Pharmacology, Biochemistry and Behavior 89 (2008) 623–626

Contents lists available at ScienceDirect

Pharmacology, Biochemistry and Behavior

journal homepage: www.elsevier.com/locate/pharmbiochembeh

Confirmation of antidepressant potential of the selective β3 adrenoceptor agonist amibegron in an animal model of depression

David H. Overstreet [a,*], Jeanne Stemmelin [b], Guy Griebel [b]

[a] Department of Psychiatry & Center for Alcohol Studies, University of North Carolina at Chapel Hill, Chapel Hill, NC 27599-7178, USA
[b] Sanofi-Aventis CNS Research, 31 Ave PV-Couturier, 92220 Bagneux, France

ARTICLE INFO

Article history:
Received 17 December 2007
Received in revised form 1 February 2008
Accepted 19 February 2008
Available online 26 February 2008

Keywords:
Amibegron
β3 adrenergic receptor
FSL rat model of depression
Forced swim test

ABSTRACT

The involvement of the noradrenergic system, particularly the β1 and β2 receptors, in depressive disorders has been frequently shown. Recently, however, it has been shown that the β3 receptor may also contribute since amibegron (SR58611A), a selective β3 receptor agonist, has antidepressant-like effects. The present experiment sought to confirm the antidepressant potential of amibegron by studying its effects in an animal model of depression, the Flinders Sensitive Line (FSL) rat. The FSL rat is innately highly immobile in the forced swim test and exhibits a decrease in immobility after chronic, not acute antidepressant treatment. FSL rats were treated for 14 consecutive days with amibegron (0.3, 1.0, or 3.0 mg/kg), fluoxetine (5 mg/kg) or desipramine (5 mg/kg) as positive controls, and vehicle, while the control strain, the Flinders Resistant Line (FRL) rats, was given either vehicle or 1.0 mg/kg amibegron. About 23–25 h after the last injection the rats were tested in the forced swim test. All doses of amibegron and the two active controls, fluoxetine and desipramine, significantly reduced immobility in the FSL rats. Thus, amibegron had a selective antidepressant-like effect in this study, confirming its antidepressant potential.

1. Introduction

A considerable literature exists on the involvement of the noradrenergic system in general, and β1 and β2 adrenergic receptors in particular, in depressive disorders (Holoubek et al., 2004; Millan, 2006; Ressler and Nemeroff, 2000). Much less has been known about the potential involvement of the β3 adrenergic receptor (Strosberg, 1997). However, the availability of amibegron (SR56811A) and other selective β3 adrenergic receptor agonists (Bianchetti and Manara, 1990; Manara and Bianchetti, 1990), has enabled a fuller testing of its behavioral effects. An earlier study showed that amibegron acted like an antidepressant in several models (Simiand et al., 1992). More recently, two separate studies have reported that amibegron has antidepressant-like effects in the forced swim test, using a modification of the Porsolt acute method (Consoli et al., 2007; Stemmelin et al., 2008). Moreover, both studies reported that amibegron was active in several animal tests reflective of anxiety (Consoli et al., 2007; Stemmelin et al., 2008). The present experiment sought to provide confirmation of amibegron's antidepressant-like effects by using an animal model of depression.

The FSL rat has been regarded as a genetic animal model of depression because it exhibits several behavioral abnormalities resembling those seen in depressed individuals, such as elevated REM sleep (Overstreet et al., 2005; Schiller et al., 1992). Moreover, the rat and the paradigm used are able to detect potential antidepressants and reject others that have been false positives, such as amphetamine and scopolamine (Overstreet et al., 1995). Early on it was shown that the FSL rat exhibits a reduction in immobility after chronic treatment with desipramine or sertraline, but not after the acute Porsolt treatment (Pucilowski and Overstreet, 1993). Thus, the FSL rat model is a very efficient system for detecting antidepressant-like effects. More recently, it has been determined that the FSL rat exhibits a reduction in social interaction behavior (Overstreet et al., 2004), suggesting that it may be more anxious than the FRL rat in this task. Because the FSL rat exhibits both depressed-like and anxious-like behavior thus resembling many depressed individuals (Himmelhoch et al., 2001), it is possible to test drug effects on both measures. In this study, we have performed exactly that.

