ELSEVIER

Electrical Stimulation of Rat Medial Prefrontal Cortex Enhances Forebrain Serotonin Output: Implications for Electroconvulsive Therapy and Transcranial Magnetic Stimulation in Depression

Georg Juckel, M.D., Anna Mendlin, M.A., and Barry L. Jacobs, Ph.D.

Decreased activity of the prefrontal cortex (PFC), as well as reduced serotonergic neurotransmission, is considered as a characteristic feature of major depression. The mechanism by which electroconvulsive therapy (ECT) and transcranial magnetic stimulation (TMS) achieve their antidepressant effects may involve changes in PFC activity. It is, however, still unclear whether these changes are accompanied by increased synaptic availability of serotonin (5-HT). In the present study, 5-HT efflux in the rat ventral hippocampus and amygdala was analyzed using in vivo microdialysis during low-current electrical stimulation of PFC and other cortical regions. Electrical stimulation of the medial PFC produced current-dependent increases in limbic 5-HT output in both urethane-anesthetized and behaving rats. No effects on 5-HT levels were seen after comparable stimulation of either the lateral parts of the PFC, the medial precentral area, the primary motor cortex or the parietal cortex. This pronounced regional specificity of the effect of medial PFC stimulation on limbic 5-HT output suggests that activation of this particular area might play a crucial role in such antidepressant treatments as ECT and TMS. ***[Neuropsychopharmacology 21:391–398, 1999]*** *© 1999 American College of Neuropsychopharmacology. Published by Elsevier Science Inc.*

KEY WORDS: *Serotonin; Prefrontal cortex; Microdialysis; Electroconvulsive therapy; Transcranial magnetic stimulation; Depression*

An extensive body of clinical evidence suggests that prefrontal cortex (PFC) abnormalities are involved in the pathophysiology of major depression. Significantly decreased cerebral blood flow as well as reduced rates of glucose metabolism have been consistently found in the PFC of depressed patients (Soares and Mann 1997; Kennedy et al. 1997; George et al. 1993). This reduction seems to be localized predominantly in the left hemisphere (Baxter et al. 1989; Martinot et al. 1990). This is supported by structural neuroimaging studies demonstrating that poststroke depression is more likely to occur following a lesion in the left PFC rather than either in the right PFC or elsewhere in the left hemisphere (Morris et al. 1996; Robinson et al. 1984). Both neuropsychological and oculomotor functioning of prefrontal cortex is impaired in depression (Goodwin 1997; Sweeney et al. 1998). Furthermore, symptom improvement and remission of depressed patients have been associated with increases in cerebral blood flow and glucose metabolism in this region (Goodwin et al. 1993; Bench et al. 1995; Bonne and Krausz 1997; Buchsbaum et al. 1997).

From the Program in Neuroscience (AM, BLJ), Princeton University, Princeton, New Jersey; and Department of Psychiatry (GJ), Ludwig-Maximilians-University, Munich, Germany.

Address correspondence to: Dr. Georg Juckel, Department of Psychiatry, Ludwig-Maximilians-University, Nussbaumstr. 7, 80336 Munich, Germany.

Received June 25, 1998; revised August 31, 1998; accepted September 21, 1998.

Published by Elsevier Science Inc.
655 Avenue of the Americas, New York, NY 10010

0893-133X/99/$–see front matter
PII S0893-133X(98)00097-9

This close association between prefrontal cortical dysfunction and depression suggests that enhancement of PFC activity might be beneficial in the treatment of this disorder. Consistent with this idea, electroconvulsive therapy (ECT), as well as transcranial magnetic stimulation (TMS), seems to achieve its antidepressant effects by altering PFC activity. ECT leads to pronounced prefrontal EEG changes, which are directly related to the therapeutic outcome. Hence, the efficacy of ECT seems to be critically linked to prefrontal cortex involvement in ECT-induced seizure activity (Sackeim et al. 1996). This is further supported by studies that have reported changes in cerebral blood flow in prefrontal cortex of ECT responders (Nobler et al. 1994; Bonne et al. 1996; Petracca et al. 1995). In recent years, repetitive TMS of the left dorsolateral prefrontal cortex has emerged as a successful antidepressant treatment (Pascual-Leone et al. 1996; George et al. 1997). Although the exact mechanism underlying the efficacy of TMS is still unclear, this stimulation procedure probably induces focal activation of PFC neurons, as revealed by simultaneous measurements of cerebral glucose metabolism and blood flow (George et al. 1995; Paus et al. 1997; Fox et al. 1997).

