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Effects of the Serotonin Reuptake Inhibitor Fluvoxamine on Yohimbine-Induced Anxiety in Panic Disorder

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Abstract. To assess the effects of the selective serotonin reuptake blocker fluvoxamine on noradrenergic function in patients with panic disorder, an intravenous yohimbine challenge test was administered to eight patients with panic disorder before and after 8 weeks of fluvoxamine treatment and to a parallel group of eight patients treated with placebo. Fluvoxamine treatment reduced yohimbine-induced anxiety while placebo treatment had no effect on this variable. Both fluvoxamine and placebo treatment had little effect on biochemical or physiologic responses to yohimbine.

Key Words. Norepinephrine, 3-methoxy-4-hydroxyphenylglycol, cortisol, blood pressure, heart rate.

There is a substantial literature implicating the serotonin (5-hydroxytryptamine; 5-HT) system as a mediator of fear responses in animals and man (Kahn et al., 1988*a*). Furthermore, the therapeutic mechanism of action of clinically effective antipanic medications may be mediated by the 5-HT system. For example, the antipanic efficacy of potent serotonin reuptake inhibitors (SRIs) such as clomipramine (Beaumont, 1977; den Boer et al., 1987), fluvoxamine (den Boer and Westenberg, 1990), zimelidine (Evans et al., 1986), and fluoxetine (Gorman et al., 1987) is well documented. In fact, two clinical studies suggest that the SRIs may be superior in clinical efficacy for panic disorder to medications that influence the norepinephrine (NE) system (den Boer and Westenberg, 1988; Modigh et al., 1992). Earlier preclinical (Redmond and Huang, 1979) and clinical studies (Charney et al., 1990) have also implicated the NE neurotransmitter system in the mediation of fear responses in animals and humans. Specifically, pharmacologic challenge with the α_2-adrenergic antagonist yohimbine has been used as a probe of NE functioning in panic disorder. In a subgroup of panic patients, yohimbine increases panic-anxiety

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symptoms and produces elevations in 3-methoxy-4-hydroxyphenylglycol (MHPG) compared with levels found in healthy control subjects (Charney et al., 1984, 1987, 1992).

Few clinical studies have examined the effects of serotoninergic medications on NE functioning in anxiety patients. In the present study, yohimbine challenges were conducted before and after chronic fluvoxamine or placebo treatment in patients who currently met *DSM-III-R* criteria for panic disorder. The behavioral, physiological, and biochemical (MHPG and cortisol) responses to yohimbine were evaluated both before and after treatment.

Methods

Patients. All patients gave written informed consent to participate in the study. They were admitted to the Yale Anxiety Disorders Clinic at the Connecticut Mental Health Center, New Haven. After completion of a screening interview and then a structured interview, the Anxiety Disorders Interview Schedule, revised version (DiNardo and Barlow, 1988), 16 patients were enrolled in and completed the study. The patients all met *DSM-III-R* criteria (American Psychiatric Association, 1987) for a principal diagnosis of panic disorder. There were nine male and seven female patients with a mean age of 36 years (SD = 12). Table 1 presents the rating data obtained at baseline and after treatment. Each patient underwent a careful medical and neurological evaluation and was found to be free from significant medical problems.

Medication Protocol. Patients received no psychotropic medication for at least 1 week before entry into the medication protocol. After a single-blind placebo period (mean = 3 weeks, range = 1-4 weeks), patients were randomized to either placebo or active fluvoxamine for 8 weeks. These patients were a subsample of patients involved in ongoing efficacy studies of fluvoxamine in panic disorder. The present study, however, was not specifically designed to evaluate the clinical efficacy of fluvoxamine. Thirty-three patients received single-blind treatment and during this time participated in one placebo and one yohimbine challenge test. Patients were randomized in the medication protocol only if they had at least one panic attack per week during the single-blind placebo phase of the study. Fluvoxamine was started at a dose of 50 mg/day (1 capsule) and was titrated to a maximum of 300 mg (6 capsules). During the study, all patients attended weekly group psychotherapy sessions lasting 60 minutes. The patients in the groups were all participants in the medication protocols. The groups consisted of between 6 and 10 patients and 2 therapists. Psychoeducation, relaxation, and supportive, but not exposure techniques were used in the treatment.

Patients received weekly ratings assessing the severity of their illness and response to treatment. An experienced research assistant, unaware of patient treatment status, completed the Hamilton Rating Scale for Anxiety (HRSA; Hamilton, 1969), the Clinician-Rated Anxiety Scale (CRAS; Sheehan, 1986), and the Clinician Global Change Scale (CGCS; Ballenger et al., 1988), a 0- to 10-point scale, in which 0 = extreme clinical worsening, 5 = no clinical change, and 10 = complete recovery. Raters administered the 24-item Hamilton Rating Scale for Depression (HRSD; Hamilton, 1960) every month during the study. The patients also completed a Panic Attack Inventory every week to record the frequency, severity, and situations in which panic attacks occurred. In addition they completed the Patient-Rated Anxiety Scale (PRAS; Sheehan, 1986) and the Symptom Checklist-90 (SCL-90; Derogatis et al., 1973) every week.

Sixteen of the 33 patients originally challenged completed the double-blind treatment protocol and participated in repeat challenge studies. Nine of the 17 patients who dropped out were randomized to receive treatment with fluvoxamine. The most common reason for discontinuation was medication side effects. Eight placebo-treated patients dropped out of the study because of lack of efficacy. Thus, the attrition rates were similar in each treatment group. The mean age of the dropout group was 42 (SD = 12) years, and there were 13 women and 4 men. The pretreatment rating data for this group were as follows: mean number of panic

attacks/week = 3.0, SD = 2; mean HRSA score = 19.6, SD = 9; mean HRSD score = 18.5, SD = 9.4; mean PRAS score = 36.9, SD = 22; mean CRAS score = 32.0, SD = 20; and mean SCL-90 score = 66.0, SD = 42. None of the dropout group participated in followup challenge tests.