2. Methods

2.1. Animals

The 80-day old FSL and FRL rats were selected from breeding colonies maintained at the University of North Carolina Bowles Center for Alcohol Studies. They were housed in groups of three in temperature- and humidity-controlled rooms under a 12:12 light: dark cycle (lights on 0700–1900). Rats were randomly divided into six (FSL) or two (FRL) groups and then given the treatments described below. Fewer FRL groups were used because previous evidence indicated that the FRL rats, which exhibit a relatively low degree of immobility, do not exhibit decreases in immobility following antidepressant treatments (Overstreet, 2002; Overstreet et al., 2005).

* Corresponding author. Bowles Center for Alcohol Studies, 3011 Thurston-Bowles Bldg., CB #7178 University of North Carolina at Chapel Hill, Chapel Hill, NC 27599-7178, USA. Tel.: +1 919 966 0505; fax: +1 919 966 5679.
E-mail address: dhover@med.unc.edu (D.H. Overstreet).

0091-3057/$ – see front matter © 2008 Elsevier Inc. All rights reserved.
doi:10.1016/j.pbb.2008.02.020

2.2. Treatments

The following six treatment groups were established for the FSL rats: Veh — isotonic saline; amibegron — 0.3, 1.0, 3.0 mg/kg; FLX — fluoxetine (5 mg/kg); DMI — desipramine (5 mg/kg). The FRL rat groups were treated with Veh or amibegron (1 mg/kg). The rats were injected intraperitoneally (i.p.) once daily for 14 consecutive days between 09.00 and 11.00 h.

2.3. Behavioral tests

Approximately 22 h after the last treatment, rats with the same treatment and similar body weights were placed in a square test arena (60×60 cm, marked with 16 15×15 cm squares on the floor) for the testing of social interaction. The amount of time spent in social interaction (grooming, licking, sniffing and crawling over or under) was recorded during a 5-min session by an experienced observer who was blind to the treatment condition. This measure provides one index of anxiety-like behavior, with more "anxious" rats spending less time in social interaction. The total number of lines crossed during the session provided a measure of general activity.

The swim tank was 18 cm in diameter and 40 cm tall. The tank was filled with enough 25 °C water so the rat could not touch bottom with its hindpaws. The rat was placed in the swim tank for a single 5-min session 23–25 h after the last treatment and the seconds of immobility were scored by an observer blind to the treatment condition and rat strain being tested (Overstreet, 1993; Zangen et al., 1997).

2.4. Statistical analyses

The data for the four measures were summarized into mean±S.E.M. for each of the 10 treatment groups. Graphical representations of the findings were compiled using Prism software. Initially, the data for each measure were subjected to one-way ANOVAs. If the ANOVA revealed significant group differences, follow-up Tukey's tests were carried out to elucidate the pattern of group differences. The GBstat software package was used for the statistical analyses. Two-way ANOVAs were conducted to compare treatment responses in the FSL and FRL rats.

3. Results

All treatments were effective in reducing the exaggerated swim test immobility of the FSL rats ($F[7,56]=10.90$, $p<0.0001$), as illustrated in Fig. 1. Importantly, the values of the various drug-treated FSL groups were similar to that of the FRL rats given vehicle (Fig. 1). However, the swim test immobility of the FRL rats given 1 mg/kg amibegron was not significantly different from the scores of the FRL rats given vehicle. Consequently, the 2-way ANOVA revealed highly significant strain ($F[1,28]=28.63$, $p<0.0001$) and interaction ($F[1,28]=27.11$, $p<0.0001$) effects. The putative antidepressant amibegron had antidepressant-like effects only in the animal model of depression that exhibits innate elevated immobility.