Apart from decreased PFC activity, a functional deficiency in serotonergic neurotransmission is considered as another characteristic feature of depression (Maes and Meltzer 1995). The therapeutic efficacy of various antidepressant treatments is thought to result from their ability to enhance serotonergic function (Blier and De Montigny 1994). It is, however, still unclear what is the relationship between serotonin (5-HT) and the activity of the prefrontal cortex in the pathophysiology and treatment of depression. Available data suggest that the PFC in depressed patients is characterized by reduced serotonergic neurotransmission. Diminished responsiveness to the serotonin-releasing drug fenfluramine was observed in the PFC of patients with major depression (Mann et al. 1996). Tryptophan depletion-induced depressive relapses in previously remitted patients were associated with a decrease in glucose metabolism of the dorsolateral PFC (Bremner et al. 1997). At present, there is no direct neurobiological evidence linking the effects of ECT and TMS on the activity of prefrontal cortex to increased synaptic availability of serotonin in the forebrain. Both ECT and electroconvulsive shocks (ECS) in animals elicit generalized seizure activity, which does not allow to relate their numerous neurochemical effects (for review, see Mann and Kapur 1994; Fochtmann 1994) to one specific anatomical area. Similarly, the inherent technical limitations of the TMS procedure used in rat studies result in the stimulation of multiple cortical areas, making it difficult to interpret the effects in terms of regional specificity. Therefore, to elucidate the relationship between changes in PFC activity and availability of 5-HT, it is necessary to measure extracellular 5-HT levels during focal electrical stimulation of the cortex. The present study used *in vivo* microdialysis, in both anesthetized and behaving rats, to examine the effects of electrical stimulation of the PFC on 5-HT output in two brain areas believed to be important in the pathophysiology of depression; namely, the ventral hippocampus and the amygdala. We also assessed the regional specificity of this relationship by comparing the effects of focal stimulation of several areas within the PFC, as well as of other cortical regions.

METHODS

Animals

Male Sprague–Dawley rats, weighing 240 to 285 g, were housed individually under controlled temperature and lighting conditions (22 ± 0.5°C; 12-h reversed light/dark cycle, white light off/dim red light on at 11:00 A.M.) with food and water available *ad libitum*. All rats were cared for and used in strict accordance to the PHS Guide for the Care and Use of Laboratory Animals. All procedures were reviewed and approved by the Institutional Animal Care and Use Committee of Princeton University.

Surgical Procedure

Rats were anesthetized with urethane (1.4 g/kg IP, 0.2 g/kg supplement if necessary) and placed in a stereotaxic frame in a flat-skull position. During surgery and acute experiments, body temperature of the animals was continuously monitored with a rectal thermometer and was maintained at 37 to 38°C by using heating lamps. Stainless steel guide cannulae (0.7 mm o.d.) were implanted in the left or right ventral hippocampus (AP −5.4 mm, ML ± 4.8 mm, from bregma, DV −3.5 mm below dura; 9-mm cannula length), or in the left amygdala (AP −2.8 mm, ML +5.0 mm, DV − 5.4 mm; 14-mm cannula length), according to the atlas of Paxinos and Watson (1986). Additional small craniotomies were performed above the cortical regions of interest for electrical stimulation. For cortical EEG recordings, a pair of stainless steel electrodes (1.5-mm diameter) was implanted at the following coordinates: AP +2 mm, ML −3 mm and AP −4 mm, ML −3 mm. For experiments in freely moving animals, rats were pretreated with atropine sulfate (0.2 mg/kg IM) and anesthetized with a mixture of ketamine HCl and xylazine (80 mg/kg and 12 mg/kg, respectively, IM). A stimulating electrode, as described below, and one hippocampal cannula were implanted, secured with skull screws and dental acrylic, and the cannula was plugged with a stainless steel stylet. Postoperatively, rats received an injection of penicillin (300,000 U/kg IM) and were allowed to recover for 4 to 7 days.

Electrical Stimulation

A bipolar stimulating electrode was made from two insulated nichrome wires (360 μm diameter; California Fine Wire, Inc., Grover Beach, CA) that were etched for 0.5 mm at the tips and had a tip separation of 0.5 mm in the anterior–posterior plane. Stimulation was applied to the following cortical areas: left or right medial prefrontal cortex (prelimbic area or CG3, abbreviations in accordance with Zilles (1985); AP + 3.2 mm, ML ± 0.5 mm, both from bregma, DV −2.5 mm below dura, adjusted for the anterior stimulating wire) as well as parietal cortex (Par1; AP + 1.7 mm, ML + 5.0 mm, DV −2.4 mm), primary motor cortex (Fr1; AP + 3.7 mm, ML +3.2 mm, DV −1.0 mm), medial precentral area (FR2; AP +4.7 mm, ML + 2.0 mm, DV −1.0 mm), dorsal part of the agranular insular cortex (AID; AP +3.7 mm, ML +4.1 mm, DV −3.5 mm), and lateral orbitofrontal cortex (LO; AP +4.2 mm, ML +2.0 mm, DV −13.7 mm), all at the left side. In the acute experiments, the electrode was placed sequentially in no more than three different cortical areas. In the experiments using behaving animals, the stimulating electrode was placed only in the left medial prefrontal cortex. Electrical stimulation was delivered by a Grass S48 Stimulator and consisted of 1 s trains of 5 ms stimuli (60 Hz, 100 and 150 μA), presented every 5 s for 20 min.

Microdialylsis and Experimental Procedure

Concentric dialysis probes (2.0 mm length of nitrocellulose membrane, 0.22 mm o.d., 6,000 Da cut-off, Spectrum, Houston, TX) were constructed as previously described (Hernandez et al. 1987). In the acute studies, the probes were implanted at the end of surgery. In the behavioral study, the rats were gently restrained on the day of the experiment without use of anesthesia for probe implantation. Dialysis probes were lowered through the guide cannulae and secured with dental acrylic, so that the probe tips extended 3 mm beyond the cannulae tips. The probe inlets were attached to a Harvard syringe microinfusion pump (Harvard Apparatus, Boston, MA; via a fluid swivel in the behavioral study), and a modified Ringers solution (147.2 mM NaC1, 4.0 mM KC1, 1.8 mM $CaC1_2$) was continuously infused at a flow rate of 1.3 μl/min. The perfusion medium contained 3 μM fluoxetine (Eli Lilly, Indianapolis, IN). Collection of 20 min perfusate samples started 3 h after microdialysis probe implantation. Immediately after obtaining a stable 3-sample baseline, electrical stimulation was administered, and three additional follow-up dialysate samples were collected.

