SEROTONIN$_{1A}$ RECEPTOR AGONISM IN THE EXPRESSION OF BEHAVIORAL DOPAMINERGIC SUPERSENSITIVITY IN SUBCHRONIC HALOPERIDOL TREATED RATS

FARHAT BATOOL* AND DARAKHSHAN J. HALEEM

Neurochemistry and Biochemical Neuropharmacology Research Laboratory, Department of Biochemistry, University of Karachi, Karachi-75270, Pakistan

ABSTRACT

The idea that serotonin (5-hydroxytryptamine; 5-HT) is contributed in schizophrenia has long been advocated and alterations in 5-HT neurotransmission has been hypothesized to modulate both the therapeutic and extrapyramidal symptoms (EPS) liability of conventional neuroleptics. The 8-hydroxy-2-(di-n-propylamino) tetralin (8-OH-DPAT), a preferential 5-HT$_{1A}$ ligand, has been reported to attenuate EPS functions of haloperidol in animals. In view of a possible role of 5-HT$_{1A}$ receptors in the management of EPS functions of a neuroleptic drug, the present study was designed to investigate behavioral responses of 8-OH-DPAT at a challenge dose of 0.5mg/kg in rats with subchronic haloperidol administration at a dose of 5mg/kg twice daily for 5 days. The intensity of 5-HT syndrome provoked by 8-OH-DPAT was taken as a measure of postsynaptic responses. In the present study administration of haloperidol at a dose of 5mg/kg twice daily for 5 days decreased locomotion significantly ($p<0.01$) in familiar (home cage) environment. Subchronic administration of haloperidol at the same dose elicited significant ($p<0.01$) cataleptic responses in rats when compared with saline treated rats. Results revealed that 8-OH-DPAT-induced hyperlocomotion ($p<0.05$) and forepaw treading ($p<0.1$) were significantly smaller in rats pre-treated with haloperidol for 5 days than repeatedly saline injected rats. Conversely, the other components of the syndrome i.e. flat body posture ($p<0.001$), hind limb abduction ($p<0.001$) and straub tail ($p<0.01$) were significantly greater in repeated haloperidol treated rats when compared with repeated saline injected rats. These findings help to demonstrate a causal link between the upregulation of DA-D$_2$ receptors and the decrease in the effectiveness of presynaptic 5-HT$_{1A}$ receptors following subchronic haloperidol administration and this may further help to yield an antipsychotic agent with an improved profile of efficacy to EPS, thereby widening its therapeutic window.

Keywords: EPS, haloperidol, 8-OH-DPAT, 5-HT$_{1A}$ receptors, 5-HT syndrome, schizophrenia.

INTRODUCTION

In the brain research, schizophrenia is a major therapeutic challenge of modern medicine, and characterized by the impairment of cognitive functions, delusions, hallucinations, and social withdrawal (Kane and Marder, 1993; Dixon *et al*., 1995; Kane, 1996; Huber and Gross, 1997; Miyamoto *et al*., 2005). Although, conventional neuroleptics control positive symptoms in schizophrenia, they also induce extrapyramidal side effects (EPS) such as dsystonia, Parkinsonism, akathisia and tardive dyskinesia (Baldessarini, 1985; Casey, 1991). These effects are mediated by blockade of dopamine (DA)-D$_2$ receptors, but their expression can be modulated by other systems, most notably, serotonin (5-hydroxytryptamine; 5-HT) (Casey, 1993). In the recent years, there has been considerable renewal of interest in the contribution of serotonergic system in the treatment of schizophrenia (Prinssen *et al*., 1999). 5-HT$_{1A}$ receptors implicated in the treatment of schizophrenia are located presynaptically in the raphe nuclei (fig. 1). Administration of 5-HT agonists to the animals elicits a variety of behavioral effects manifested by an increase in the activity of an animal (Yamada *et al*., 1989). The behavioral effects are often described as hyperactivity syndrome and various components of the syndrome include forepaw treading, head weaving, flat body posture, hind limb abduction and straub tail (Smith and Peroutka, 1986). The syndrome is elicited by the binding of 5-HT agonists with the presynaptic receptors (see fig. 1). Reported studies have shown that postsynaptic 5-HT$_{1A}$ receptors are implicated in the 5-HT syndrome elicitation, particularly forepaw treading (Tricklebank *et al*., 1984; Goodwin and Green, 1985). Syndrome can be induced in rats by treatment that increase synaptic levels of 5-HT (Yamada *et al*., 1998) or by some drugs that directly stimulate 5-HT$_{1A}$ receptors (Bantick *et al*., 2004) in the brain. It is also reported that 8-OH-DPAT administration induces a 5-HT$_{1A}$ receptor-mediated behavioral syndrome in the rodent species (Backus *et al*., 1990) and rats (Ensler *et al*., 1993). Evidence has shown that the administration of selective 5-HT$_{1A}$ receptor agonist 8-OH-DPAT induced both the complete 5-HT syndrome with all its components and partial syndrome like forepaw treading (Bardin *et al*., 2007; Haleem *et al*, 2002b). It is suggested that forepaw treading is thought to be one of the components of 5-HT syndrome most clearly associated with 5-HT$_{1A}$ receptor activation (Meltzer *et al*., 2003). It is also suggested that