Yohimbine Test Procedure. The first two challenges were performed after patients had been free of medication for at least 1 week. Before the challenges, 4 out of 16 patients had been treated with a benzodiazepine (3 alprazolam, 1 clonazepam), 1 had received imipramine, and the remaining 11 had been free of medication for 6 or more months. Patients participated in one active yohimbine and one placebo challenge administered in a double-blind, placebo-controlled balanced sequence. Fifty percent of each treatment group started with an active yohimbine day. The mean number of days between the first and second tests was 4 (range = 1-12 days). Yohimbine, 0.4 mg/kg, was administered as an intravenous infusion of 100 ml given over 10 minutes on the active test day, while a solution of normal saline was infused on the placebo day. After the two tests were completed, patients began the medication protocol. After single-blind treatment for a mean of 13 days, the patients continued to receive medication for 8 weeks, at which time the challenges were repeated in the original sequence. The average time between the third and fourth challenge was 3 days (range = 1-7 days).

The challenge studies were conducted in a quiet, well-lighted room on the Clinical Neuroscience Research Unit. The patients were instructed to fast overnight before arriving on the Unit at 8 a.m. on each test day. Patients remained in the fasting state until 2 p.m. on each day. The testing period lasted about 5 hours, with patients lying supine in bed with their heads raised for most of this time. Patient were able to stand to go to the bathroom and also to have blood pressure readings taken. They were not permitted to sleep. An intravenous cannula was inserted in a forearm vein for blood sampling 60 minutes before the yohimbine infusion. Free plasma MHPG and cortisol determinations were made from blood samples that were drawn at 15 minutes before the infusion and then 15, 30, 45, 60, 90, 120, 180, and 240 minutes after the infusion. Samples for cortisol were not taken at 240 minutes, while samples for MHPG were not drawn at the 15-, 30-, or 45-minute time points. Sitting and standing blood pressures and heart rates were measured at about the same times as the blood sampling.

Behavioral rating scales were administered during each test day. Patient-rated visual analog scales (VAS) were used to evaluate 13 different mood states (talkative, happy, drowsy, nervous, sad, calm, depressed, anxious, energetic, fearful, mellow, high, and angry) (Charney and Heninger, 1985). Each mood state could be rated along a 100-mm horizontal line, with 0 representing the absence of the mood state and 100 representing the most ever experienced. Patients marked their rating as a perpendicular line along each scale. The Panic Attack Symptom Scale (PASS; 27 items including 15 *DSM-III-R* panic symptoms; 1 = none, 2 = mild, 3 = moderate, 4 = severe) and the Panic Attack Questionnaire (Charney and Heninger, 1985), both self-report instruments, were also administered. Patients, clinicians, and laboratory staff were unaware of the composition and sequence of the yohimbine infusions. Behavioral VAS and PASS ratings were performed 15 minutes before the intravenous infusion and then 15, 30, 60, 90 120, and 240 minutes after the infusion.

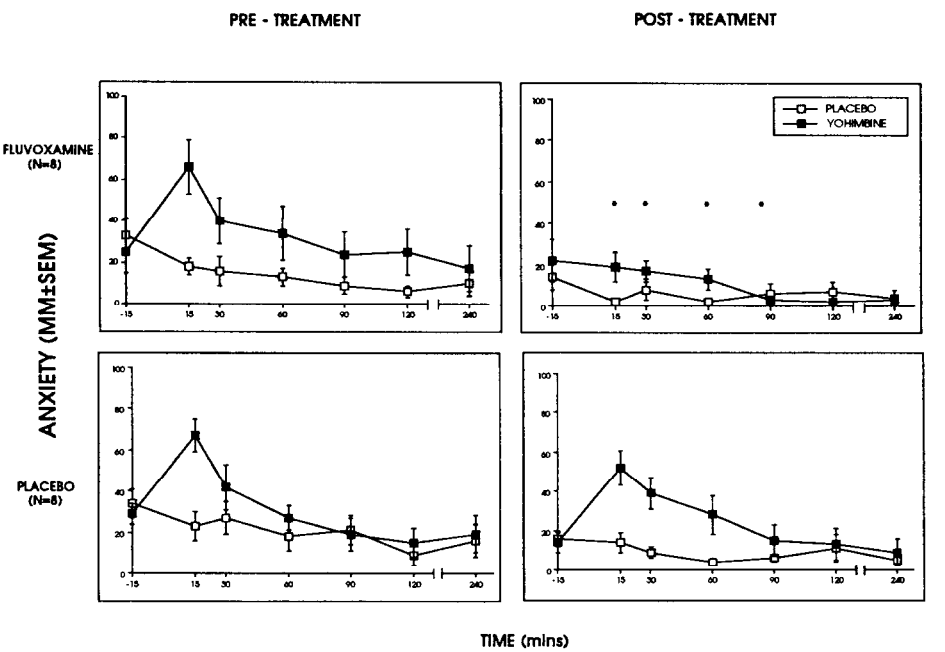
Patients were judged to have a panic attack on test days if they met the following three research criteria for an attack: (1) a 25 mm or more increase in VAS anxiety above baseline in the first 60 minutes following infusion, (2) an increase in four or more *DSM-III-R* symptoms of a panic attack (from PASS) above baseline, and (3) self-report that the attack was similar to their naturally occurring panic attacks (Charney and Heninger, 1985; Woods et al., 1991).