Chromatography

A reversed phase high-performance liquid chromatography system coupled with electrochemical detection (HPLC-ECD) was used for the analysis of serotonin. The mobile phase (0.15 M chloroacetic acid, 0.12 M NaOH, 0.18 mM EDTA, 60 ml/l acetronitrile, 1.0 mM sodium octane sulfate) was delivered at a flow rate of 1.0 ml/min onto a 10 cm × 3.2 mm ODS 3-μm column (BAS Inc., West Lafayette, IN). Perfusate samples were manually injected (model 7125 injector, Rheodyne Inc., Cotati, CA) and analyzed using a dual potentiostat electrochemical detector (model 400 EG&G, Princeton Applied Research Corp., Princeton, NJ), with the potentials applied to the parallel working electrodes set at 610 and 590 mV relative to an Ag/AgC1 reference electrode. A Shimadzu model C-R3A integrator (Kyoto, Japan) was used to analyze the output from the detector. Identification and quantification of 5-HT in the samples was achieved by comparison of the retention time and peak height to those of a standard solution containing 5-HT. The detection limit for 5-HT was approximately 1 pg based on a signal-to-noise ratio of 3:1. *In vitro* probe recovery was determined by immersing the probes in a standard solution containing 10 pg of 5-HT and perfusing them for at least 4 h with a Ringers solution at a flow rate of 1.3 μl/min. The relative recovery was 13.2 ± 3.8% for 5-HT ($n = 8$).

Histology

For verification of stimulating electrode and microdialysis probe placement, animals were perfused intracardially with 10% formalin in saline under deep phenobarbital anesthesia (100 mg/kg, IP). Brains were removed, serial frozen sections (75-μm thick) were cut using a microtome, mounted on glass slides, and stained with neutral red. The slides were examined under a microscope, and only the data from the animals with correct placement of both electrode and probe were reported.

Statistical Analysis

To minimize between-subject variability, levels of extracellular 5-HT were expressed as a percentage of the mean of the three baseline samples. All values are expressed as means ± SEM. Data were analyzed using either one-way or two-way analysis of variance (ANOVA) with repeated-measures ("time" as a within-subject factor and "group" as a between-subject factor), followed by post hoc comparisons (Student–Newman–Keuls' test).

RESULTS

Electrical stimulation of the left medial prefrontal cortex (mPFC) increased serotonin output in both the ipsilateral ventral hippocampus and the amygdala in a cur-

