Pergamon

Pharmacology Biochemistry and Behavior, Vol. 47, No. 4, pp. 985–987, 1994

0091-3057/94 $6.00 + .00

0091-3057(93)E0024-X

RAPID COMMUNICATION

Lithium Treatment Restores Clonidine's Effect in an Animal Model of Depression

CHARANJIT S. AULAKH,[1] JAMES L. HILL AND DENNIS L. MURPHY

Laboratory of Clinical Science, National Institute of Mental Health, Bethesda, MD 20892

Received 3 June 1993

AULAKH, C. S., J. L. HILL AND D. L. MURPHY. *Lithium treatment restores clonidine's effect in an animal model of depression.* PHARMACOL BIOCHEM BEHAV 47(4) 985–987, 1994.—IP administration of various doses of clonidine produces significant increases in growth hormone levels in the Wistar rats but not in the Fawn-Hooded (FH) rats, a rat strain suggested to be a genetic model of depression. However, short-term lithium treatment restores clonidine's effect on growth hormone levels in the Fawn-Hooded rats. Potentiation of clonidine's effect on growth hormone levels following short-term lithium treatment appears most likely due to increased serotonergic function as a consequence of enhanced 5-HT concentrations at postsynaptic 5-HT_{1C} receptor sites. Thus, the reversal of a deficit state in Fawn-Hooded rats by lithium treatment supports earlier studies suggesting this rat strain to represent a genetic model of depression.

α_2-Adrenergic heteroreceptors 5-HT_{1C} receptors Depression Growth hormone Fawn-Hooded rat

THE lithium ion is effective clinically for the treatment of acute manic illness, for prophylaxis in manic-depressive (bipolar) and unipolar depressive disorders, and, in conjunction with antidepressant drugs, for the treatment of resistant depressive illness (6). There are numerous studies demonstrating an interaction of lithium with the serotonergic and the noradrenergic neurotransmitter mechanisms (18).

The Fawn-Hooded (FH) rat strain is associated with a hemorrhagic disorder known as platelet storage pool deficiency analogous to that in the Chediak-Higashi syndrome of humans. The blood platelets from FH rats have decreased numbers and contents of dense granules, decreased concentrations of serotonin, diminished uptake of serotonin, and also manifest functional subsensitivity to serotonin agonists (2). Recently, the FH rat strain has been suggested to represent a genetic model of depression and alcoholism (1,13).

We have recently demonstrated that clonidine stimulates growth hormone (GH) secretion by activation of α_2-adrenergic heteroreceptors present on 5-HT nerve terminals which, in turn, enhance 5-HT activity via stimulation of postsynaptic 5-HT_{1C} receptors to promote growth hormone (GH)-releasing factor (1). Furthermore, we demonstrated that IP administration of various doses (50–200 μg) of clonidine produced significant increases in GH levels in the Wistar rat strain but not in the FH rat strain, suggesting that either α_2-adrenergic heteroreceptors or 5-HT_{1C} receptors or both were functionally subsensitive in the FH rat strain relative to the Wistar rat strain (1). The purpose of the present study was to investigate if lithium treatment would restore clonidine's effect on GH levels in the FH rat strain. Therefore, we studied the effects of clonidine on GH levels in saline-treated, short-term lithium-treated, and long-term lithium-treated FH rats.

MATERIALS AND METHODS

Male FH rats weighing approximately 250 g were used. They were housed six per cage in a temperature-controlled (25 ± 1°C) room with a 12-h light/dark cycle (lights on at 0700). The animals had free access to food and water at all times. Separate groups of animals were used for saline treatment, short-term lithium treatment, and long-term lithium treatment.

For lithium treatment, the animals were given rat chow containing lithium carbonate (1.65 g/kg) for 3 days (short-

[1] Requests for reprints should be addressed to C. S. Aulakh, Building 10, Room 3D41, NIH Clinical Center, NIMH, 9000 Rockville Pike, Bethesda, MD 20892.

term treatment) and 21 days (long-term treatment). The selection of lithium diet was based on our previous work (4). Plasma levels of lithium in rats maintained on this diet were found to be 0.8 ± 0.07 meq/l in our previous study in the Wistar rats (4). All the saline-treated, short-term lithium-treated, and long-term lithium-treated animals were challenged with saline or clonidine (50 μg/kg). Saline or clonidine was injected IP between 1015 and 1100. The animals were sacrificed 30 min after saline or clonidine injection between 1045 and 1130.