Biochemical Methods. The preparation procedure for the MHPG assay was adapted from the method developed by Dekirmenjian and Maas (1974). Free plasma MHPG measurements were carried out using gas chromatographic mass spectrophotometry. Cortisol levels were determined by radioimmunoassay (Clinical Assays, Inc., Cambridge, MA). Cortisol and MHPG samples were assayed in duplicate and then averaged to give the values that are reported.

Data Analysis. Data were entered into computer files and analyzed using Standard Analysis Systems (SAS) and Biomedical Programs (BMDP). An analysis of variance (ANOVA) with repeated measures was applied to examine the effects of yohimbine on behavioral, biochemical, and autonomic measures before and after treatment with fluvoxamine or placebo. Thus, the three within-group factors included challenge administration (yohimbine vs. placebo) (Y), time within challenge (T), and before and after chronic treatment with either medication condition (PP). The single between-groups factor was treatment condition (fluvoxamine vs. placebo) (G). The central question of the data analysis, whether or not chronic administration of fluvoxamine altered patients' responses to yohimbine, was addressed by evaluating the four-way ANOVA interaction of Y × T × PP × G. Visual inspection of the behavioral data revealed that little change occurred after 120 minutes and that data up to that point approximated a quadratic curve (Fig. 1). Therefore, data up to and including the 120-minute time point were analyzed. ANOVA was performed with and without a quadratic assumption with respect to time, after the method of orthogonal polynomials described by Winer (1971).

Significant interactions that emerged from ANOVA or quadratic ANOVA were analyzed further by the relevant post hoc paired *t* test or Student's *t* test. Thus, for each test day at each time point, the score for each variable was subtracted from the baseline value to generate a change score. Then the change score for the placebo test day was subtracted from that obtained during the yohimbine day to generate a statistic called the net yohimbine effect. Post hoc comparisons were performed on the net yohimbine effect scores where indicated by the preceding ANOVAs.

Fig. 1. Mean anxiety scores (± SEM) following yohimbine and placebo administration before and after fluvoxamine or placebo treatment



A significant difference between groups ($p < 0.05$) on the change in net yohimbine effect from pretreatment to posttreatment is indicated by an asterisk.

Results

Five of eight fluvoxamine patients and five of eight placebo patients had panic attacks following active yohimbine challenge, before treatment (Table 1). There were no panic attacks elicited by placebo challenge. After treatment with fluvoxamine, the total number of panic attacks in response to yohimbine challenge was one in eight patients. In contrast, four of eight patients from the placebo group responded to yohimbine with a panic attack (Table 1). No patients had panic attacks on the placebo challenge day before or after chronic medication treatment. Application of the binomial test (Siegel, 1956) to evaluate the effect of treatment condition on yohimbine-provoked panic attacks within each group was significant ($p < 0.01$) for the fluvoxamine group but not for the placebo group ($p < 0.35$). Analysis between the fluvoxamine and placebo groups for differences in panic responses at the end of chronic treatment was not statistically significant (Fisher's exact test, $p < 0.13$).

Table 1 summarizes the response to treatment for each treatment group. On a variety of outcome measures, there were no significant differences between the fluvoxamine and placebo groups either before or after treatment. A treatment responder was defined as a patient who had experienced an 80% decrease in panic attacks from baseline and had received a rating of 8 (much improved from baseline) or higher on the CGCS. There were six fluvoxamine responders and five placebo responders.

Table 1. Clinical characteristics of patients in relation to treatment

	Placebo treatment ($n = 8$)			Fluvoxamine treatment ($n = 8$)	
Clinical data	Pre-treatment	Post-treatment	***	Pre-treatment	Post-treatment
Total panic attacks/W	5.4 ± 3.4	1.5 ± 3.2*		4.6 ± 2.5	1.1 ± 2.8**
HRSA	21 ± 9	11 ± 8.0*		20 ± 7	12 ± 9*
HRSD	20 ± 6	13 ± 11		19 ± 9	11 ± 7*
CRAS	39 ± 25	16 ± 13*		33 ± 15	17 ± 15**
PRAS	44 ± 36	19 ± 21		45 ± 35	23 ± 34
SCL-90	76 ± 78	39 ± 4.5		70 ± 4.5	47 ± 87
Number of treatment responders	—	5/8		—	6/8
Medication (mg)	—	243 ± 81		—	206 ± 67
CGCS	—	7.8 ± 1.4		—	7.8 ± 2.0

Note. All data are reported as mean ± SD except for responder state, which is expressed as a fraction of 8. HRSA = Hamilton Rating Scale for Anxiety. HRSD = Hamilton Rating Scale for Depression. CRAS = Clinician-Rated Anxiety Scale. PRAS = Patient-Rated Anxiety Scale. SCL-90 = Sympton Checklist-90. CGCS = Clinician Global Change Scale.

*$p < 0.05$, paired t test, pre vs. post.
**$p < 0.01$, paired t test, pre vs. post.
***$p <$ All between groups comparisons, pre vs. pre and post vs. post, were NS.