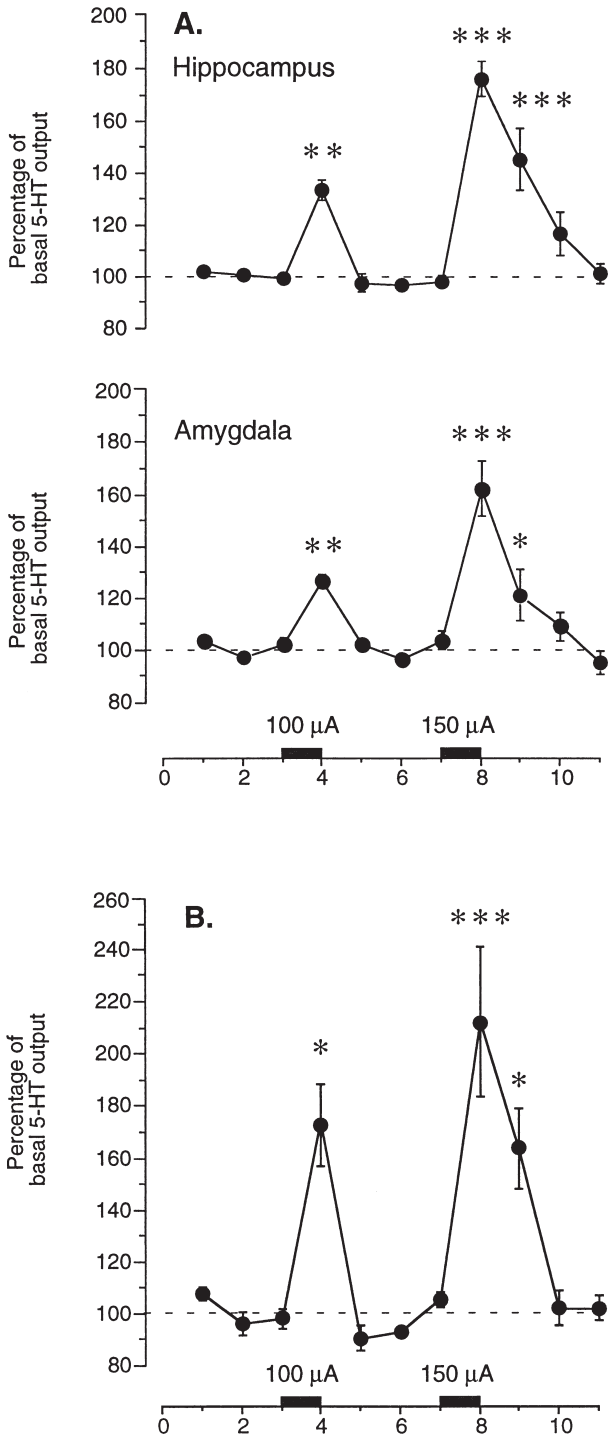

Figure 1. The effects of electrical stimulation of the left medial prefrontal cortex (100 µA and 150 µA) on forebrain 5-HT efflux (20-min samples) in urethane-anesthetized rats **(A)** and in behaving rats **(B)**. A. Medial PFC stimulation produced a current-dependent increase in 5-HT output in the ipsilateral ventral hippocampus (top panel, $n = 6$) and amygdala (bottom panel, $n = 6$) of anesthetized rats. B. The effects of mPFC stimulation on 5-HT efflux in the ipsilateral ventral hippocampus were more pronounced in behaving rats ($n = 3$). Each point represents mean values ± SEM. Data are expressed as percentages of the three baseline samples. Horizontal bars represent time of electrical stimulation (20 min). *$p < .05$ compared to baseline; **$p < .01$ compared to baseline; ***$p < .001$ compared to baseline; Student–Newman–Keuls' multiple comparisons test.

rent-dependent manner. In the hippocampus (Figure 1A), 5-HT efflux increased by 33 ± 4% ($F_{(10,50)} = 24.7$, $p < .01$) and by 75 ± 7% ($p < .001$) above baseline at currents of 100 µA and 150 µA, respectively. In the amygdala, 5-HT levels increased by 26 ± 3% ($F_{(10,50)} = 15.6$, $p < .01$) and by 61 ± 11% ($p < .001$) above baseline at currents of 100 µA and 150 µA, respectively. In both areas, the increase in 5-HT output produced by the 150 µA current was significantly greater than that produced by the 100 µA current ($p < .001$). In behaving animals (Figure 1B), electrical stimulation of the left mPFC enhanced serotonin output over baseline values in the ipsilateral hippocampus by 73 ± 16% at 100 µA ($F_{(10,20)} = 7.7$, $p < .05$) and by 112 ± 39% at 150 µA ($p < .001$). In all cases, the effect of the higher current was more prolonged. Thus, 5-HT efflux in the first poststimulation sample was still significantly elevated after 150 µA, but not after 100 µA (Figure 1).

Similarly, 150 µA stimulation of the right mPFC in three anesthetized rats produced a 70 ± 8% increase in 5-HT output in the ipsilateral hippocampus ($F_{(6,12)} = 24.7$, $p < .001$). The magnitude of this increase was not significantly different from that produced by ipsilateral stimulation of the left mPFC ($F_{(6,42)} = 1.4$, $p = .24$). Furthermore, stimulation of the right mPFC in four anesthetized rats (150 µA) produced a 50 ± 8% increase in 5-HT levels in the contralateral hippocampus ($F_{(6,18)} = 18.6$, $p < .001$). This increase was less pronounced than that observed during ipsilateral stimulation on the left side at the same current (50 ± 8% vs. 75 ± 7%; $F_{(6,48)} = 2.5$, $p < .01$).

To assess the regional specificity of the effects of mPFC stimulation, the same stimulation procedure (using 150 µA current) was carried out in a number of other cortical areas: agranular insular cortex, lateral orbitofrontal cortex, medial precentral area Fr2, primary motor cortex, and parietal cortex. As shown in Figure 2, 5-HT efflux in the left hippocampus was unaffected by stimulation of any of these regions, except for mPFC ($F_{(30,138)} = 12.4$, $p < .001$).

Electrical stimulation of the left mPFC did not induce any discernible changes in EEG or behavior. EEG activity (mostly in the theta band), as observed under urethane anesthesia, was not affected by stimulation at either 100 or 150 µA currents. In the study with freely moving animals, no overt motor or behavioral reactions were observed at either stimulation level.

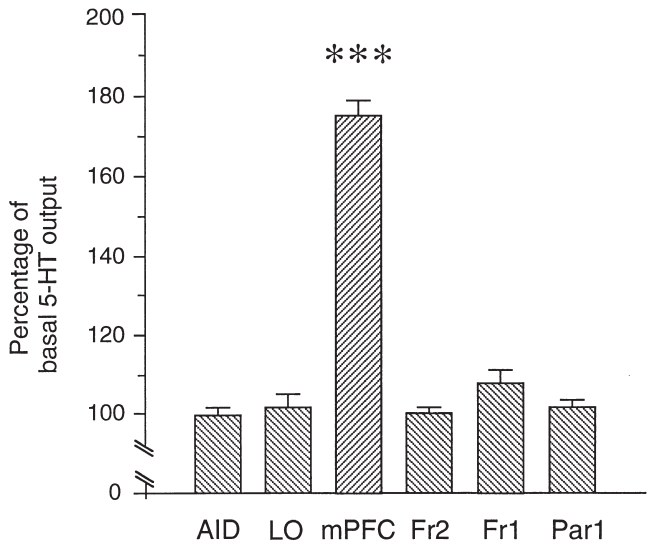

Figure 2. Comparison of the effects of ipsilateral electrical stimulation (150 μA) of various cortical areas on 5-HT output in the left ventral hippocampus. Abbreviations: AID, dorsal part of the agranular insular cortex ($n = 5$); LO, lateral orbitofrontal cortex ($n = 4$); mPFC, medial prefrontal cortex ($n = 6$); Fr2, medial precentral area ($n = 5$); Fr1, primary motor cortex ($n = 4$); Par1, parietal cortex ($n = 5$); each column represents mean values ± SEM. Data are expressed as percentages of the three baseline samples. ***$p < .001$ compared to baseline; Student–Newman–Keuls' multiple comparisons test.