The rats were sacrificed by decapitation, and trunk blood was collected in centrifuge tubes containing 0.5 ml of ethylenediaminetetraacetic acid (EDTA). Following centrifugation, plasma samples were collected and stored at −70°C. The plasma concentrations of GH were measured by radioimmunoassay as described elsewhere (7).

Drugs

The drugs, clonidine HCl (Research Biochemicals, Inc., Natick, MA) and lithium carbonate (Bio-serv, Frenchtown, NJ) were used in the study. Clonidine was dissolved in 0.9% saline. All drug doses given in the text refer to the salt.

Data Analysis

A two-way analysis of variance (ANOVA) was used to examine both the effects of lithium treatment and the effects of clonidine compared to a saline control. Bonferroni-corrected post hoc *t* tests were used after a significant overall treatment effect was found. In the above analysis, the $\log_{10}$ transformations of the GH data were used because of variance homogeneity in the raw data. All data are reported as mean ± 1 SEM.

RESULTS

The effects of saline treatment and short- and long-term lithium treatment on clonidine's effect on GH levels are shown in Fig. 1. ANOVA showed an overall significant clonidine effect, $F(1, 25) = 5.6$, $p < 0.05$, as well as a significant lithium treatment effect, $F(2, 26) = 8.96$, $p < 0.01$. Further analysis revealed that clonidine administration produced significant increases in GH levels in short- but not long-term lithium-treated or saline-treated animals (Fig. 1).

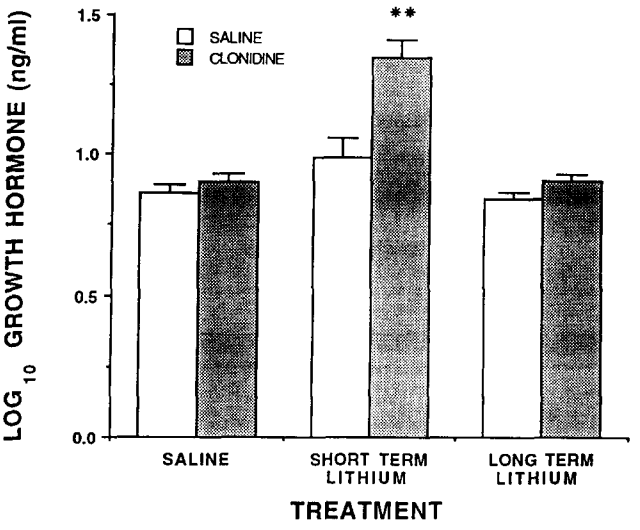

FIG. 1. Effects of saline or clonidine (50 μg/kg) on growth hormone levels in saline-treated, short-term lithium-treated, and long-term lithium-treated Fawn-Hooded rats. Values are expressed as means ± SEMs from six animals. $^{**}p < 0.01$, significantly different from control (saline + saline) animals.

DISCUSSION

The present study demonstrates that clonidine administration produced significant increases in GH levels in short-term lithium-treated but not saline-treated or long-term lithium-treated FH rats. Clonidine stimulates GH secretion by activation of α_2-adrenergic heteroreceptors present on 5-HT nerve terminals which, in turn, enhance 5-HT activity via stimulation of postsynaptic 5-HT_{1C} receptors to promote GH-releasing factor (1).