Inspection of salient VAS variables like anxiety (Fig. 1) and nervousness before each challenge infusion revealed similar baseline levels in each treatment group, irrespective of phase of treatment. There were no significant baseline differences between fluvoxamine and placebo groups on any VAS or PASS variables, before or

after medication treatment. ANOVA with repeated measures was performed on the combined group pretreatment data. There were significant Y × T interactions for 5 of 13 VAS mood symptoms. After yohimbine, patients were more nervous ($F = 7.96$; $df = 5, 70$; $p < 0.0001$), anxious ($F = 9.92$; $df = 5, 70$; $p < 0.0001$) (Fig. 1), and fearful ($F = 8.15$; $df = 5, 70$; $p < 0.0001$), and less happy ($F = 2.95$; $df = 5, 70$; $p < 0.05$) and calm ($F = 3.81$, $df = 5, 70$; $p < 0.01$). There were also significant Y × T interactions for 17 of 27 PASS symptoms. After yohimbine, patients generally experienced more nausea ($F = 3.12$; $df = 5, 70$; $p < 0.05$), perspiration ($F = 7.76$; $df = 5, 70$; $p < 0.0001$), palpitations ($F = 4.3$; $df = 5, 70$; $p < 0.005$), parasthesias ($F = 4.1$; $df = 5, 70$; $p < 0.005$), anorexia ($F = 2.42$; $df = 5, 70$; $p < 0.05$), dizziness ($F = 4.21$; $df = 5, 70$; $p < 0.005$), faintness ($F = 9.47$; $df = 5, 70$; $p < 0.0001$), piloerection ($F = 3.95$; $df = 5, 70$; $p < 0.01$), hot and cold flashes ($F = 6.15$; $df = 5, 70$; $p < 0.0001$), and tremulousness ($F = 18.29$; $df = 5, 70$; $p < 0.0001$). In addition, they reported an increase in lacrimation ($F = 5.49$; $df = 5, 70$; $p < 0.001$), feelings of unreality ($F = 6.04$; $df = 5, 70$; $p < 0.0001$), weakness ($F = 5.76$; $df = 5, 70$; $p < 0.001$), dyspnea ($F = 5.33$; $df = 5, 70$; $p < 0.001$), fear of losing control ($F = 7.6$; $df = 5, 70$; $p < 0.0001$), muscular incoordination ($F = 9.2$; $df = 5, 70$; $p < 0.0001$), and muscle twitching ($F = 9.15$; $df = 5, 70$; $p < 0.0001$). Generally, the peak symptom effects were observed 15 minutes after yohimbine infusion. Table 2 shows the net yohimbine effect for all VAS and PASS variables at this time point.

A comparison of the effects of treatment condition on yohimbine responses was performed by examination of the four-way interaction of Y × T × PP × G for all VAS and PASS variables. Two of the five VAS mood variables significantly affected by yohimbine before treatment differed between treatment groups. Patients in the fluvoxamine group compared with patients in the placebo group reported less anxiety (quadratic ANOVA: $F = 10.10$; $df = 1, 14$; $p < 0.01$) (Fig. 1) and less nervousness (quadratic ANOVA: $F = 4.61$; $df = 1, 14$; $p < 0.05$) (Table 2) in response to yohimbine, after medication treatment. Post hoc Student's t test, performed on the net yohimbine effect after treatment minus the before-treatment net effect, fluvoxamine group vs. placebo, was significant at 15, 30, 60, and 90 minutes ($p < 0.05$) after challenge for self-reported anxiety (Fig. 1), with a marked reduction in the fluvoxamine and not the placebo group. The same comparison for the variable of nervousness was not significant at 15 minutes (Table 2) but was significant ($p < 0.05$) at 60 minutes after the challenge (fluvoxamine: mean = -50 mm, SD = 65; placebo: mean = 13 mm, SD = 48). An analysis of peak (15 minutes after yohimbine) anxiety responses confirmed the above findings. The drop in the peak net yohimbine anxiety response in the fluvoxamine group following treatment was -45 mm (SD = 34) compared with -4 mm (SD = 22) in the placebo-treated group. This comparison was significant ($p < 0.02$) by Student's t test (Table 2, comparison d). ANOVA revealed that 2 of the 17 PASS variables originally affected by yohimbine before treatment were significantly different after fluvoxamine compared with placebo treatment. The fluvoxamine group reported significantly less anorexia (quadratic ANOVA; $F = 5.09$; $df = 1, 14$; $p < 0.05$) and feelings of weakness (quadratic ANOVA; $F = 5.97$; $df = 1, 14$; $p < 0.05$) in response to yohimbine. Post hoc t tests confirmed these findings for weakness only (Table 2, comparison d) at the 15-minute postchallenge time point.

Table 2. Effects of fluvoxamine or placebo treatment on Visual Analog Scale (VAS) mood symptoms and Panic Attack Symptom Scale (PASS) symptoms 15 minutes after yohimbine administration in panic disorder patients