DISCUSSION

The present study is the first to demonstrate that low-current electrical stimulation of the medial prefrontal cortex, but not of the lateral parts of the PFC or of other cortical areas, enhances serotonin levels in such subcortical areas as the hippocampus and amygdala. This highly specific effect was current-dependent, not-lateralized, and was found in both anesthetized and awake rats. The absence of motor or behavioral reactions during low-current stimulation of mPFC in behaving animals is in agreement with previous studies (Fuster 1997; Taber and Fibiger 1993). The effect of prefrontal stimulation on the 5-HT output was more pronounced in these rats than in the urethane-anesthetized rats.

The mechanism by which stimulation of mPFC neurons enhances 5-HT efflux in terminal areas may involve activation of the dorsal and median raphe nuclei. Consistent with this hypothesis, it has been demonstrated that rat medial prefrontal cortex (prelimbic area) has dense efferent projections to both the dorsal and the median raphe nuclei (Aghajanian and Wang 1977; Beckstead 1979; Wyss and Sripanidkulchai 1984; Sesack et al. 1989; Behzadi et al. 1990; Peyron et al. 1998). Thus, it is possible that PFC stimulation activates the raphe nuclei leading to increased -HT levels in terminal areas. It cannot, however, be dismissed that this effect is mediated by local activation of the nerve terminals, because medial and lateral prefrontal cortex project to both the hippocampus and amygdala in rats (Groenewegen et al. 1997). Nevertheless, because ipsilateral mPFC stimulation in the present study produced similar increases in 5-HT output in the hippocampus and amygdala, and because contralateral stimulation also enhanced 5-HT efflux, the effects of mPFC stimulation on extracellular 5-HT levels seem to be mediated by a central structure, such as the raphe nuclei. In this context, the fact that the effects of contralateral stimulation were smaller than those of insilateral stimulation could be explained by the known lateralization of forebrain projections of serotonergic cells in the raphe nuclei (Jacobs and Azmitia 1992).

The effect of mPFC electrical stimulation on serotonergic neurotransmission was found to be highly specific. Stimulation of such cortical areas outside the PFC as the medial precentral area Fr2, the primary motor, and the parietal cortex did not affect serotonin output in either hippocampus or amygdala. One possible explanation for this is that the prefrontal cortex is the only cortical area with direct projections to the midbrain raphe nuclei in rats (Aghajanian and Wang 1977; Behzadi et al. 1990; Marcinkiewicz et al. 1989; Peyron et al. 1998), as well as in primates (Arnsten and Goldman-Rakic 1984). It is noteworthy that there is still open debate as to whether the medial precentral area Fr2, located in the dorsomedial, or so-called shoulder region of the rat frontal cortex, belongs to the PFC (Uylings and Van Eden 1990; Preuss 1995). In the present study, Fr2 was considered as part of the sensorimotor cortex, rather than that of the PFC, according to several anatomical criteria (Zilles and Wree 1995; Reep et al. 1987; Donoghue and Wise 1982). On the other hand, the areas around the rhinal sulcus, such as the lateral orbitofrontal cortex (LO) and the dorsal part of the agranular insular cortex (AID), constitute the lateral prefrontal cortex in rats, because they have reciprocal connections to the mediodorsal thalamic nucleus (Zilles and Wree 1995; Fuster 1997) which is a main anatomical criterion for belonging to the PFC. Although LO and AID have projection fibers to the dorsal raphe nucleus in rats (Peyron et al. 1998; Beckstead 1979), these fibers, unlike those originating from mPFC, probably have no functional relevance for the serotonergic neurotransmission, because electrical stimulation of AID and LO did not alter hippocampal 5-HT output in the present study. There are two reasons for this functional difference between medial and lateral PFC: first, the medial PFC sends significantly more efferent fibers to the dorsal raphe nucleus than LO and AID; and second, only the medial PFC projects to the whole of the dorsal raphe nucleus (Peyron et al. 1998).

The present findings have important implications for the understanding of human pathology, because it is well established that the medial PFC in rats is analo-

gous to the medial prefrontal cortex in primates, including humans (Uylings and Van Eden 1990; Preuss 1995). As far as the human dorsolateral PFC is concerned, three cortical areas can be considered as analogous to this clinically relevant region in the human brain: LO, Fr2, and AID. However, the rat LO probably corresponds to the primate orbitofrontal cortex; whereas, Fr2 presumably belongs to the sensorimotor rather than to the PFC. Because of its anatomical position close to the upper frontal pole, the agranular insular cortex AID is the most promising candidate for a rat analog of the human dorsolateral prefrontal cortex. Our study demonstrates that stimulation of none of these three areas produces any effect on 5-HT output, suggesting that the dorsolateral PFC does not seem to influence serotonergic neurotransmission. The finding that low-current electrical stimulation of the rat medial PFC enhances 5-HT output in such limbic structures as the hippocampus and amygdala underlines the importance of the medial PFC for pathophysiology and treatment of depression. Prefrontal abnormalities in depressed patients were found mostly in the medial parts of PFC (Drevets et al. 1997; Ebert and Ebmeier 1996; Mann et al. 1996; Buchsbaum et al. 1997; Agren and Reibring 1994). It was suggested that one of the core symptoms of depression, namely, motor and affective apathy, is related specifically to dysfunctions of the medial PFC (Cummings 1995; Saint-Cyr et al. 1995). This is supported by positron emission tomography (PET) studies in healthy subjects demonstrating that medial PFC is involved in the regulation of mood and emotions (Reiman 1997).