**Corresponding author:* e-mail: batool@uok.edu.pk

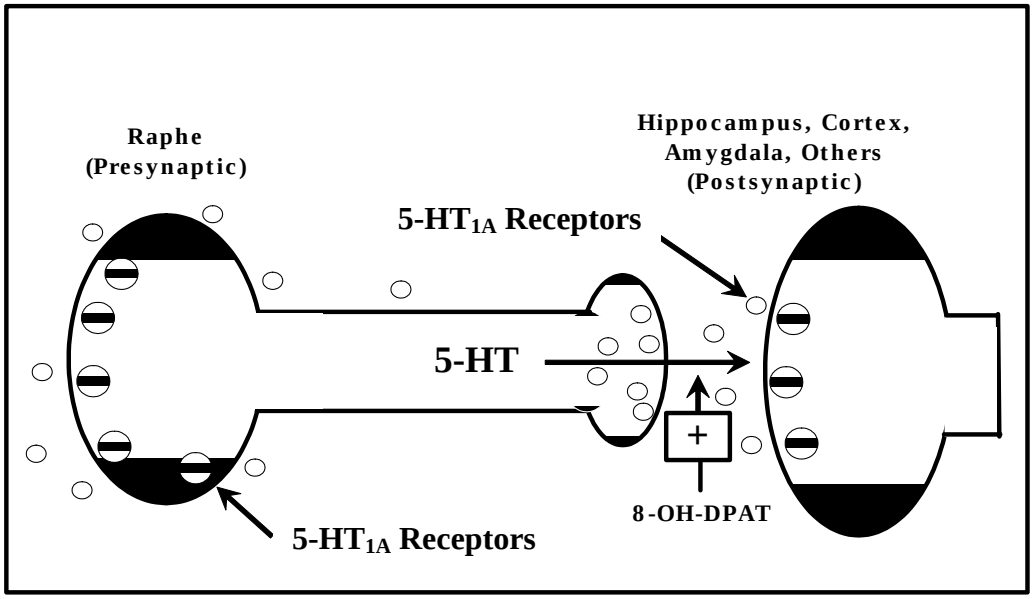

Fig. 1: Schematic representation of a serotonergic neuron summarizing the potential importance of actions at pre- and postsynaptic 5-HT$_{1A}$ receptors in the management of schizophrenia (Full agonist 8-OH-DPAT stimulate both pre- and postsynaptic populations of 5-HT$_{1A}$ receptors).

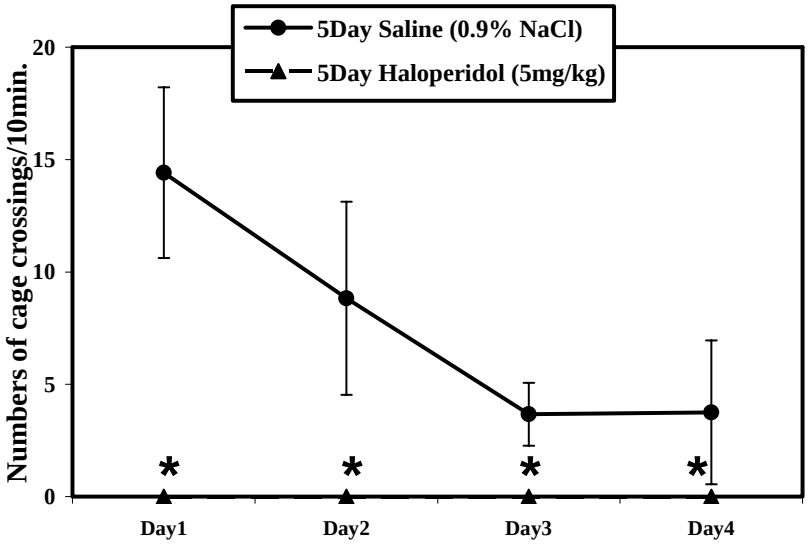

Fig. 2: Effects of haloperidol (5.0mg kg^{-1}) on daily changes of Home cage activity. Values are means ± SD. Significant differences by Newman-keuls test: *p<0.01 from similar day saline injected rats, +p<0.01, ++p<0.05 from 1st day haloperidol injected rats following two-way ANOVA (repeated measure design).

acute administration of 8-OH-DPAT attenuate the cataleptic side effects of haloperidol (Prinssen *et al*., 1999). Previous studies have demonstrated that the activation of 5-HT$_{1A}$ receptors, probably those located presynaptically on 5-HT containing cell bodies (fig. 1), reduces the catalepsy induced by haloperidol (Christofferson and Meltzer, 1998; Hicks, 1990). 5-HT$_{1A}$ receptor agonism is accountable in these behavioral responses that may be mediated, to some extent, by effects of DA release in various brain regions (Watanabe *et al*., 1993). 8-OH-DPAT has the ability to decrease stimulated DA release in medial prefrontal cortex and

nucleus accumbens and this may contribute to the antipsychotic efficacy of D_2 receptor antagonism (Ishikane *et al*., 1997). In consequence, stimulation of 5-HT_{1A} receptors abolishes stimulated DA release and consequent behaviors (Kuroki *et al*., 1996). Our observations demonstrate that activation of 5-HT_{1A} receptors takes an imperative part in the relatively low or non-cataleptogenic liability seen with compounds reported to have dual D_2/ 5-HT_{1A} actions. This confirms previous data reported by Prinssen *et al* (1998), showing that barricade of 5-HT_{1A} receptors by WAY100635 pretreatment "unmasks" this 5-HT_{1A} receptor influence.

