60.4	10.5**
	S.D	±20.46	±3.48	±25.5	±6.6	±21.6	±6.25

Values are means ± S.D, n=6, Significant differences by Mann-Whitney U-test
*P<0.05, **P<0.01 from similarly treated saline injected rats. +P<0.05 from similarly injected water treated rats.

Table 2: Effect of m-CPP 3mg/kg on food intake measured 2hr and 4hr post-injection.

Parameter		Water treated rats		Caffeine treated rats		Theophylline treated rats	
		Saline	m-CPP	Saline	m-CPP	Saline	m-CPP
2 Hours	Mean	5.25	0*	2.8^{++++}	0*	3.32^{++}	0*
	S.D	±0.78	±0.0	±1.02	±0.0	±1.699	±0.0
4 Hours	Mean	8.1	0.615*	3.5^{+++}	0.47*	$4.85^{+\#}$	$1.34^{++\#*}$
	S.D	±4.16	±0.307	±1.66	±0.22	±1.32	±0.811

receptors located postsynatically (Gibson *et al*., 1996) produced greater hypolocomotive and hypophagic effects in theophylline treated animals. These effects of m-CPP in caffeine treated animals were comparable to water treated group.

The aim of the present study was to determine the 5-HT2C receptors responsiveness in rats after long term administration of caffeine and theophylline. The results are consistent that long term theophylline administration down regulates post synaptic 5-HT-2C receptors dependent functions. While long term caffeine administration doesn't alter 5-HT-2C receptor dependent functions.

The arylpiperazie compound m-CPP is currently the agonist of choice to examine the functions of 5 HT2C receptors (Murphy *et al*., 1991; Kennett, 1993; Baxter *et al*., 1995). The drug increases 5-HT release (Hikiji *et al*., 2004) via stimulation of postsynaptic 5 HT2C receptors (Gibson *et al*., 1996).

It is well established that activation of the serotonergic system results in the suppression of feeding behavior (Bickerdike, 2003; Haleem, 1996). The hypothalamus is the region of the brain known to have role in the regulation of appetite (Hikiji *et al*., 2004). The 5-HT-2C receptor subtype has also been implicated extensively in the regulation of ingestive behavior. Researches show that increase in the functional activity of 5-HT at post synaptic 5-HT-2C sites elicited anxiety in experimental animals (Kitchener *et al*., 1994). In the present work, administration of m-CPP decreased food intake 2 and 4 h post-injection (fig. 2). Other authors have also reported decreased food intake (hypophagia) following the administration of m-CPP (Fone *et al*., 1998; Hewitt *et al*., 2002).

Studies have shown that acute administration of m-CPP induce hypolocomotive, hypophagic and anxiogenic effects (Samanin *et al*., 1979; Kennett and Curzon, 1988; Kennett *et al*., 1997; Sills *et al*., 1985; Freo *et al*., 1993; Kennett *et al*., 1995; Fone, *et al*., 1996), that is consistent with our present findings (figs.1-2). In addition, important finding of the present study is that hypophagic as well as anxiogenic – like behavior of m-CPP were smaller in theophylline than water and caffeine treated rats. Possibly by development of postsynatic subsensitivity (Lucki *et al*., 1989). It is suggested that increase in availability of 5-HT at post synaptic site may be involved in the down regulation or subsensitization of 5-HT-2C receptor dependent responses observed in the present study (fig. 2).

In conclusion the present study show that the anxiogenic and anorexiogenic effect of coffee are not produced via changes in 5-HT-2C receptor function. It is how ever possible that availability of 5-HT at the receptor sites is increased to produce the reported anxiogenic and anorexiogenic effects. The present results also tends to show that theophylline may attenuate 5-HT-2C receptor dependent anorexia and anxiety suggesting that tea which contains both caffeine and theophylline may not produce adverse effect on appetite and mood.

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