There are several behavioral (8), electrophysiological (5), food intake (4), and neuroendocrine (12) studies demonstrating enhanced 5-HT function in the brain following short-term lithium treatment. In biochemical studies, short-term lithium treatment has been shown to increase neuronal uptake of the 5-HT precursor tryptophan (10) and enhance the release of 5-HT from nerve endings (16). Thus, the ability of clonidine to significantly increase GH levels in short-term lithium-treated FH rats appears most likely due to enhanced concentrations of 5-HT at postsynaptic 5-HT_{1C} receptors, although enhanced sensitivity of α_2-heteroreceptors or 5-HT_{1C} receptors cannot be excluded. In contrast to short-term treatment, long-term lithium treatment causes downregulation of 5-HT_1 and 5-HT_2 receptors in the cortex, hippocampus, and striatum (9); decreases activity of the 5-HT synthetic enzyme tryptophan hydroxylase (10); desensitizes 5-HT autoreceptors mediating 5-HT release (17); and increases $G_{i\alpha}$ in the hypothalamus and hippocampus (11). These delayed changes may explain the failure of long-term lithium treatment to significantly enhance clonidine's effect on GH levels in the FH rats. Alternatively, changes in the catecholaminergic neurotransmitter system following lithium treatment (18) may be responsible for short- versus long-term lithium treatment differences. The possibility that long-term lithium treatment may have caused a phase shift in adrenergic, serotonergic, or GH rhythms seems unlikely, since long-term lithium treatment did not alter baseline GH levels in the present study.

In a previous report from this laboratory, we have demonstrated that similar treatment with lithium causes decreases in food intake and body weight gain and increases in water intake in Wistar rats (4). Therefore, it can be argued that these effects of lithium may be responsible for the observed increases in GH levels in the short-term lithium-treated FH animals in the present study. However, this possibility seems unlikely, since decreases in food intake and body weight gain occur during both short- and long-term lithium treatment (4), whereas only short-term lithium treatment produced increases in GH levels in the present study. Furthermore, short-term lithium treatment potentiates the effect of fenfluramine, which acts presynaptically to release serotonin (4), but not the effect of the postsynaptic 5-HT_1 receptor agonist m-chlorophenylpiperazine (m-CPP) on food intake (3). Therefore, potentiation of clonidine's effect on GH levels in FH rats following short-term lithium treatment appears most likely due to enhanced concentrations of 5-HT at postsynaptic 5-HT_{1C} receptors.

Finally, it is of interest to note that the FH rats have higher baseline levels of corticosterone and also manifest functional

subsensitivity to 5-HT agonists (2). Depressed patients have been reported to have higher baseline levels of cortisol and also manifest functional subsensitivity to 5-HT agonist-induced increases in plasma prolactin and hypothermia and, furthermore, also manifest blunted GH responses to clonidine compared to normal controls (15). Brain serotonin changes have been implicated in the etiology of affective illness and mode of action of antidepressant and antimanic drugs. Thus, the FH rat strain may prove to be a useful genetic model for depression and other neuropsychiatric disorders with possible abnormalities in serotonergic function. However, caution must be taken, since the therapeutic effects of lithium are manifested after long-term treatment, whereas only short-term lithium treatment enhanced clonidine's effect on GH levels in FH rats in the present study. On the other hand, it is of note that, compared to pretreatment, the prolactin response to L-tryptophan was significantly enhanced in 13 affective disorder patients after short-term (less than one week) lithium treatment, whereas no effect was observed after long-term (more than three weeks) lithium treatment (14). Thus, lithium's ability to enhance presynaptic 5-HT function might interact with postsynaptic 5-HT receptors sensitized by long-term tricyclic antidepressant treatment, resulting in greater improvement than would be obtained with either drug alone (5).

ACKNOWLEDGEMENT

The authors would like to thank Wilma Davis for her valuable assistance in the editing of this manuscript.