It is suggested that 8-OH-DPAT partly attenuated haloperidol-induced dyskinesia due to its weak dopamine agonistic properties (Ishikane *et al*., 1997). Furthermore, systemic administration of 8-OH-DPAT has been reported to augment extracellular dopamine levels in the ventral tegmental area (VTA) and nucleus accumbens (Prinssen *et al*., 1999). Experiments have shown that activity of dopaminergic neurons in the VTA can be increased by the administration of 8-OH-DPAT and it can also abolished by the selective destructions of 5-HT neurons. The evidence suggest that the activation of 5-HT_{1A} autoreceptors in the raphe region are under the influence of stimulation of dopaminergic neurons, that ultimately result in reduced serotonergic input to dopaminergic neuron and a subsequent disinhibition of firing of dopaminergic neurons (Ellenbroek *et al*., 1994). Reported studies have shown that pretreatment with 5-HT_{1A} agonist 8-OH-DPAT did not change dopamine release in nucleus accumbens and striatum produced by haloperidol administration (Prinssen *et al*., 2000). However, 5-HT_{1A} agonist 8-OH-DPAT increase cortical DA release which may underlie efficacy against negative symptoms and reduce DA D_2 antagonist induced extrapyramidal symptoms (Kapur and Ramington, 1996). The decrease in stimulation of DA release in medial prefrontal cortex and nucleus accumbens is explainable on the basis of 5-HT1A receptors selectivity of 8-OH-DPAT and may contribute to the antipsychotic effect of neuroleptics like haloperidol (Ichikawa and Meltzer, 1999). These data support the hypothesis that frontal cortex dopamine neurons may be a common site for antipsychotic action while decreased release of DA in the striatum may be associated with the development of extrapyramidal side effects (Ahlenius, 1989).

In an attempt to determine behavioral responses of 8-OH-DPAT and a role of 5-HT_{1A} receptors in the management of EFS functions, the present study is designed to evaluate the behavioral responses produced by 8-OH-DPAT in rats with subchronic haloperidol treatment. We also tested the hypothesis that whether the repeated administration of haloperidol could modify the sensitivity of presynaptic 5-HT_{1A} receptors stimulated by 8-OH-DPAT. The results are discussed in the context of a causal link between the upregulation of DA-D_2 receptors and the decreases in the effectiveness of presynaptic 5-HT_{1A} receptors following the repeated administration of haloperidol. Taken

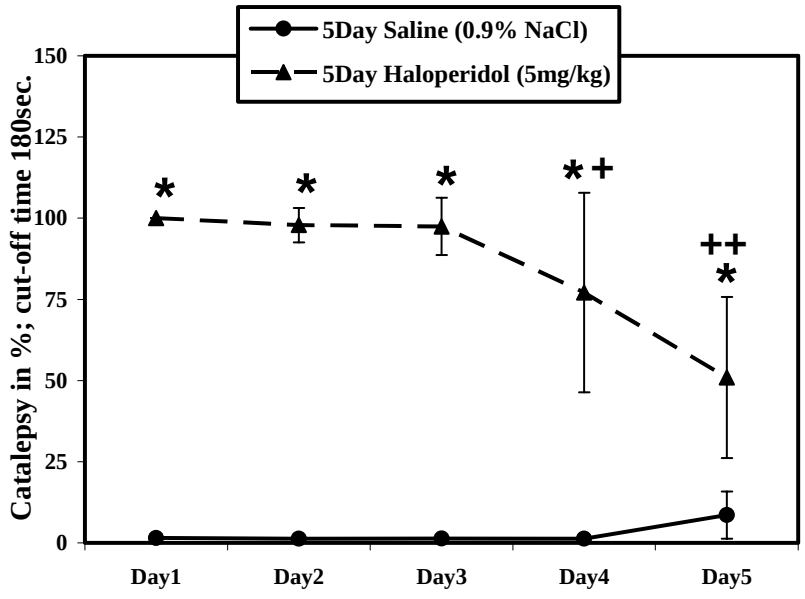

Fig. 3: Effects of haloperidol (5.0mg kg^{-1}) on daily changes of Catalepsy. Values are means ± SD. Significant differences by Newman-keuls test: *p<0.01 from similar day saline injected rats, +p<0.01, ++p<0.05 from 1st day haloperidol injected rats following two-way ANOVA (repeated measure design).