	Net yohimbine effect +			
	Placebo ($n = 8$)		Fluvoxamine ($n = 8$)	
Symptom	**Pretreatment**	**Posttreatment**	**Pretreatment**	**Posttreatment**
VAS				
Talkative	-7 ± 36	-15 ± 22	-21 ± 31	-8 ± 28
Happy	-20 ± 34	-18 ± 31	-22 ± 36	2 ± 14
Drowsy	5 ± 34	-5 ± 14	5 ± 25	-4 ± 12
Nervous	43 ± 28*	30 ± 39	50 ± 48*	8 ± 39
Sad	7 ± 22	6 ± 25	-1 ± 22	0 ± 0
Calm	-23 ± 22*	-28 ± 36	-40 ± 25*	-16 ± 14*b
Depressed	1 ± 8	3 ± 8	2 ± 18	1 ± 14
Anxious	49 ± 25*	40 ± 28*	55 ± 44*	10 ± 36cd
Fearful	39 ± 31*	27 ± 31	31 ± 20*	-6 ± 4.5
Mellow	-18 ± 28	-17 ± 42	-24 ± 39	-18 ± 22
High	9 ± 17	13 ± 34	-6 ± 22	16 ± 28
Energetic	-2 ± 31	-7 ± 12	-8 ± 42	-6 ± 36
Angry	-1 ± 6	1 ± 6	-7 ± 17	0 ± 0
PASS				
Nausea	0.4 ± 0.8	0.5 ± 0.8	0.6 ± 1.1	0.8 ± 0.8*
Urinary frequency	0.0 ± 0.8	0.5 ± 0.8	0.1 ± 1.1	0.3 ± 0.6
Perspiration	0.8 ± 0.8*	0.6 ± 0.8*	2.0 ± 1.1*a	1.0 ± 1.1*
Palpitations	1.0 ± 1.1*	0.8 ± 0.8*	1.4 ± 1.4*	0.6 ± 0.8*
Paresthesias	1.4 ± 1.7	1.1 ± 1.1*	0.9 ± 1.4	0.3 ± 1.1
Restless	-0.1 ± 1.4	0.9 ± 0.8*	0.4 ± 2.2	0.8 ± 1.1
Chest discomfort	0.1 ± 0.6	0.1 ± 0.3	0.1 ± 0.3	0.4 ± 0.6
Choking	0.3 ± 0.8	0.6 ± 0.8*	0.5 ± 0.8	0.5 ± 0.6*
Anorexia	0.4 ± 0.9	1.1 ± 1.1*	0.8 ± 1.7	0.3 ± 0.8
Dizziness	0.5 ± 0.6*	0.8 ± 1.1	1.0 ± 1.4	0.4 ± 0.8
Faintness	0.8 ± 1.4	1.4 ± 0.8*	2.3 ± 1.1*	1.1 ± 1.1*
Tremulousness	2.0 ± 1.1*	18. ± 1.1*	2.1 ± 2.0*	1.0 ± 0.8*
Goosebumps	0.8 ± 1.1	0.5 ± 0.8	1.6 ± 2.0	0.5 ± 0.8
Hot and cold flashes	0.9 ± 0.8*	1.0 ± 1.1*	1.6 ± 1.1*	0.9 ± 0.8*
Lacrimation	1.1 ± 1.4	0.6 ± 1.1	0.8 ± 1.4	0.5 ± 0.8
Rhinorrhea	0.6 ± 1.4	0.4 ± 0.8	0.3 ± 1.1	0.3 ± 0.6
Muscle aches	0.4 ± 1.1	0.1 ± 0.6	0.3 ± 0.8	0.0 ± 0.0
Unreal feelings	1.4 ± 1.4	1.3 ± 1.1*	0.6 ± 1.1	0.5 ± 0.8
Weakness	0.8 ± 1.7	0.8 ± 0.8*	1.9 ± 1.1*a	0.3 ± 0.8cd
Headache	0.3 ± 0.8	0.4 ± 0.6	-0.1 ± 0.3	0.3 ± 0.8
Dyspnea	1.0 ± 1.1*	0.4 ± 0.6	0.9 ± 1.1	0.4 ± 0.8
Fear of death	-0.5 ± 0.8	0.1 ± 0.3	0.3 ± 0.6	-0.1 ± 0.3
Fear of losing control	1.6 ± 1.7*	1.1 ± 0.8*	1.1 ± 1.1*	0.3 ± 0.8
Fear of going crazy	0.4 ± 1.1	0.3 ± 0.6	0.4 ± 0.8	-0.1 ± 0.3
Irritability	0.9 ± 1.1	0.5 ± 0.8	-0.4 ± 0.8	0.3 ± 0.8
Incoordination	1.1 ± 1.1*	0.9 ± 1.1	1.8 ± 1.4*	0.5 ± 0.8b
Muscle twitching	1.0 ± 1.4	0.6 ± 1.1	2.1 ± 1.4*	0.5 ± 0.6*b

+ This statistic is derived as follows: (15 minute post-yohimbine challenge score minus baseline score) minus (15 minute post-placebo challenge score minus baseline score). Means ± SD are reported. Units are on a 0-100 mm scale for VAS. For PASS, 1 = none, 2 = mild, 3 = moderate, 4 = severe. Fifteen of 27 PASS symptoms are *DSM-III-R* panic symptoms. *$p < 0.05$, paired t test, 2-tailed, for net yohimbine effect.

a. $p < 0.05$, Student's t test, 2-tailed, for net yohimbine effect pretreatment, fluvoxamine group vs. placebo group.
b. $p < 0.05$, net yohimbine effect posttreatment minus pretreatment, paired t test, 2-tailed.
c. $p < 0.01$, net yohimbine effect posttreatment minus pretreatment, paired t test, 2-tailed.
d. $p < 0.05$, net yohimbine effect posttreatment minus pretreatment, fluvoxamine group vs. placebo group, Student's t test, 2-tailed.

The effects of yohimbine on physiologic measures, before treatment, were similar in both groups. An ANOVA on the pretreatment data set revealed significant Y × T interactions for sitting ($F = 3.73$; $df = 5, 70$; $p < 0.01$) and standing ($F = 5.9$; $df = 5, 70$; $p < 0.0001$) systolic and sitting ($F = 2.31$; $df = 5, 70$; $p < 0.05$) and standing ($F = 4.69$; $df = 5, 70$, $p < 0.001$) diastolic blood pressure. Table 3 presents the magnitude of these pressor effects of yohimbine. Evaluation of the effects of treatment on yohimbine-induced physiologic changes was performed by examination of the four-way interaction of Y × T × PP × G. This was significant for sitting systolic blood pressure (quadratic ANOVA: $F = 11.75$; $df = 1, 14$; $p < 0.01$). However, the significance of the drop in the net yohimbine effect at the 15-minute postchallenge time point for this variable following chronic fluvoxamine treatment (fluvoxamine group: mean = -16 mmHg, SD = 17; placebo group: mean = -1 mmHg, SD = 31) was not confirmed by Student's t test ($p < 0.3$) (Table 3).