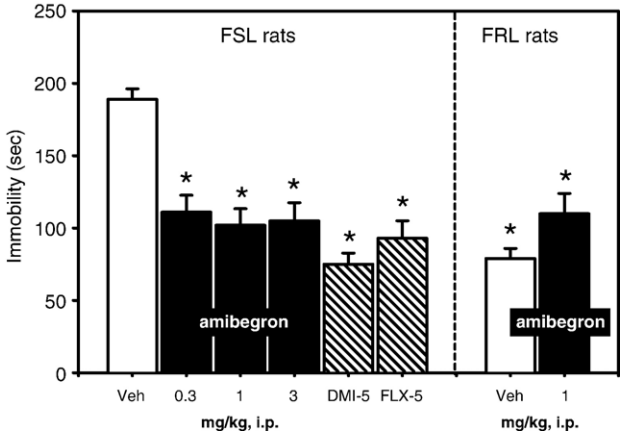

Fig. 1. The effects of amibegron, desipramine, or fluoxetine on immobility in the forced swim test. Rats were given their appropriate treatments i.p. once daily for 14 consecutive days and the test was carried out about 22–25 h after the last treatment. Data represent mean+S.E.M. *$p<0.05$, all treatments different from FSL-Veh. Abbreviations: FSL Flinders Sensitive Line; FRL Flinders Resistant Line; DMI desipramine; FLX fluoxetine; Veh vehicle (isotonic saline).

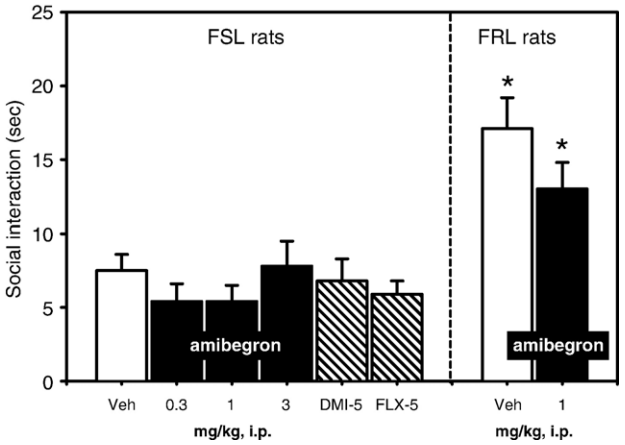

Fig. 2. The effects of amibegron, desipramine, and fluoxetine on social interaction behavior. The test was conducted about 22 h after the last of 14 consecutive daily treatments. Data represent mean+S.E.M. *$p<0.05$, all FRL different from all FSL. See Fig. 1 for abbreviations.

Although the FSL rats also exhibit an innate reduction in social interaction behavior, none of the drug treatments altered this behavior, as illustrated in Fig. 2. However, because the FRL rats engage in more social interaction behavior than the FSL rats, the overall 1-way ANOVA was highly significant ($F[7,56]=8.26$, $p<0.0001$). Additional testing with a 2-way ANOVA confirmed the strain difference ($F[1,28]=29.26$, $p<0.0001$) and the lack of drug effect ($F[1,28]=3.84$, $p>0.05$). Thus, the abnormally low social interaction behavior of the FSL rat was not corrected by the drugs used in this study.

The present finding partially confirms the suggestion that locomotor activity is independent from social interaction behavior. While there is a clear strain difference for the latter, there is none for the former, as illustrated in Fig. 3. Not only is there no strain difference, but there is also no drug effect ($F[7,56]=1.18$, $p>0.05$). Thus, the behavior that is relatively normal in the FSL rat is not affected by the known antidepressants or putative antidepressant.

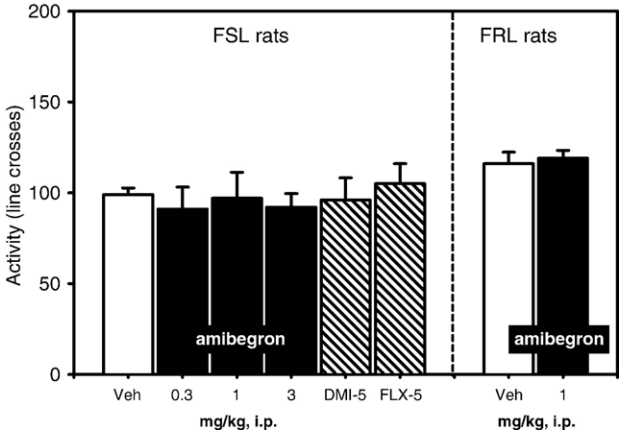

Fig. 3. The effects of amibegron, desipramine, and fluoxetine on locomotor activity (line crosses). Behavior was recorded simultaneously with social interaction. Data represent mean+S.E.M. See Fig. 1 for abbreviations.