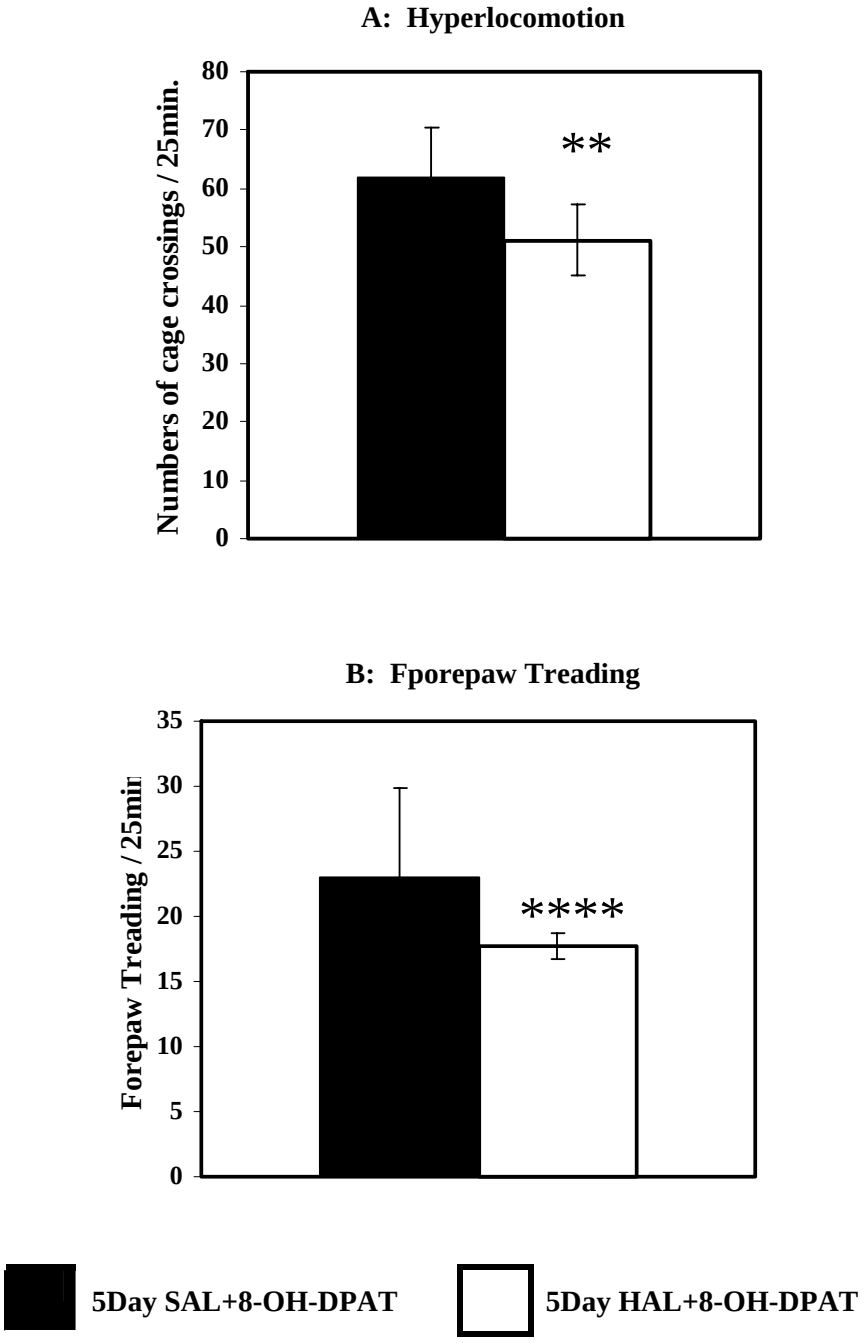

Fig. 4: Effects of 8-OH-DPAT (0.5mg kg^{-1}) elicited hyperactivity syndrome (hyperlocomotion 4A and forepaw treading 4B) in 5 day haloperidol and 5 day saline treated rats. Values are means ± SD (total of four scoring periods of 25min duration). *p<0.001, **p<0.05, ***p<0.01, ****p<0.1 by t-test.

together, these data proposed a well-documented correlation with clinical data in humans to further support the concept of mixed D_2/ 5-HT_{1A} receptor pharmacology and the diverse contribution of 5-HT ligands to the mechanism of action of a neuroleptic in the treatment of schizophrenia.

MATERIALS AND METHODS

Behavioral Methods

Animals

Male Albino-Wistar rats with an average weight of 180±20 gms on arrival purchased from Agha Khan