The high regional specificity of medial PFC stimulation on 5-HT output observed in this study also has implications for such antidepressant treatments as ECT and TMS, although because of our focal approach, the stimulation parameters used here were somewhat different from those of ECT or TMS (20 trains of 1 ms stimuli at 20 Hz, lasting for 2 or 10 s, administered over 20 min) in humans. Our findings suggest that efficacy of ECT in depression might depend upon the involvement of the medial PFC. To activate this deeply located area by outside electrical stimulation, it seems to be necessary to induce generalized seizures involving all cortical areas. This could explain the lack of efficacy of subconvulsive ECT and the need for eliciting generalized seizures to produce an antidepressant effect (Sackeim 1994). Our results also indicate that transcranial magnetic stimulation of the medial PFC, rather than the commonly targeted dorsolateral PFC, may result in a greater success in treatment of depressed patients. Regarding the possible synaptic mechanisms involved in the effects of ECT and TMS, it remains to be elucidated how the transient alterations in 5-HT levels induced by electrical stimulation could lead to long-term effects relevant for the antidepressant response in animals, single administration of both TMS (Ben-Shachar et al. 1997) and ECS (Zis et al. 1992), increases forebrain 5-HT levels; whereas, repeated ECS does not affect either hippocampal 5-HT efflux (Gur et al. 1997) or the firing rate of 5-HT neurons (Blier and Bouchard 1992). There is, however, sufficient evidence demonstrating that chronic, but not single or subconvulsive ECS, induces pronounced sensitization of postsynaptic 5-HT_{1A} receptors, especially at hippocampal pyramidal cells (de Montigny 1984; Chaput et al. 1991; Blier and de Montigny 1994). Thus, the transient alterations of 5-HT concentration in the synaptic cleft evoked by repeated administration of either ECS or another type of electrical stimulation might cumulatively result in postsynaptic 5-HT receptor adaptations and, consequently, in the therapeutic effects of chronic ECS or ECT. Finally, stimulation of medial PFC has diverse neurochemical effects, affecting several neurotransmitter systems besides 5-HT. Electrical stimulation of this area in rats enhances the levels of forebrain dopamine (Taber and Fibiger 1993, 1995; Murase et al. 1993) and acetylcholine (Taber and Fibiger 1994; Consolo et al. 1996). It was also demonstrated that medial PFC stimulation activates noradrenergic neurons in the locus coeruleus (Aston-Jones et al. 1991; Jodo et al. 1998). Because stimulation of the medial prefrontal cortex affects all these neurotransmitter systems relevant to depression, any treatment activating the medial PFC might be therapeutically successful for this disorder.

ACKNOWLEDGMENTS

G. J. was a recipient of the DGPPN-Solvay/Duphar-Award for Psychiatric Research. The study was supported by the NIMH (MH23433). The authors are deeply grateful to Dr. C. A. Fornal for his helpful suggestions and comments throughout the course of this study. The encouragement and advice of Dr. P. Mavrogiorgou and Dr. F. J. Martin are greatly appreciated.

REFERENCES

Aghajanian GK, Wang RY (1977): Habenular and other midbrain raphe afferents demonstrated by a modified retrograde tracing technique. Brain Res 122:229–242

Agren H, Reibring L (1994): PET studies of presynaptic monoamine metabolism in depressed patients and healthy volunteers. Pharmacopsychiatry 27:2–6

Arnsten AFT, Goldman-Rakic PS (1984): Selective prefrontal cortical projections to the region of the locus coeruleus and raphe nuclei in the rhesus monkey. Brain Res 306:9–18

Aston-Jones G, Chiang C, Alexinsky T (1991): Discharge of noradrenergic locus coeruleus neurons in behaving rats and monkeys suggests a role in vigilance. Prog Brain Res 88:501–520

Baxter LR, Schwartz JM, Phelps ME, Mazziotta JC, Guze BH, Selin CE, Gerner RH, Sumida RM (1989): Reduction of

prefrontal cortex glucose metabolism common to three types of depression. Arch Gen Psychiat 46:243–250

Beckstead RM (1979): An autoradiographic examination of corticocortical and subcortical projections of the mediodorsal-projection (prefrontal) cortex in the rat. J Comp Neurol 184:43–62

Behzadi G, Kalen P, Parvopassu F, Wiklund L (1990): Afferents to the median raphe nucleus of the rat: Retrograde cholera toxin and wheat germ conjugated horseradish peroxidase tracing, and selectin D-(3H)aspartate labelling of possible excitatory amino acid inputs. Neuroscience 37:77–100

Bench CJ, Frackowiak RS, Dolan RJ (1995): Changes in regional cerebral blood flow on recovery from depression. Psychol Med 25:247–261

Ben-Shachar D, Belmaker RH, Grisaru N, Klein E (1997): Transcranial magnetic stimulation induces alterations in brain monoamines. J Neural Transm 104:191–197

Blier P, Bouchard C (1992): Effect of repeated electroconvulsive shocks on serotonergic neurons. Eur J Pharmacol 211:365–373

Blier P, de Montigny C (1994): Current advances and trends in the treatment of depression. TIPS 15:220–226

Bonne O, Krausz Y (1997): Pathophysiological significance of cerebral perfusion abnormalities in major depression—Trait or state marker? Eur Neuropsychopharmacol 7:225–233