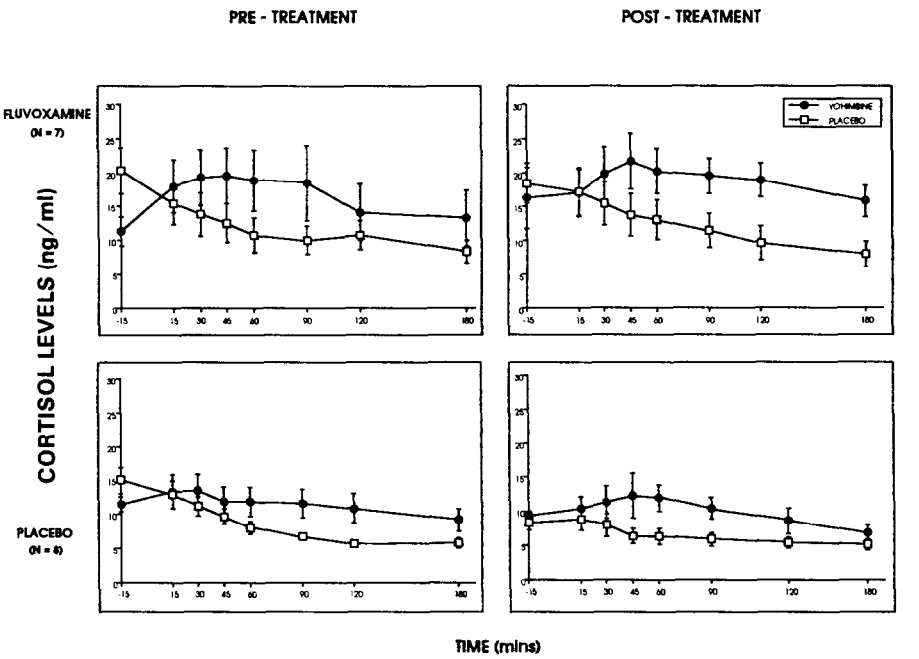
Table 3. Effects of fluvoxamine or placebo treatment on physiologic variables 15 minutes after yohimbine administration in panic disorder patients

Physiologic variable	Net yohimbine effect +			
	Placebo ($n = 8$)		Fluvoxamine ($n = 8$)	
	Pretreatment	Posttreatment	Pretreatment	Posttreatment
Systolic blood pressure (sitting)	15 ± 17*	14 ± 17*	21 ± 14*	6 ± 17[a]
Systolic blood pressure (standing)	16 ± 17*	13 ± 8*	27 ± 14*	6 ± 28
Diastolic blood pressure (sitting)	4 ± 14	3 ± 6	10 ± 8*	7 ± 8
Diastolic blood pressure (standing)	7 ± 14	9 ± 12	10 ± 14	-1 ± 8
Heart rate (sitting)	5 ± 6	9 ± 14	4 ± 14	2 ± 12
Heart rate (standing)	8 ± 8	3 ± 12	1 ± 12	1 ± 12

+Net yohimbine effect in mmHg ± SD.
*$p < 0.05$, paired t test, 2-tailed, for the net yohimbine effect.
[a]$p < 0.05$, paired t test, 2-tailed, for pretreatment minus posttreatment net yohimbine effect.

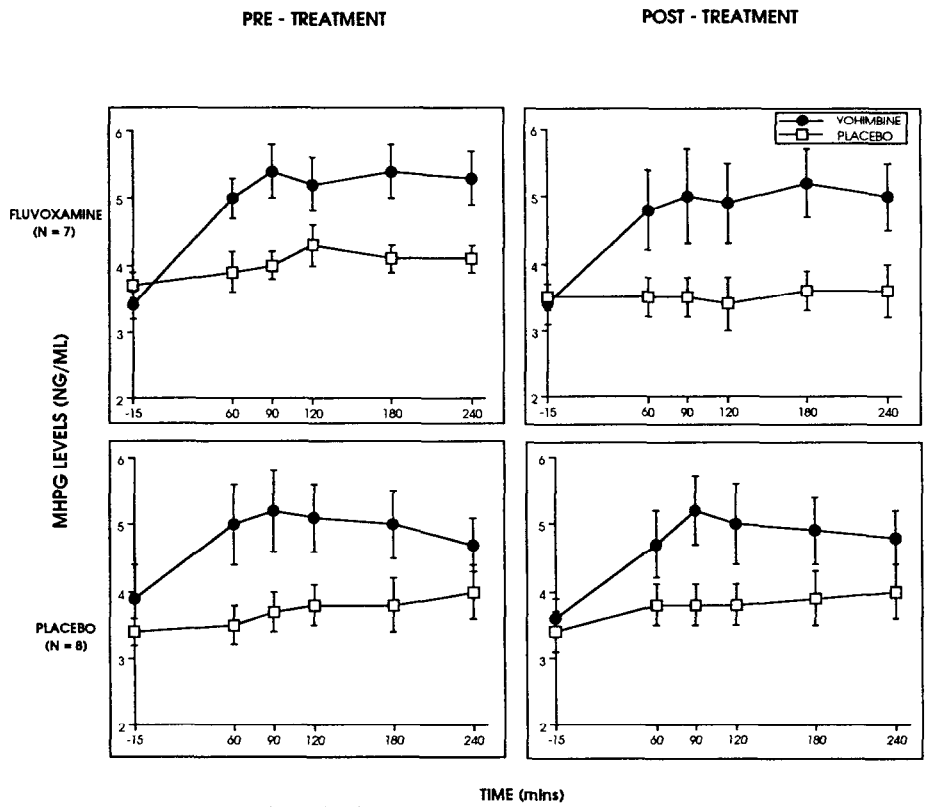
At baseline, analysis of MHPG and cortisol levels revealed no significant differences between fluvoxamine and placebo groups, before or after treatment (Figs. 2 and 3). The Y × T interaction was highly significant for both cortisol ($F = 15.42$; $df = 6, 78$; $p < 0.0001$) and MHPG ($F = 15.54$; $df = 5, 65$; $p < 0.0001$). Figs. 2 and 3 present the time courses of cortisol and MHPG responses to yohimbine and placebo challenge. Finally, a comparison of the effect of treatment on yohimbine-induced MHPG and cortisol changes between groups was performed. The Y × T × PP × G interaction was found to be not significant for MHPG ($F = 0.56$; $df = 5, 65$; $p =$ NS) or cortisol ($F = 1.33$; $df = 6, 78$; $p =$ NS).