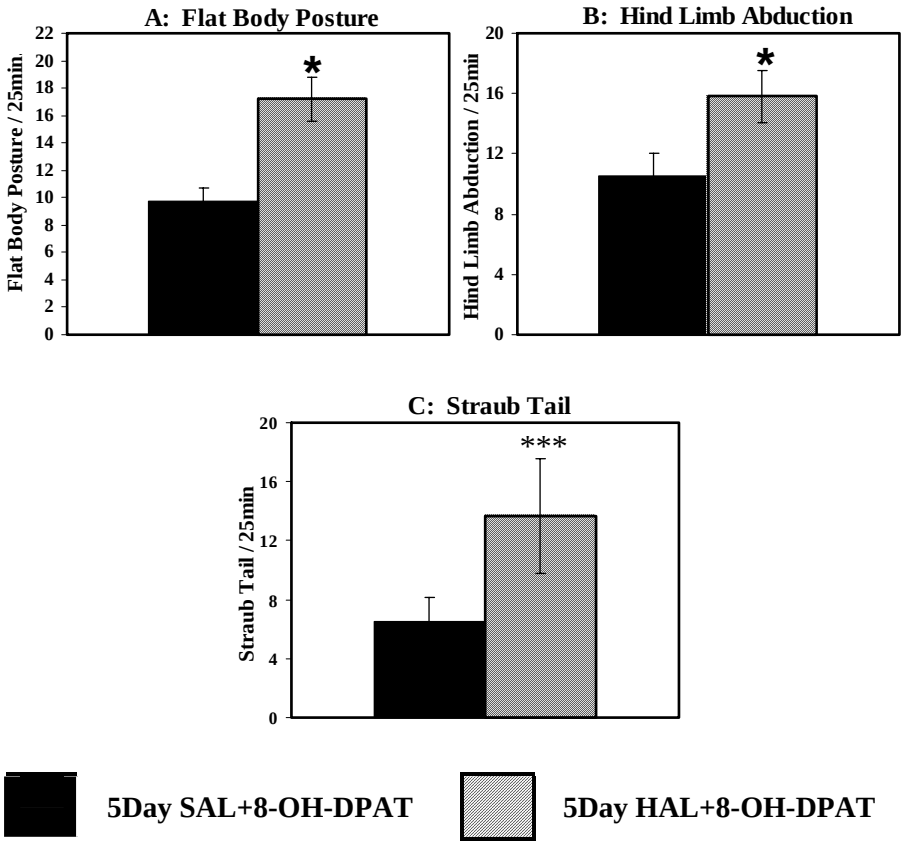

Fig. 5: Effects of 8-OH-DPAT (0.5mg kg^{-1}) elicited 5-HT syndrome (flat body posture 5A, hind limb abduction 5B and straub tail 5C) in 5 day haloperidol and 5 day saline treated rats. Values are means ± SD (total of four scoring periods of 25min duration). *p<0.001, **p<0.05, ***p<0.01, ****p<0.1 by t-test.

University (AKU), were group-housed (2 rats per cage) in an animal-keeping environmentally controlled room (ambient temperature 21±1°C and relative humidity 55±5%) on a 12:12h light/ dark cycle (lights on at 7:00AM). A 5 day acclimatization period was allowed before animals were used in experiments. After this period, and 24h before the behavioral tests, the animals were individually housed in an environmentally controlled test room in transparent Perspex cages (dimensions 26×26×26 cm W×L×H). Food (standard rat diet) and tap water were continuously available to animals during experiment. The rats used for the treatment were all experimentally naive animals. All experimental protocols were approved by and performed in strict accordance with the Guide for the Care and Use of Laboratory Animals (Institute of Laboratory Animal Resources on Life Sciences, US National Research Council, 1996) and the Local Ethical Committee guidelines for animal research.

Drugs and injections

Haloperidol (Serenace; manufactured under license from G.D. Searle and Co. U.S.A, by Searle Pakistan Ltd. Laboratories) available in 5 mg/ml/kg ampoules was used in the present study. R(+) 8-Hydroxy-2-(di-n-propylamino) tetralin hydrobromide (8-OH-DPAT) purchased from RBI Chemicals dissolved in saline (0.9% NaCl) was also used in the treatment. In each experimental group, animals received either drugs or their appropriate vehicle. Route of administration for vehicle or drugs was intraperitoneal (i.p.). Drug doses were calculated as the free base.

Experimental protocol

In order to evaluate the reciprocal relationship between the effects of haloperidol and 8-OH-DPAT, animals were randomly assigned as test and control groups (n=12 animals in each group). Animals of test groups were injected with haloperidol at a dose of 5 mg/kg twice daily for 5 days between 8:00-9:00 am (1st injection) and 3:00-4:00pm (2nd injection). At the same time control animals were injected with saline (0.9% NaCl) 1mg/kg twice daily for 5 days. On the 6th day of experiment, the groups were divided to two (1) 5day saline plus 8-OH-DPAT and (2) 5day haloperidol plus 8-OH-DPAT. Followings the injections of 8-OH-DPAT at a dose of 0.5mg/kg to these

two groups behavioral syndrome were monitored for 25 minutes.

Monitoring of motor activity in a familiar environment
The activity boxes used in the present investigation were specifically designed Perspex home cages (26×26×26 cm) with saw-dust covered floor and the experiment was conducted in a separate quiet room. The procedure used was essentially as described by Batool *et al*. (2001). Controls and treated rats were observed in their home cages for 10 minutes to monitor the activity in terms of numbers of cage crossings for each rat in all sessions.