Bonne O, Krausz Y, Shapira B, Bocher M, Karger H, Gorfine M, Chisin R, Lerer B (1996): Increased cerebral blood flow in depressed patients responding electroconvulsive therapy. J Nucl Med 137:1075–1080

Bremner JD, Innis RB, Salomon RM, Staib LH, Ng CK, Miller HL, Bronen RA, Krystal JH, Duncan J, Rich D, Price LH, Malison R, Dey H, Soufer R, Charney DS (1997): Positron emission tomography measurement of cerebral metabolic correlates of tryptophan depletion-induced depressive relapse. Arch Gen Psychiat 54:364–374

Buchsbaum MS, Wu J, Siegel BV, Hackett E, Trenary M, Abel L, Reynolds C (1997): Effect of sertraline on regional metabolic rate in patients with affective disorder. Biol Psychiat 41:15–22

Chaput Y, de Montigny C, Blier P (1991): Presynaptic and postsynaptic modifications of the serotonin system by long-term administration of antidepressant treatments. Neuropsychopharmacology 5:219–229

Consolo S, Baldi G, Giorgi S, Nannini L (1996): The cerebral cortex and parafascicular thalamic nucleus facilitate in vivo acetylcholine release in the rat striatum through distinct glutamate receptor subtypes. Eur J Neurosci 8:2702–2710

Cummings JL (1995): Anatomic and behavioral aspects of fronto-subcortical circuits. Ann NY Acad Sci 769:1–13

De Montigny C (1984): Electroconvulsive shock treatments enhance responsiveness of forebrain neurons to serotonin. J Pharmacol Exper Ther 228:230–234

Donoghue JP, Wise SP (1982): The motor cortex of the rat: Cytoarchitecture and microstimulation mapping. J Comp Neurol 212:76–88

Drevets WC, Price JL, Simpson JR, Todd RD, Reich T, Vannier M, Raichle ME (1997): Subgenual prefrontal cortex abnormalities in mood disorders. Nature 386:824–827

Ebert D, Ebmeier KP (1996): The role of the cingulate gyrus in depression: From functional anatomy to neurochemistry. Biol Psychiat 39:1044–1050

Fochtmann LJ (1994): Animal studies of electroconvulsive therapy: Foundations for future research. Psychopharmacol Bull 30:321–444

Fox P, Ingham R, George MS, Mayberg H, Ingham J, Roby J, Martin C, Jerabek P (1997): Imaging human intracerebral connectivity by PET during TMS. Neuroreport 8:2787–2791

Fuster JM (1997): The Prefrontal Cortex: Anatomy, Physiology, and Neuropsychology of the Frontal Lobe, 3rd ed. Philadelphia, Lippincott-Raven

George MS, Ketter TA, Post RM (1993): SPECT and PET imaging in mood disorders. J Clin Psychiatry 54:6–13

George MS, Wassermann EM, Williams WE, Callahan A, Ketter TA, Basser P, Hallett M, Post RM (1995): Daily repetitive transcranial magnetic stimulation (rTMS) improves mood in depression. Neuroreport 6:1853–1856

George MS, Wassermann EM, Kimbrell TA, Little JT, Williams WE, Danielson AL, Greenberg BD, Hallett M, Post RM (1997): Mood improvement following daily left prefrontal repetitive transcranial magnetic stimulation in patients with depression: A placebo-controlled crossover trial. Am J Psychiat 154:1752–1756

Goodwin GM (1997): Neuropsychological and neuroimaging evidence for the involvement of the frontal lobes in depression. J Psychopharmacol 11:115–122

Goodwin GM, Austin MP, Dougall N, Ross M, Murray C, O'Carroll RE, Moffoot A, Prentice N, Ebmeier KP (1993): State changes in brain activity shown by the uptake of 99mTc-exametazime with single photon emission tomography in major depression before and after treatment. J Affect Disord 29:243–253

Groenewegen HJ, Wright CI, Uylings HBM (1997): The anatomical relationships of the prefrontal cortex with limbic structures and the basal ganglia. J Psychopharmacol 11:99–106

Gur E, Lerer B, Newman ME (1997): Chronic electroconvulsive shock and 5-HT autoreceptor activity in rat brain: an in vivo microdialysis study. J Neural Transm 104: 795–804

Hernandez L, Lee F, Hoebel BG (1987): Simultaneous microdialysis and amphetamine infusion in the nucleus accumbens and striatum of freely moving rats: Increase in extracellular dopamine and serotonin. Brain Res Bull 19:623–628

Jacobs BL, Azmitia EC (1992): Structure and function of the brain serotonin system. Physiol Rev 72:165–229

Jodo E, Chiang C, Aston-Jones G (1998): Potent excitatory influence of prefrontal cortex activity on noradrenergic locus coeruleus neurons. Neuroscience 83:63–79

Kennedy SH, Havanmard M, Vaccarino FJ (1997): A review of functional neuroimaging in mood disorders: Positron emission tomography and depression. Can J Psychiat 42:467–475

Maes M, Meltzer HY (1995): The serotonin hypothesis of

major depression. In Bloom FE, Kupfer DJ (eds), Psychopharmacology: The Fourth Generation of Progress. New York, Raven Press, pp 933–944

Mann JJ, Kapur S (1994): Elucidation of biochemical basis of the antidepressant action of electroconvulsive therapy by human studies. Psychopharmacol Bull 30:445–453

Mann JJ, Malone KM, Diehl DJ, Perel J, Cooper TB, Mintun MA (1996): Demonstration in vivo of reduced serotonin responsivity in the brain of untreated depressed patients. Am J Psychiat 153:174–182