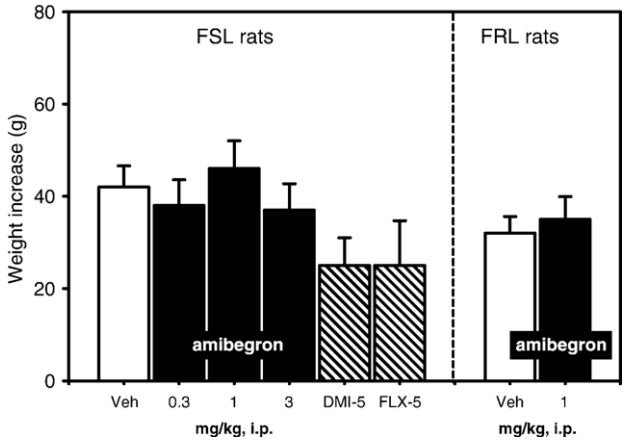

Fig. 4. The effects of amibegron, desipramine, and fluoxetine on increase in body weight. Weights were taken at Day 1 and Day 14 and the increase in body weight is plotted. Data represent mean+S.E.M. See Fig. 1 for abbreviations.

Although there is a trend for rats treated with FLX or DMI to gain less weight than the other rats, as illustrated in Fig. 4, there are no significant group differences in the degree of body weight change ($F[7,56]=1.59$, $p>0.05$). Thus, amibegron does not appear to have any negative effects on appetite that could compromise its use as an antidepressant.

4. Discussion

The present findings have confirmed the antidepressant potential of amibegron by demonstrating that all doses used reduced the exaggerated immobility (Fig. 1). In fact, the exaggerated swim test immobility of the FSL rats was reduced as much by amibegron as by the positive controls, desipramine and fluoxetine. Similar results were reported by Stemmelin et al. (2008) in the acute Porsolt model as well as the chronic mild stress model, and by Consoli et al. (2007) and Simiand et al. (1992) using the forced swim test. However, the present findings include one important difference from the previous studies. The dose of 0.3 mg/kg amibegron was effective in our study but not in the earlier studies (Consoli et al., 2007; Simiand et al., 1992; Stemmelin et al., 2008). This observation is consistent with other evidence that the FSL rat model is more sensitive to the effects of antidepressants than the acute models (Overstreet et al., 2005; Porsolt et al., 1977; Pucilowski and Overstreet, 1993). Indeed, both an older and a newer review of the forced swim test model indicated that lower doses were possible when chronic treatment was employed (Borsini and Meli, 1988; Cryan et al., 2005). Another important distinction between the earlier and present swim test paradigms relates to the time between drug treatment and behavioral test. The earlier studies tended to test the rat about 60 min after the last drug treatment, whereas we tested the rat after 23–25 h. Our rationale is that chronic treatment will lead to adaptive changes that will support a behavioral change outlasting any acute effects.

In any case, it is clear that amibegron has antidepressant potential because the exaggerated swim test immobility of the FSL rats is reduced following chronic treatment.

Because of the very low social interaction behavior and the exaggerated forced swim test immobility, the FSL rat appears to be an appropriate model for the many depressed individuals with comorbid anxiety (Himmelhoch et al., 2001). However, the anxiety status of the FSL rat is unclear. The FSL and FRL rats do not differ in the elevated plus maze, another index of anxiety-like behavior (Schiller et al., 1991; Overstreet et al., 1995). Comparison of the two strains on other behavioral tasks reflective of anxiety has not been conducted as yet. Therefore, the degree of anxiety-related behavior in the FSL rat is unclear.

Despite the uncertainty about the degree of anxiety in the FSL rats, it is clear that they do exhibit very low levels of social interaction behavior and would thus be regarded as being anxious in this task (File and Seth, 2003). Furthermore, none of the treatments induced a change in social interaction behavior. This finding is at odds with previous reports showing anxiolytic effects of amibegron (Consoli et al., 2007; Stemmelin et al., 2008). Most of these earlier tests involved acute administration of amibegron shortly before the test, whereas the present study employed a chronic treatment protocol with testing carried out 22 h after the last treatment. In addition, previous work with the FSL rats showed anxiolytic effects of desipramine and fluoxetine in the social interaction test (Overstreet et al., 2004; Overstreet and Griebel, 2004). We should not conclude, therefore, that amibegron does not have anxiolytic effects. Rather, the conditions of the present experiment were not appropriate to elicit the anxiolytic effects of amibegron.