Rat catalepsy determinations
Catalepsy, defined as the acceptance and retention of abnormal posture, was measured by means of bar test. Bar test determinations were carried out by gently removing rats (n=12) from their home cages and placing their forepaws over a horizontal bar, fixed at a height of 10cm with heads of animals towards upward on an inclined surface at an angle of 60^{o} with the hind limbs abducted (Batool *et al*., 2000 and Batool *et al*., 2001). The length of time during which the animal retained this position was recorded by measuring the time from the placement of the rat until removal of one of its forepaws. In the present study, testing was performed 30 minutes after the drug administration for 5days consecutively to monitor daily changes on catalepsy and the time to withdrawal of legs by the rats were measured. The intensity of catalepsy was assessed by counting the time and the animal remained in this position with a maximal "cut-off" 180sec. Rats were removed from the bar if their latency on the bar test exceeded by 180sec.

8-OH-DPAT-Elicited 5-HT syndrome
On the last day of experiment 8-OH-DPAT (0.5mg/kg) was administered as a challenge dose to the groups of animals pretreated with 5day saline and 5day haloperidol (5mg/kg). The 5-HT syndrome behaviors were observed in animals for a total scoring period of 25 min, 5min post injections of 8-OH-DPAT. During the observation period, intermittent behaviors like hyperlocomotion (circling around the perimeter of the cage) and forepaw treading (hitting forepaw on the floor like piano playing) were scored. Continuous behaviors such as flat body posture, hind limb abduction and straub tail (snake like tail) were monitored on a scale of relative intensity (0-4) for 25 min. (Haleem and Khan, 2003).

STATISTICAL ANALYSIS

The results presented in this investigation are as means $\pm$ SD. Drug-induced behavioral responses i.e., daily changes in catalepsy and numbers of cage crossings in home cages in 5day saline and 5day haloperidol treated rats were statistically analyzed by two-way ANOVA (repeated measure design). Posthoc comparisons were done by Newman-Keuls test. Data on the effects of 8-OH-DPAT elicited 5-HT syndrome was analyzed by two-tailed t-test. Values $p<0.05$ were considered statistically significant.

RESULTS

Effects of repeated haloperidol administration (5mg/kg twice daily for 5days) on daily changes of home cage activity
Fig. 2 shows the effects of repeated administration of haloperidol at a dose of 5 mg/kg twice daily for 5 days on daily changes of Home Cage Activity (fig. 2) in rats.

Data on home cage activity (as numbers of cage crossings/10min; fig. 2) analyzed by two-way ANOVA (repeated measure design) showed a significant repeated ($F=17.28$; $df=4,88$; $p<0.01$) and single drug effect ($F=43.0$; $df=1,22$; $p<0.01$) and significant interaction between repeated and single drug effect ($F=26.65$; $df=4,88$; $p<0.01$).

Newman-Keuls test performed on Home cage Activity showed that repeated administration of haloperidol significantly ($p<0.01$) decreased numbers of cage crossings in rats injected with haloperidol at a dose of 5 mg/kg twice daily for 4 days. All the decreases were comparable following 1st, 2nd, 3rd and 4th daily administration of haloperidol.

Effects of repeated haloperidol administration (5mg/kg twice daily for 5days) on daily changes of catalepsy
Fig. 3 shows the effects of repeated administration of haloperidol at a dose of 5mg/kg twice daily for 5 days on daily changes of Catalepsy (fig. 3) in rats. Data on daily changes of Catalepsy in percentage (%) (fig. 3) analyzed by two-way ANOVA (repeated measure design) showed a significant repeated ($F=14.91$; $df=4,88$; $p<0.01$) and significant drug effect ($F=612.33$; $df=1,22$; $p<0.01$) and significant interaction ($F=25.76$; $df=4,88$; $df=p<0.01$) between repeated and single drug treatment.

Posthoc comparisons done by Newman-Keuls test showed that significant ($p<0.01$) cataleptic effect (immobile posture on an inclined surface) was produced on 1st, 2nd and 3rd day of haloperidol administration i.e, 100% effect, but the decreases following 4th and 5th day of injections were significant ($p<0.05$) but not 100% as compared to the decreases observed following 1st day of haloperidol injections in rats.

8-OH-DPAT elicited 5-HT syndrome in 5 day saline and 5 day haloperidol injected rats
Fig. 4(A and B) show the 8-OH-DPAT elicited 5-HT syndrome (hyperlocomotion, A and forepaw treading, B) in 5 day saline and 5 day haloperidol injected rats. Analysis by t-test showed that 8-OH-DPAT elicited hyperlocomotion ($p<0.05$; fig. 4A) and forepaw treading

($p<0.1$; fig. 4B) were significantly lower in 5day haloperidol injected rats than 5day saline injected rats.

Fig. 5 (A, B and C) show the 8-OH-DPAT elicited 5-HT syndrome (flat body posture, A; hind limb abduction, B and straub tail, C) in 5day saline and 5day haloperidol injected rats. 8-OH-DPAT elicited flat body posture ($p<0.001$; fig. 5A), hind limb abduction ($p<0.001$; fig. 5B) and straub tail ($p<0.01$; fig. 5C) were significantly increased in rats repeatedly injected with haloperidol for 5 days when injected with 8-OH-DPAT (0.5 mg/kg) on the 6^{th} day before killing when compared with 5day saline injected rats.