Fig. 2. Mean plasma cortisol levels (± SEM) following yohimbine and placebo administration in relation to treatment with fluvoxamine or placebo



An analysis was performed comparing the responses to the posttreatment yohimbine test of treatment nonresponders ($n = 5$) to the responses of placebo ($n = 5$) or fluvoxamine ($n = 6$) treatment responders. In relation to panic attacks, none of six fluvoxamine responders experienced a yohimbine-induced panic attack compared with three of five treatment nonresponders (Fisher's exact test, $p < 0.06$). In contrast, two of five placebo responders had yohimbine panic attacks, and comparison with the nonresponders was not significant (Fisher's exact test, $p < 0.4$). With respect to the net yohimbine effect on VAS anxiety at 15 minutes postinfusion, there were no significant differences between fluvoxamine responders (mean = 16.7 mm, SD = 33) and treatment nonresponders (mean = 15.6 mm, SD = 47; Student's t test, $t = 0.1$, $df = 9$, $p < 0.97$) or between placebo responders (mean = 43.4 mm, SD = 20) and treatment nonresponders ($t = -1.3$, $df = 8$, $p < 0.3$). MHPG responses to yohimbine were also examined across the above groups. The MHPG increase from baseline to the 90-minute postinfusion time point was calculated for each patient, for the posttreatment yohimbine test. There were no significant differences between the MHPG responses of the nonresponders ($n = 4$; mean = 1.8 ng/ml, SD = 1.6) and the MHPG responses of the fluvoxamine responders ($n = 6$; mean = 1.7 ng/ml, SD = 1.4; $t = 0.12$, $df = 8$, p = NS), nor in comparison to the placebo responders ($n = 5$; mean = 1.5 ng/ml, SD = 0.9; $t = 0.39$, $df = 7$, p = NS).

Fig. 3. Mean plasma MHPG levels (± SEM) following yohimbine and placebo administration in relation to treatment with fluvoxamine or placebo.



MHPG = 3-methoxy-4-hydroxyphenylglycol.

Discussion

The findings of the study can be summarized as follows: There was a greater numerical reduction in the yohimbine-induced panic attack rate observed in the patients who completed 8 weeks of fluvoxamine treatment compared with those who were treated with placebo. As determined by self-report measures, patients treated with fluvoxamine experienced a significant reduction in yohimbine-induced anxiety and nervousness following treatment. In contrast, these findings were not observed in the placebo-treated group. There was little effect of treatment condition on somatic symptom responses to yohimbine in either group, suggesting that fluvoxamine had selectively diminished the subjective anxiety component of yohimbine-induced panic. There were no significant alterations in physiologic and biochemical responses to yohimbine (MHPG and cortisol) as a result of either treatment condition. Clinical improvement was similar in both treatment groups. Fluvoxamine treatment responders had a 0% (0/6) yohimbine-induced panic rate following treatment compared with treatment nonresponders (placebo and fluvoxamine nonresponders), who had a 60% (3/5) rate.

Sampling (Type I) error could have accounted for the findings of the present study, especially in view of the high attrition rate and the small number of patients who completed the study. The fact that the fluvoxamine patient group tended to have greater PASS symptom responses to yohimbine before treatment (Table 2) also raises the possibility that this group represents a different subpopulation of panic disorder patients that is not strictly comparable to the placebo group. The data should only be seen as providing preliminary evidence of a fluvoxamine effect on yohimbine-induced anxiety until replication work is done with a larger sample to exclude or confirm this important possibility.

A number of other factors, including passage of time, differences in patient characteristics, pharmacokinetic interactions, and awareness of medication vs. placebo condition due to side effects could potentially account for the present data. If the blunting by fluvoxamine of the VAS effects of yohimbine were simply an effect of time, we would have expected to observe similar behavioral changes to the yohimbine in the placebo treatment group as well. Furthermore, there did not seem to be differences between the two treatment groups in clinical profile before and after treatment that might explain the behavioral finding (Table 1). A pharmacokinetic interaction between yohimbine and fluvoxamine could have been responsible for the observed effects. If this had occurred, however, the other nonbehavioral effects of yohimbine should also have been diminished. The fact that the MHPG and cortisol signals were similar in the placebo- and fluvoxamine-treated patients before and after treatment suggests that similar amounts of yohimbine were crossing the blood-brain barrier, independent of treatment condition. The impact of medication side effects "unblinding" the fluvoxamine treatment could potentially have influenced the level of subjective anxiety a patient reported during the yohimbine test. A patient who believed that he or she was in the active medication condition might have been expected to experience less anticipatory anxiety before the yohimbine test and less anxiety during the test. Inspection of Fig. 1 reveals that baseline levels of VAS anxiety were similar in both treatment groups before test days, before and after treatment, a finding that tends to argue against knowledge of treatment/placebo condition as an explanatory factor.