The lack of effect of amibegron on locomotor activity is entirely consistent with previous reports (Consoli et al., 2007; Stemmelin et al., 2008). Thus, the antidepressant effects of amibegron in this and other studies and the anxiolytic effects in other studies are selective effects. The lack of changes in locomotor activity suggests that amibegron does not have any behaviorally toxic effects. The similar weight change seen in this study is consistent with that conclusion as is the many studies carried out by Stemmelin et al. (2008). Thus, amibegron appears to be a safe β3 adrenergic receptor agonist with strong antidepressant potential.

The neurochemical mechanisms underlying amibegron's antidepressant-like effects cannot be elucidated solely on the basis of the present findings. However, a comparison of the neurochemical effects of amibegron with the neurochemical pathology of the FSL rats reveals some intriguing possibilities. A key difference between the two rat strains is that the FSL rats exhibit elevated REM sleep, just as do depressed individuals Benca et al., 1996; Shiromani et al., 1988). And amibegron reduced REM sleep (Stemmelin et al., 2008), as did antidepressants (Sharpley and Cowen, 1995). The effects on REM sleep could be mediated by increasing serotonergic tone (Bakalian and Fernstrom, 1990) and amibegron and other β-adrenergic agonists increase serotonin or its precursor tryptophan (Claustre et al., 2007; Lenard et al., 2003). Interestingly, the FSL rats have abnormal serotonin function (Zangen et al., 1997), including a reduction in serotonin synthesis (Hasegawa et al., 2006), that is restored following chronic antidepressant treatment (Zangen et al., 1997). Thus, amibegron might be having its antidepressant-like effects in the FSL rats by normalizing their serotonergic tone. Alternatively, amibegron might be working by normalizing the abnormal noradrenergic tone in the FSL rats (Claustre et al., 2007; Zangen et al., 1999). Further conjoint behavioral and neurochemical studies in the FSL rats with amibegron may help resolve this issue.

References

Bakalian MJ, Fernstrom JD. Effects of L-tryptophan and other amino acids on electroencephalographic sleep in the rat. Brain Res 1990;528:300–7.

Benca RM, Overstreet DH, Gilliland MA, Russell D, Bergmann BM, Obermeyer WH. Increased basal REM sleep but no difference in dark induction or light suppression of REM sleep in Flinders Rats with cholinergic supersensitivity. Neuropsychopharmacology 1996;15:45–51.

Bianchetti A, Manara L. In vitro inhibition of intestinal motility by phenylethanolaminotetralines: evidence of atypical beta-adrenoceptors in rat colon. Br J Pharmacol 1990;100:831–9.

Borsini F, Meli A. Is the forced swim test a suitable model for revealing antidepressant activity? Psychopharmacology 1988;94:147–60.

Claustre Y, Leonetti M, Sarhan M, Santucci V, Bougault I, Desvignes C, et al. Effects of β3-adrenoceptor agonist SR58611A (amibegron) on serotonergic and noradrenergic transmission in the rodent: relevance to its antidepressant/anxiolytic-like profile. Program no. 170.15. Neuroscience Meeting PlannerSan Diego, CA: Society for Neuroscience; 2007.

Consoli D, Leggio GM, Mazzola C, Micale V, Drago F. Behavioral effects of the β3 adrenoceptor agonist SR58611A: is it the putative prototype of a new class of antidepressant/anxiolytic drugs? Eur J Pharmacol 2007;573:139–47.

Cryan JF, Valentino RJ, Lucki I. Assessing substrates underlying the behavioral effects of antidepressants using the modified rat forced-swimming test. Neurosci Biobehav Rev 2005;29:547–69.

File SF, Seth P. A review of 25 years of the social interaction test. Eur J Pharmacol 2003;463:35–53.

Hasegawa S, Nishi K, Watanabe A, Overstreet DH, Diksic M. Brain 5-HT synthesis in the Flinders Sensitive Line rat model of depression: an autoradiographic study. Neurochem Int 2006;48:358–66 [Jan 18 Electronic publication PMID: 16377459].

Himmelhoch J, Levine J, Gershon S. Historical overview of the relationship between anxiety disorders and affective disorders. Depress Anxiety 2001;14:53–66.

Holoubek G, Noldner M, Treiber K, Muller WE. Effect of chronic antidepressant treatment on beta-receptor coupled signal transduction cascade. Which effect matters most? Pharmacopsychiatry 2004;37(suppl 2):S113–9.

