LETTER TO THE EDITOR

FENFLURAMINE-INDUCED HEAD-TWITCH RESPONSE IN MICE AND ITS MODIFICATION BY CERTAIN DRUGS INFLUENCING THE CENTRAL 5-HYDROXYTRYPTAMINE FUNCTION

Sir,

(Received on May 23, 1983)

While studying the behavioural effects of fenfluramine in our laboratory we observed that the drug induces head-twitches in mice. Since head-twitches can be due to increased activity of central 5-hydroxytryptamine (5-HT) neuronal systems (1, 9) and since fenfluramine produces selective, prolonged depletion of brain 5-HT (2, 4), we have studied the effects of pretreatment with methysergide, p-chlorophenylalanine (PCPA) and clomipramine, the drugs known to influence central 5-HT mechanisms, on fenfluramine-induced head twitches in mice. Effect of clomipramine on 5-hydroxytryptophan (5-HTP)-induced head twitches was also determined.

Male albino mice, 20 to 30 *g*, were used in groups of 10 for each treatment. They had free access to tap water and a diet locally composed as per Haffkine Institute's specifications. Each animal was used once only. All observations were made at ambient room temperature (27°-30°C), between 10.00 and 16.00 hr in a noiseless, diffusely illuminated room.

Fenfluramine or 5-HTP-induced head twitches was studied in control or drug-pretreated mice by the method of Orikasa and Kisara (10). The number of head twitches was counted for 2 min at 10 min intervals, between 10 and 92 min, starting immediately after drug administration (fenfluramine, ip/5-HTP, iv).

The statistical significance of differences between mean cumulative scores of various groups was assessed by the Student's unpaired t-test.

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, in a dose-dependent manner (Table I). The dose of 15 *mg/kg* also induced abduction and extension of hind limbs, and higher doses were therefore not used.

Methysergide (5, 10 *mg/kg*), PCPA (100 *mg/kg/day* X 3 days) and clomipramine

(5, 10 *mg/kg*) induced neither gross behavioural changes nor head twitches in mice. Effect of these pretreatments on head twitches induced by various doses of fenfluramine is shown in Table I alongwith effect of clomipramine pretreatment on 5-HTP-induced head twitches.

TABTLE I : Head twitch responses induced by fenfluramine (or 5-hydroxytryptophan, 5-HTP) in mice pretreated with normal saline or with drugs, viz., methysergide (METH), p-chlorophenylalanine (PCPA) or clomipramine (CIMI).

Response to[a]	*Pretreatment*[b] *(mg/kg)*	*Number of head twitches*[c] *Mean±S.E.M.*
Fenfluramine	1. Normal saline (controls)	15.2±2.5
5 *mg/kg*, ip	2. METH 5	0
	3. PCPA 300	0
	4. CIMI 5	0
Fenfluramine	1. Normal saline (controls)	34.7±2.2
10 *mg/kg*, ip	2. METH 5	3.6±1.9**
	3. METH 10	0
	4. PCPA 300	0
	5. CIMI 5	9.2±2.7**
	6. CIMI 10	0
Fenfluramine	1. Normal saline (controls)	40.2±2.9
15 *mg/kg*, ip	2. METH 5	5.9±1.6**
	3. METH 10	0
	4. PCPA 300	3.4±1.4**
	5. CIMI 5	14.6±2.3**
	6. CIMI 10	3.2±1.8**
5-HTP	1. Normal saline (controls)	52.4±3.4
100 *mg/kg*, iv	2. CIMI 5	79.9±2.9*
	3. CIMI 10	87.2±2.5**

[a]dl-Fenfluramine HCl (Walter Bushnell) was given as aqueous solution, and 5-HTP (Sigma) as a solution in normal saline.

[b]dl-PCPA (Sigma, 100 *mg/kg/day*) was given ip as suspension in 1% methyl-cellulose solution on 3 consecutive days; animals were used 24 hr after last injection. Methysergide hydrogen maleinate (Sandoz) and clomipramine HCl (Ciba-Geigy) were given ip as aqueous solutions 30 min before fenfluramine.

[c]See text for methodology. There were 10 mice per group. Values differ significantly from controls. (*$P<0.01$, **$P<0.001$).

Pretreatment with methysergide, a 5-HT receptor antagonist (3) and PCPA, a specific brain 5-HT depletor (8) effectively prevented fenfluramine-induced head twitches in mice. Magnitude of blockade depended upon intensity of pretreatments and on dose of fenfluramine (Table I). These results suggest that fenfluramine induced head

twitches involve 5-HT receptors, but the activation is indirect, mediated through release of endogenous 5-HT. Our suggestion is consistent with the reports that fenfluramine causes a rapid release of 5-HT (5) and induces a 5-HT mediated behavioural syndrome in rats, which is antagonised by PCPA pretreatment (12).

In view of the proposed indirect mode of action of fenfluramine it was of interest to compare its effects with that of 5-HTP, which increases brain 5-HT concentration (1,9) with resultant activation of 5-HT receptors. 5-HTP also induced head-twitch response, which was qualitatively indistinguishable from that produced by fenfluramine. Clomipramine pretreatment increased the number of 5-HTP induced head twitches, in accord with earlier reports, because of its ability to block the neuronal reuptake of 5-HT (7, 11). In contrast the pretreatment blocked the effect of fenfluramine. Clomipramine pretreatment has been reported to antagonize the anorectic and 5-HT depleting action of fenfluramine (6), and the antagonism has been explained on the basis that clomipramine, by blocking the uptake of fenfluramine into 5-HT nerve terminals, prevents the release and depletion of 5-HT by fenfluramine. The antagonism of fenfluramine-induced head twitches by clomipramine may have a similar explanation. Further, as clomipramine produced opposite effects on fenfluramine- and 5-HTP-induced head twitches; possibly fenfluramine and 5-HTP enter the 5-HTergic neurones through different uptake mechanims.

In conclusion, fenfluramine apparently induces head twitches in mice indirectly by releasing 5-HT from 5-HTergic neurones with a resultant stimulation of 5-HT receptors.

ACKNOWLEDGEMENTS

The authors are grateful to the Dean, V. M. Medical College for providing facilities, and to Walter Bushnell, Ciba-Geigy and Sandoz Ltd., for their generous gifts of the drugs used. The technical assistance of Mr. S.S. Chavan is gratefully acknowledged.

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