It is possible that a 5-HT effect of fluvoxamine that was unrelated to the NE system could fully explain our findings. There are little data on the effect of yohimbine administration on 5-HT function. However, yohimbine has a significant affinity for the 5-HT1a receptor (Fozard et al., 1987) and, when given in high doses to animals (0.8-1.6 mg/kg, i.v.), may function as an agonist at this site to suppress central NE neurotransmission (McCall et al., 1991). The biochemical and physiologic data in the present study suggest that yohimbine activated rather than suppressed NE transmission. Therefore, while it is possible that the behavioral effects seen in our study were the result of reversal by fluvoxamine of the 5-HT effects of yohimbine, the above preclinical data suggest otherwise. Clinical studies have documented anxiety symptom exacerbations to the 5-HT challenge agents fenfluramine (Targum and Marshall, 1989) and meta-chlorophenylpiperazine (Charney et al., 1987; Kahn et al., 1988*b*). It has been observed (Targum, 1992) that these clinical changes resemble anticipatory anxiety rather than true panic. Thus, the

5-HT system may play an important role in mediating anticipatory anxiety. In this context, fluvoxamine treatment may be influencing yohimbine-induced anxiety by inhibiting anticipatory fear mechanisms. However, the finding of similar baseline levels of anxiety before and after treatment in both treatment groups (Fig. 1) tends to argue against this hypothesis.

If it is assumed that a specific interaction between 5-HT and NE systems accounts for the findings of the present study, there are a variety of mechanisms by which the putative interaction(s) could occur. Preclinical studies suggest that 5-HT/NE interactions occur at the level of the locus ceruleus (LC) with the dorsal raphe nuclei (Leger et al., 1979; Baraban and Aghajanian, 1980). Segal (1979) found that direct application of 5-HT to LC neurons led to a tonic inhibition of electrical activity, while recent work argues for phasic 5-HT inhibition of LC function mediated by an excitatory amino acid (EAA) pathway from the nucleus paragigantocellularis (Aston-Jones et al., 1991), possibly through a 5-HT1a receptor. In this context, it is remarkable that fluvoxamine has been found to alter EAA receptor mRNA expression throughout the brain (Havard et al., 1991). Another potential mechanism within the LC involves corticotropin releasing factor (CRF), which is known to increase LC firing in rats (Valentino and Foote, 1988). Antidepressants like desipramine, mianserin, phenelzine, and sertraline (Valentino et al., 1990; Melia and Duman, 1991) exert a functional antagonism to the effects of CRF on the LC. Other possible mechanisms include down-regulation by fluvoxamine of tyrosine hydroxylase levels within the LC (Nestler et al., 1990) and down-regulation of substance P, which can cause excitation of LC neurons (Riley et al., 1991).

In relation to the current study, it would be premature to designate a location for the presumed 5-HT/NE interaction(s) contributing to the observed effects. To the extent that MHPG provides some measure of LC activity, we did not find an effect of fluvoxamine on baseline MHPG or evoked MHPG response, suggesting that fluvoxamine had little effect at the level of the LC. However, a recent study (W.K.G., unpublished data) in fluvoxamine-treated obsessive-compulsive patients found a modest reduction in cerebrospinal fluid MHPG levels after treatment, calling into question the accuracy of peripheral MHPG measures in the present study. Cortisol levels and physiologic variables also were not significantly altered by fluvoxamine more than by placebo. Since we found significant effects of fluvoxamine on the mood component of the yohimbine response, and little effect on the somatic symptom component, it is tempting to postulate that fluvoxamine is modulating NE stimulation at a cortical level through one or more of the interactive mechanisms discussed. It is possible, however, that the modulating effects of fluvoxamine on the LC or subcortical regions are subtle and not well measured by indirect markers like plasma MHPG or vital signs. The preclinical work of Aston-Jones et al. (1991) is relevant here as it supports the notion of phasic rather than tonic 5-HT inhibition of LC function. Accordingly, other indices of LC function like cerebral blood flow may provide a more direct and sensitive measure of cerebral NE function in response to yohimbine (Woods et al., 1988).

The role of the NE system with respect to clinical improvement of panic disorder requires further evaluation. From the present data, however, it seems that placebo

medication responders, who completed the study, improved through mechanisms that were independent of the NE system or did not have a sufficiently potent NE effect to alter baseline MHPG or any of the behavioral, physiological, or biochemical responses to yohimbine. A favorable response to fluvoxamine treatment was associated with a reduction in yohimbine-induced panic attacks but not in the yohimbine-induced MHPG signal, suggesting that clinical improvement to fluvoxamine was associated with partial modulation of NE functioning.

The current investigation was a preliminary evaluation of the clinical significance of 5-HT/NE interactions in relation to human anxiety. Further study of the biological relationships between the 5-HT and NE systems in the anxiety disorders may provide more precise insights into the pathophysiology of human anxiety and may lead to better explanations for the mechanisms of action of the diversity of pharmacologic and behavioral treatments that are becoming available for these conditions.

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