Lenard NR, Gettys TW, Dunn AJ. Activation of beta2- and beta3-adrenergic receptors increases brain tryptophan. J Pharmacol Exp Ther 2003;305:653–9.

Manara L, Bianchetti A. Further heterogeneity of the beta-adrenoceptor. The phenylethanolaminotetralines: new selective agonists for atypical beta-adrenoceptors. Trends Pharmacol Sci 1990;11:229–30.

Millan M. Multi-target strategies for the improved treatment of depressive states: conceptual foundations and neuronal substrates, drug discovery and therapeutic application. Pharmacol Ther 2006;110:135–370.

Overstreet DH. The Flinders Sensitive Line rats: a genetic animal model of depression. Neurosci Biobehav Rev 1993;17:51–68.

Overstreet DH. Behavioral characteristics of rat lines selected for differential hypothermic responses to cholinergic or serotonergic agonists. Behav Genet 2002;32:335–48.

Overstreet DH, Griebel G. Antidepressant-like effects of CRF_1 receptor antagonist SSR125543 in an animal model of depression. Eur J Pharmacol 2004;497:49–53.

Overstreet DH, Pucilowski O, Rezvani AH, Janowsky DS. Administration of antidepressants, diazepam and psychomotor stimulants further confirms the utility of Flinders Sensitive Line rats as an animal model of depression. Psychopharmacology 1995;121:27–37.

Overstreet DH, Keeney A, Hogg S. Antidepressant effects of citalopram and CRF receptor antagonist CP-154,526 in a rat model of depression. Eur J Pharmacol 2004;492:195–201.

Overstreet DH, Friedman EF, Mathe' AM, Yadid G. The Flinders Sensitive Line rat: a selectively bred putative animal model of depression. Neurosci Biobehav Rev 2005;29(4–5):739–59 [Epub 2005 Apr 22].

Porsolt R, Le Pichon, Jalfre M. Depression. A new animal model sensitive to antidepressant treatments. Nature 1977;266:730–2.

Pucilowski O, Overstreet DH. Effect of chronic antidepressant treatment on responses to apomorphine in selectively bred rat strains. Brain Res Bull 1993;32:471–5.

Ressler KJ, Nemeroff CB. Role of serotonergic and noradrenergic systems in the pathophysiology of depression and anxiety disorders. Depress Anxiety 2000;12:1098–106.

Schiller GD, Daws LC, Overstreet DH, Orbach J. Absence of anxiety in an animal model of depression with cholinergic supersensitivity. Brain Res Bull 1991;26:443–7.

Schiller GD, Pucilowski O, Wienicke C, Overstreet DH. Immobility-reducing effects of antidepressants in a genetic animal model of depression. Brain Res Bull 1992;28:821–3.

Sharpley AL, Cowen PJ. Effect of pharmacologic treatments on the sleep of depressed patients. Biol Psychiatry 1995;37:85–98.

Shiromani PJ, Overstreet DH, Levy D, Goodrich CA, Campbell SS, Gillin JC. Increased REM sleep in rats selectively bred for cholinergic hyperactivity. Neuropsychopharmacology 1988;1:127–l33.

Simiand J, Keane PE, Guitard J, Langlois X, Gonalons N, Martin P, et al. Antidepressant profile in rodents of SR 58611A, a new selective agonist for atypical beta-adrenoceptors. Eur J Pharmacol 1992;219:193–201.

Stemmelin J, Cohen C, Terranova JP, Lopez-Grancha M, Pichat P, Bergis O, et al. Stimulation of the β3-adrenoceptor as a novel treatment strategy for anxiety and depressive disorders. Neuropsychopharmacology 2008;33:574–87 [Electronic publication 2007 Apr 25].

Strosberg AD. Structure and function of the beta 3-adrenergic receptor. Annu Rev Pharmacol Toxicol 1997;37:421–50.

Zangen A, Overstreet DH, Yadid G. High serotonin and 5-Hydroxyindoleacetic acid levels in limbic regions of a rat model of depression: normalization by chronic antidepressant treatment. J Neurochem 1997;69:2477–83.

Zangen A, Overstreet DH, Yadid Y. Increased catecholamine levels in specific brain regions of a rat model of depression: normalization by chronic antidepressant treatment. Brain Res 1999;824:243–50.