DISCUSSION

Recently, attention has turned to activation of serotonin$_{1A}$ receptors as a promising component of antipsychotic drug action. 5-HT_{1A} receptor agonists are thought to enhance the antipsychotic-like effects of DA-D_2 receptor antagonists, while reducing their potential to produce extrapyramidal side effects. The present study was two-fold: first, previously reported rats administered with long-term haloperidol would develop dopaminergic supersensitivity evaluated by locomotion and catalepsy behavior, and second, to investigate behavioral responses expressed by the acute challenge dose of 8-OH-DPAT in these paradigms. In the line with these observations, number of investigations reported that the administration 5-HT_{1A} agonists could abolish acute cataleptogenic effects of traditional antipsychotic drugs (Balsara *et al*., 1979; Invernizzi *et al*., 1988; Hicks, 1990). This study shows that administration of 5-HT_{1A} agonist, 8-OH-DPAT, at a dose that preferentially stimulates somatodendrtitic 5-HT_{1A} receptors (Haleem *et al*., 2007) could reverse the induction of EPS functions by haloperidol. The results, therefore, support the notion that the activation of somatodendritic 5-HT_{1A} receptors may be of use in the treatment of EPS induced by haloperidol or other antipsychotic drugs, also suggesting that a drug which incorporates both D_2 antagonism and 5-HT_{1A} agonism would exhibit antipsychotic activity but little or no EPS.

Our results have demonstrated that haloperidol-induced deficits of locomotor activity were not abolished by a single administration of 8-OH-DPAT. The apparent failure of 8-OH-DPAT to attenuate haloperidol-induced deficits of locomotor activity, however, can be explainable in terms of large decreases of locomotor activity in haloperidol treated rats at a selective dose (fig. 2) or might be decreases in the inhibitory serotonergic influence on the activity of dopaminergic neurons (Samad *et al*., 2007). Neuroleptic drug-induced EPS are one of the major limitations to effective neuroleptic treatment (Baldessarini, 1985). These disorders have both motor (objective) and mental (subjective) aspects, which must be considered in any evaluation and differential diagnosis of treatment-related side effects (Casey, 1994). Acute EPS are commonly explained on the basis of dopamine D_2 receptor antagonism, whereas, serotonin agonist often reduce or prevent catalepsy in rodents (Kleven *et al*., 1996). It has also been reported that there was a direct relationship between the impairment of motor function induced by the single haloperidol administration and the increment of general activity observed after withdrawal from repeated drug administration. Our results were considered to be a consequence of the supersensitivity of central dopaminergic pathway, and the ability of 8-OH-DPAT to reverse haloperidol-induced catalepsy may be consequently decreased central 5-HT transmission induced by the stimulation of inhibitory somatodendritic 5-HT_{1A} autoreceptors within the raphe nuclei (Invernizzi *et al*., 1988; Needham *et al*., 1994).

In agreement with previous studies (Balsara *et al*., 1979; Invernizzi *et al*., 1988), we reported that administration of 5-HT_{1A} agonists attenuate the cataleptic side effects of traditional antipsychotic haloperidol. Reported studies have also shown that 5-HT_{1A} agonists buspirone, ipsapirone and 8-OH-DPAT all potently reversed catalepsy in experimental animals (Haleem *et al*., 2007). It is therefore, suggested that 5-HT_{1A} receptor sites are important in the serotonergic modulation of haloperidol-induced catalepsy (Millan, 2000). A concept of the contribution of serotonin receptor subtypes in the behavioral effects of neuroleptic drugs emerged because of the hypothesis that prolonged blockade of central dopaminergic receptors by neuroleptics cause subsequent behavioral effect that may be due to the development of enhanced receptor sensitivity (Creese *et al*., 1977; Vonvoigtlander *et al*., 1975). The 5-HT_{1A} receptor is one of the several subtypes of receptors for serotonin that exist in the brain (Bantick *et al*., 2004) and is located presynaptically on cell bodies in the raphe (fig. 1), where its activation inhibits the firing rate of raphe neurons (Wadenberg, 1996). Striatum is a projection area of dorsal raphe nuclei and enrich of dopaminergic neurons. Hence, dopamine induced membrane depolarization in the dorsal raphe serotonergic neurons is mediated by the activation of dopamine D_2 like receptors (Lucas and Bonhomme, 1997). The dopamine system has traditionally been considering crucial to the control of motor activity (Bishnoi *et al*., 2007). With respect to the anatomical site of action a view has developed that the striatum is involved in the control of motor activity. Serotonin inhibits dopamine neurotransmission at the level of origin of dopamine system in the midbrain and in the terminal region (Sandyk and Fischer, 1988). Serotonin agonists with selectivity towards 5-HT_{1A} receptors could release dopamine neurotransmission from the inhibitory influence of 5-HT to alleviate EPS functions of haloperidol (Kapur and Ramington, 1996; Millan *et al*., 1998). Another supporting evidence show that exposure to haloperidol for 21 days in experimental animals caused an upregulation

in the striatum but not in the cortex of D_2 receptors (Invernizzi *et al*., 1988). These results suggest that dopamine-regulating mechanisms in the striatum following repeated administration of haloperidol may involve in the expression of behavioral dopaminergic supersensitivity and suggest that EPS functions of neuroleptics may be due in part to blockade of D_2 receptors in the striatum (Kapur and Ramington, 1996; Millan *et al*., 1998).

