*SHORT COMMUNICATION*

# ANTAGONISM OF APOMORPHINE-INDUCED CAGE CLIMBING BEHAVIOUR AND METHAMPHETAMINE STEREOTYPY BY FENFLURAMINE IN MICE

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**Summary :** Pretreatment with fenfluramine (5 and 10 *mg/kg*, ip) in doses which induced head twitches was found to antagonize apomorphine-induced cage climbing behaviour and methamphetamine stereotypy in mice. Since fenfluramine (5 and 10 *mg/kg*) did not induce catalepsy it indicates that fenfluramine lacks postsynaptic striatal and mesolimbic dopamine receptor blocking activity and it is possible that the fenfluramine-induced enhancement of central 5-hydroxytryptamine neuronal transmission may be responsible for its antagonistic effect on apomorphine-induced climbing behaviour and methamphetamine stereotypy.

**Key words :** fenfluramine, apomorphine, methamphetamine, climbing behaviour, stereotypy, mice

## INTRODUCTION

Apomorphine-induced cage climbing behaviour in mice occurs as a result of direct stimulation of postsynaptic striatal and mesolimbic dopamine (DA) receptors by apomorphine (3) while methamphetamine stereotypy results from activation of postsynaptic striatal and mesolimbic DA receptors by released DA (11). Recently, Costall *et al.* (3) have reported that quipazine, a drug that increases central 5-hydroxytryptamine (5-HT) neuronal transmission by stimulating the release (5) and inhibiting the reuptake of 5-HT (7), antagonised apomorphine-induced cage climbing behaviour in mice. Since fenfluramine also increases 5-HT transmission in brain by stimulating the release and inhibiting the reuptake of 5-HT (4, 12) we decided to investigate the effect of pretreatment with fenfluramine on apomorphine-induced cage climbing behaviour and on methamphetamine stereotypy in mice. Further, we have investigated whether fenfluramine induces catalepsy in mice.

## MATERIAL AND METHODS

Male albino mice, 20 to 30 g, in groups of ten for each treatment and allowed free access to a standard diet and tap water were used. Each animal was used once only.

All observations were made between 10.00 and 16.00 hr at 27 to 30°C in a noiseless, diffusely illuminated room. During the experiments the animals were placed in individual cages made of wire netting, measuring 27 cm x 20 cm and 15 cm high, 30 min before drug treatment to allow adaptation to the new environment.

The effect of fenfluramine pretreatment on apomorphine-induced cage climbing behaviour was studied by the method of Costall *et al.* (3). The animals were individually tested for climbing behaviour taking 'the percentage of time spent climbing during the 30 min period after the first climb' as a measure of climbing ('climbing index'). Further, the maximum time (in min) spent in a single climb throughout the duration of the apomorphine effect was also recorded.

The intensity of methamphetamine stereotypy was assessed over a 30 sec observation period at 10 min intervals throughout its duration, according to the scoring system of Ozawa and Miyauchi (9), as follows : no stereotypy (0) ; discontinuous sniffing (1) ; continuous sniffing (2) ; discontinuous biting, gnawing or licking (3) ; continuous biting, gnawing or licking (4).

Animals were tested for catalepsy by the method of Ahtee and Buncombe (1) by placing both front paws on a 4 cm high wooden block and measuring the time that the animal maintained this posture. They were tested for catalepsy 0.5, 1.0 and 2.0 hr after fenfluramine treatment and were considered cataleptic if they maintained this imposed posture for more than 10 sec.

The drugs used were ; dl-fenfluramine HCl (Walter Bushnell), apomorphine HCl (Sigma) and methamphetamine HCl (Burroughs Wellcome). Fenfluramine and methamphetamine were dissolved in distilled water, while apomorphine was dissolved in distilled water containing 0.2 *mg/ml* ascorbic acid. All drugs were injected ip in a volume of 0.1 *ml/10 g* body weight. Doses quoted refer to the salt. Fenfluramine (or distilled water, in control groups) was injected 30 min before apomorphine or methamphetamine.

Effects of fenfluramine pretreatment on apomorphine-induced climbing behaviour were statistically analysed by the two-tailed Student's t–test, and on methamphetamine stereotypy by the Mann-Whitney U-Test for non-parametric data.

## RESULTS

Fenfluramine (2.5 *mg/kg*) did not induce head twitches while in higher doses (5, 10 and 15 *mg/kg*) it induced head twitches, in the form of brisk, jerky side to side shaking movement of the head. As 15 *mg/kg* fenfluramine induced abduction and extension

of hind limbs, animals receiving this dose were not tested for catalepsy and this dose was not used for drug interaction studies. Further, fenfluramine (2.5, 5 and 10 *mg/kg*) did not induce catalepsy when the mice were tested upto 2 hr after injection.