REFERENCES

1. Aulakh, C. S.; Hill, J. L.; Lesch, K. P.; Murphy, D. L. Functional subsensitivity of 5-hydroxytryptamine$_{1C}$ or alpha$_2$-adrenergic heteroreceptors mediating clonidine-induced growth hormone release in the Fawn-Hooded rat strain relative to the Wistar rat strain. J. Pharmacol. Exp. Ther. 262:1038–1043; 1992.
2. Aulakh, C. S.; Wozniak, K. M.; Hill, J. L.; DeVane, C. L.; Tolliver, T. J.; Murphy, D. L. Differential neuroendocrine responses to the 5-HT agonist m-chlorophenylpiperazine in Fawn-Hooded rats relative to Wistar and Sprague-Dawley rats. Neuroendocrinology 48:401–406; 1988.
3. Aulakh, C. S.; Zohar, J.; Wozniak, K. M.; Hill, J. L.; Murphy, D. L. Long-term lithium treatment in rats attenuates m-chlorophenylpiperazine-induced decreases in food intake but not locomotor activity. Psychopharmacology 98:448–452; 1989.
4. Aulakh, C. S.; Zohar, J.; Wozniak, K. M.; Hill, J. L.; Murphy, D. L. Differential effect of lithium treatment on fenfluramine-induced decreases in food intake and locomotor activity in rats. J. Psychopharmacol. 5:149–154; 1991.
5. Blier, P.; deMontigny, C. Short-term lithium administration enhances serotonergic neurotransmission: Electrophysiological evidence in the rat CNS. Eur. J. Pharmacol. 113:69–79; 1985.
6. deMontigny, C.; Cournoyer, G.; Morissetti, R.; Langlois, R.; Caille, G. Lithium carbonate addition in tricyclic antidepressant-resistant unipolar depression. Arch. Gen. Psychiatry 40:1327–1334; 1983.
7. Eden, S.; Albertson-Wikland, K.; Isaksson, O. Plasma levels of growth hormone in female rats of different ages. Acta Endocrinol. 88:676–690; 1978.
8. Grahme-Smith, D. G.; Green, A. R. The role of brain 5-hydroxytryptamine in the hyperactivity produced in rats by lithium and monoamine oxidase inhibitors. Br. J. Pharmacol. 52:19–26; 1974.
9. Hotta, I.; Yamawaki, S.; Segawa, T. Long-term lithium treatment causes serotonin receptor downregulation via serotonergic presynapses in rat brain. Neuropsychobiology 16:19–26; 1986.
10. Knapp, S.; Mandell, A. J. Short-term and long-term lithium administration: Effects on the brain's serotonergic biosynthetic systems. Science 180:645–647; 1973.
11. Lesch, K. P.; Aulakh, C. S.; Tolliver, T. J.; Hill, J. L.; Wolozin, B. L.; Murphy, D. L. Differential effects of long-term lithium and carbamazepine administration on $G_{s\alpha}$ and $G_{i\alpha}$ protein in rat brain. Eur. J. Pharmacol. 207:355–359; 1991.
12. Meltzer, H. Y.; Simonovic, M.; Sturgeon, R. D.; Fang, V. S. Effect of antidepressants, lithium, and electroconvulsive shock treatment on rat serum prolactin levels. Acta Psychiatr. Scand. 290:100–121; 1981.
13. Overstreet, D. H.; Rezvani, A. H.; Janowsky, D. S. Genetic animal models of depression and ethanol preference provide support for cholinergic and serotonergic involvement in depression and alcoholism. Biol. Psychiatry 31:919–936; 1992.
14. Price, L. H.; Charney, D. S.; Delgado, P. L.; Heninger, G. R. Lithium treatment and serotonergic function: Neuroendocrine and behavioral responses to intravenous l-tryptophan in affective disorder patients. Arch. Gen. Psychiatry 46:13–19; 1989.
15. Siever, L. J.; Trestman, R. L.; Coccaro, E. F.; Bernstein, D.; Gabriel, S. M.; Owen, K.; Moran, M.; Lawrence, T.; Rosenthal, J.; Horvath, T. B. The growth hormone response to clonidine in acute and remitted depressed male patients. Neuropsychopharmacology 6:165–177; 1992.
16. Treiser, S. L.; Cascio, C. S.; O'Donohue, T. L.; Thoa, N. B.; Jacobowitz, D. M.; Kellar, K. J. Lithium increases serotonin release and decreases serotonin receptors in the hippocampus. Science 213:1529–1531; 1981.
17. Wang, H. Y.; Friedman, E. Chronic lithium: Desensitization of autoreceptors mediating serotonin release. Psychopharmacology 94:312–314; 1988.
18. Wood, A. J.; Goodwin, G. M. A review of the biochemical and neuropharmacological actions of lithium. Psychol. Med. 17:579–600; 1987.