It has been shown that chronic administration of neuroleptic drugs may have different effects on cerebral dopamine systems. Initial antagonism of dopamine mediated behavior, such as stereotype, disappears, and may be replaced by supersensitivity to dopamine agonists (Casey, 1993). It is suggested that repeated treatments of rats with neuroleptics such as haloperidol, results in behavioral supersensitivity of dopaminergic receptors indicated by upregulation of DA receptors in nigrostriatal pathway. In the present study, we also assessed the ability of 8-OH-DPAT to produce componenets of the 5-HT_{1A} behavioral syndrome in haloperidol treated rats. Our data demonstrate that 8-OH-DPAT elicited flat body posture, hind limb abduction and straub tail following 5days haloperidol administration were greater in haloperidol injected rats than 5day saline injected rats (figs. 4A, B and C).

It is also reported that haloperidol produced a progressive decline in the locomotion (Haleem *et al*., 2002) and this is in general agreement with our results (fig. 2B). Prinssen *et al* (1999) reported that 5-HT_{1A} agonist 8-OH-DPAT attenuated the EPS of haloperidol. In addition, decreases in forepaw treading and hyperlocomotion were seen in animals preinjected with haloperidol and administered with single injection of 8-OH-DPAT in the last day of treatment (fig. 3A). A decrease in the intensity of 8-OH-DPAT-induced hyperactivity syndrome by haloperidol suggests that DA-D_2 receptors also contribute in the expression of the syndrome (Tricklebank *et al*., 1984). These results suggest that the inability of 8-OH-DPAT to induce forepaw treading was due to an interaction between DA-D_2 and 5-HT_{1A} receptors also reported by Hinds and Kilpartrick (1994). Reported study have also shown that repeated administration of haloperidol increases the density of D_2 receptors (Wadenberg *et al*., 2001), as repeated administration of haloperidol induces striatal DA receptor supersensitivity. We tested the modulatory effect of 8-OH-DPAT on rats preinjected with haloperidol for 5days. Our data shows that 8-OH-DPAT elicited flat body posture was greater in haloperidol treated rats on the comparison with saline treated rats (fig. 4A).

In present investigation we found that repeated administration of a high dose of haloperidol was found to cause a long-lasting effect on both dopaminergic and serotonergic neurons that paralleled severe impairment of locomotor performance (fig. 2B). We also tested the hypothesis that 5-HT syndrome is mediated by postsynaptic 5-HT_{1A} receptors. It is reported that 5-HT_{1A} postsynaptic receptors become supersensitized after repeated administration of haloperidol (Haleem and Khan, 2003). Two components of 5-HT syndrome are due to action at postsynaptic sites (flat body posture and forepaw treading (Yamada *et al*., 1989). The results of present study indicate that serotonergic agonists behave as typical partial agonist for D_2 receptors and acute administration of 8-OH-DPAT attenuate cataleptic side effects of neuroleptics.

8-OH-DPAT, a selective serotonin agonist at 5-HT_{1A} receptors, preferentially stimulates somatodendritic 5-HT_{1A} receptors at a dose of 0.5 mg/kg. Stimulation of somatodendritic 5-HT_{1A} receptors by a selective 5-HT_{1A} agonist 8-OH-DPAT, resulting in a decrease in the availability of serotonin at postsynaptic site, could release DA neurotransmission from the inhibitory influence of 5-HT, and so attenuate haloperidol-induced catalepsy (Haleem *et al*., 2004; Haleem *et al*., 2007). It is suggested that induction of EPS functions by haloperidol could be abolished by the coadministration of 5-HT_{1A} agonist, 8-OH-DPAT, signifying a role of preferentially somatodendritic or postsynaptic 5-HT_{1A} receptors in unmasking of EPS and in the expression of behavioral dopaminergic supersensitivity in haloperidol treated rats. The consequence of such behavioral responses to chronic neuroleptic therapy may be of importance to understanding of schizophrenia and these issues will be addressed in future studies.

CONCLUSION

In conclusion, the present investigation suggests that 5-HT_{1A} agonist 8-OH-DPAT may abolish the extrapyramidal effects of traditional neuroleptics. This study shows an interaction of dopamine D_2 and 5-HT_{1A} receptors in the manifestation of at least one of the components of 5-HT syndrome elicited by the administration of 8-OH-DPAT in repeated haloperidol injected rats. It is conceivable from our results that D_2/5-HT_{1A} mechanism alters the antipsychotic-like effects of neuroleptics and attenuate EPS functions. Therefore, it may be interesting to monitor the results in detail, correlate with clinical data in humans to further support the concept, and indicate marked diversity in the contribution of mixed D_2/ 5-HT_{1A} receptor pharmacology and the role of 5-HT_{1A} ligands in the mechanism of action of neuroleptics leading to new avenues of exploration in the treatment of schizophrenia.

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