Pretreatment with 2.5 *mg/kg* fenfluramine did not significantly influence apomorphine (0.75 and 1 *mg/kg*)-induced climbing behaviour (Table I) and methamphetamine (6 and 7 *mg/kg*) stereotypy (Table II). However, pretreatment with 5 and 10 *mg/kg*

TABLE I : Effect of pretreatment with fenfluramine (FEN) on apomorphine (APO)-induced cage climbing behaviour in mice.

| *Treatment (dose mg/kg)* | *Climbing index (%) Mean ± S.E.M.* | *Maximum time (min) Mean ± S.E.M.* |
|---|---|---|
| 1. APO 0.75 | 46.7±3.4 | 6.5±0.7 |
| 2. FEN 2.5+APO 0.75 | 44.9±2.9 | 6.2±0.9 |
| 3. FEN 5+APO 0.75 | 28.2±2.7* | 3.3±0.3* |
| 4. FEN 10+APO 0.75 | 7.8±2.2** | 1.1±0.6** |
| 1. APO 1 | 72.9±3.3 | 12.2±0.7 |
| 2. FEN 2.5+APO 1 | 71.3±3.6 | 11.5±0.8 |
| 3. FEN 5+APO 1 | 54.2±2.9* | 8.8±0.4* |
| 4. FEN 10+APO 1 | 34.3±2.7** | 6.2±0.5** |

*P<0.05, **P<0.01 (Student's t - test). Numerals following the drugs indicate their doses (*mg/kg*).

TABLE II : Effect of pretreatment with fenfluramine (FEN) on methamphetamine (MAM)-induced stereotyped behaviour in mice.

| *Treatment (dose mg/kg)* | *Intensity score Mean ± S.E.M.* |
|---|---|
| 1. MAM 6 | 2.8±0.13 |
| 2. FEN 2.5+MAM 6 | 2.4±0.16 |
| 3. FEN 5+MAM 6 | 2.0±0.00** |
| 4. FEN 10+MAM 6 | 1.4±0.16*** |
| 1. MAM 7 | 3.9±0.10 |
| 2. FEN 2.5+MAM 7 | 3.6±0.16 |
| 3. FEN 5+MAM 7 | 3.2±0.13* |
| 4. FEN 10+MAM 7 | 2.6±0.16*** |

*P<0.05 **P<0.02 ***P<0.001 (Mann-Whitney U-Test). Numerals following the drugs indicate their doses *(mg/kg)*.

fenfluramine significantly ($P<0.05$ or less) decreased the intensity of apomorphine (0.75 and 1 *mg/kg*)-induced climbing behaviour (Table I) and also of methamphetamine (6 and 7 *mg/kg*) stereotypy (Table II).

## DISCUSSION

Classical neuroleptics, like haloperidol, induce catalepsy and antagonize apomorphine-induced cage climbing behaviour and stereotypy and methamphetamine stereotypy by blocking postsynaptic striatal and mesolimbic DA receptors (3, 10, 11). Though fenfluramine was effective in antagonising apomorphine-induced climbing behaviour and methamphetamine stereotypy it however, failed to induce catalepsy in mice. Since fenfluramine did not induce catalepsy it indicates that fenfluramine most probably lacks postsynaptic DA receptor blocking activity and that it antagonises apomorphine-induced climbing behaviour and methamphetamine stereotypy by some other mechanism.

Recently, the induction of cage climbing and stereotyped responses by DA agonists has been shown to be influenced by drugs affecting the central 5-HT systems (3, 13, 14). Pretreatment with quipazine, a 5-HT agonist, antagonised apomorphine-induced climbing behaviour, while pretreatment with methysergide, a 5-HT antagonist, potentiated it (3). Similarly, pretreatment with 5-HT precursors, L-tryptophan and 5-hydroxytryptophan, antagonised amphetamine and apomorphine-induced stereotyped behaviour, while pretreatment with p-chlorophenylalanine, a specific depletor of brain 5-HT, and methysergide potentiated it (2, 6, 8, 13, 14). These results suggest that the central 5-HT systems may have an opposing inhibitory effect upon the central DA systems involved in the mediation of climbing and stereotyped behaviours. Since fenfluramine stimulates the release and inhibits the reuptake of 5-HT (4, 12) it is possible that the fenfluramine-induced enhancement of central 5-HT neuronal transmission may be responsible for its antagonistic effect on apormorphine-induced climbing behaviour and methamphetamine stereotypy.

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