Marcinkiewicz M, Morcos R, Chretien M (1989): CNS connections with the median raphe nucleus: Retrograde tracing with WGA-apoHRP-Gold complex in the rat. J Comp Neurol 289:11–35

Martinot J, Hardy P, Feline A, Huret J, Mazoyer B, Attar-Levy D, Pappata S, Syrota A (1990): Left prefrontal glucose hypometabolism in the depressed state: A confirmation. Am J Psychiat 147:1313–1317

Morris PL, Robinson RG, Raphael B, Hopwood MJ (1996): Lesion location and poststroke depression. J Neuropsychiat Clin Neurosci 8:399–403

Murase S, Grenhoff J, Chouvet G, Gonon FG, Svensson TH (1993): Prefrontal cortex regulates burst firing and transmitter release in rat mesolimbic dopamine neurons studied in vivo. Neurosci Lett 157:53–56

Nobler MS, Sackeim HA, Prohovnik I, Moller JR, Mukherjee S, Schnur DB, Prudic J, Devanand DP (1994): Regional cerebral blood flow in mood disorders, III. Treatment and clinical response. Arch Gen Psychiat 51:884–897

Pascual-Leone A, Rubio B, Pallardo F, Catala MD (1996): Rapid-rate transcranial magnetic stimulation of left dorsolateral prefrontal cortex in drug-resistant depression. Lancet 347:233–237

Paus T, Jech R, Thompson CJ, Comeau R, Peters T, Evans AC (1997): Transcranial magnetic stimulation during positron emission tomography: A new method for studying connectivity of the human cerebral cortex. J Neurosci 17:3178–3184

Paxinos G, Watson C (1986): The Rat Brain in Stereotaxic Coordinates, 2nd ed. San Diego, Academic Press

Petracca G, Migliorelli R, Vazquez S, Starkstein SE (1995): SPECT findings before and after ECT in a patient with major depression and Cotard's syndrome. J Neuropsychiat Clin Neurosci 7:505–507

Peyron C, Petit JM, Rampon C, Jouvet M, Luppi PH (1998): Forebrain afferents to the rat dorsal raphe nucleus demonstrated by retrograde and anterograde tracing methods. Neuroscience 82:443–468

Preuss TM (1995): Do rats have prefrontal cortex? The Rose–Wooley–Akert program reconsidered. J Cogn Neurosci 7:1–24

Reep RL, Corwin JV, Hashimoto A, Watson RT (1987): Efferent connections of the rostral portion of medial agranular cortex in rats. Brain Res Bull 19:203–221

Reiman EM (1997): The application of positron emission tomography to the study of normal and pathologic emotions. J Clin Psychiat 58:4–14

Robinson RG, Kubos KL, Starr LB, Rao K, Price TR (1984): Mood disorder in stroke patients. Importance of location of lesion. Brain 107:81–93

Sackeim HA (1994): Central issues regarding the mechanisms of action of electroconvulsive therapy: Directions for future research. Psychopharmacol Bull 30:281–308

Sackeim HA, Luber B, Katzman GP, Moeller JR, Prudic J, Devanand DP, Nobler MS (1996): The effects of electroconvulsive therapy on quantitative electroencephalograms. Arch Gen Psychiat 53:814–824

Saint-Cyr JA, Taylor AE, Nicholson K (1995): Behavior and the basal ganglia. Adv Neurol 65:1–28

Sesack SR, Deutch AY, Roth RH, Bunney BS (1989): Topographical organization of the efferent projections of the medial prefrontal cortex in the rat: An anterograde tract-tracing study with *Phaseolus vulgaris leucoagglutinin*. J Comp Neurol 290:213–242

Soares JC, Mann JJ (1997): The functional neuroanatomy of mood disorders. J Psychiat Res 31:393–432

Sweeney JA, Strojwas MH, Mann JJ, Thase ME (1998): Prefrontal and cerebellar abnormalities in major depression: Evidence from oculomotor studies. Biol Psychiat 43:584–594

Taber MT, Fibiger HC (1993): Electrical stimulation of the medial prefrontal cortex increases dopamine release in the striatum. Neuropsychopharmacology 9:271–275

Taber MT, Fibiger HC (1994): Cortical regulation of acetylcholine release in rat striatum. Brain Res 639: 354–356

Taber MT, Fibiger HC (1995): Electric stimulation of the prefrontal cortex increases dopamine release in the nucleus accumbens of the rat: modulation by metabnotropic glutamate receptors. J Neurosci 15:3896–3904

Uylings HBM, Van Eden CG (1990): Qualitative and quantitative comparison of the prefrontal cortex in rat and in primates, including humans. Progr Brain Res 85:31–62

Wyss JM, Sripanidkulchai K (1984): The topography of the mesencephalic and pontine projections from the cingulate cortex of the rat. Brain Res 293:1–15

Zilles K (1985): The Cortex of the Rat: A Stereotaxic Atlas. Berlin, Springer

Zilles K, Wree A (1995): Cortex: Areal and laminar structure. In Paxinos G (ed), The Rat Nervous System, 2nd ed. San Diego, Academic Press, pp 649–685

Zis AP, Nomikos GG, Brown EE, Damsma G, Fibiger HC (1992): Neurochemical effects of electrically and chemically induced seizures: An in vivo microdialysis study in the rat hippocampus. Neuropsychopharmacology 7:189–195