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Neonatal Treatment With Clomipramine Increased Immobility in the Forced Swim Test: An Attribute of Animal Models of Depression

JAVIER VELAZQUEZ-MOCTEZUMA[1] AND OSCAR DIAZ RUIZ

Departamento Biologia de la Reproduccion, Universidad Autonoma Metropolitana-Iztapalapa, Mexico City C.P. 09340, Mexico

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VELAZQUEZ-MOCTEZUMA, J. AND O. DIAZ RUIZ. *Neonatal treatment with clomipramine increased immobility in the forced swim test: An attribute of animal models of depression.* PHARMACOL BIOCHEM BEHAV **42**(4) 737-739, 1992.—The forced swimming test in rats has been identified as a suitable model for detecting antidepressant activity of several drugs regardless of their mode of action. On the other hand, a number of animal models of human endogenous depression have been proposed. Recently, it has been reported that perinatal administration of clomipramine in rats elicits behavioral changes in adulthood that resemble human endogenous depression. In the present study, we showed that in this new animal model of depression immobility was increased when animals were submitted to the forced swimming test. This finding supports the notion that the amount of immobility during the forced swimming test is directly proportional to a depressive state in the rat.

Forced swimming test Animal models Depression Clomipramine

IN the last decades, several animal models of psychiatric illness have been proposed as screening tests for new therapeutic drugs, as well as models to analyze the etiology, biochemistry, and symptomatology of human pathology [for review, see (7,18)]. Among the screening tests for antidepressant drugs, the most widely used and validated is the forced swimming test (FST), in which the immobility of the rat is decreased by those drugs having an antidepressant action and even by nonpharmacological antidepressant treatments such as REM sleep deprivation and electroconvulsive shock (1,2,10,11).

On the other hand, Mirmiran et al. (8) analyzed the effect of REM sleep deprivation during perinatal stages on development. They administered clomipramine to pups after birth and observed behavioral changes during adulthood. Vogel and Vogel (16) realized that these behavioral changes closely resemble the clinical signs of human endogenous depression. Thus, they proposed that neonatal treatment with clomipramine in rats can be used as a new animal model of human endogenous depression. The authors validated their proposal on the facts that rats display behavioral alterations, such as decreased sexual behavior (9), aggressiveness (12), and pleasure-seeking behaviors (13), as well as alterations in REM sleep parameters (15). Moreover, as in other animal models of depression these rats showed a positive response to antidepressant treatment (14).

In this study, we analyzed the possibility that the altered mood in this new animal model of depression would be detected by the FST as an increased amount of immobility.

METHOD

Wistar rats from our colony were used. The method to produce this new animal model of depression has been fully described elsewhere (9). In brief, 3 days postnatally male pups were cross-fostered and from days 8-21 they were treated with subcutaneously injected clomipramine hydrochloride (Anafranil, CIBA-GEIGY Corp.) (15 mg/kg) (n = 14) or saline (n = 11) twice daily. Female pups were eliminated from the experiment. At day 28, subjects (Ss) were separated from their foster mothers and housed in a room with a 12 L:12 D cycle (lights on 1000 h) with food and water available ad lib. Each cage contained rats with the same treatment. At 6 months of

[1] Address requests for reprints to Javier Velazquez-Moctezuma, M.D., Departamento Biologia de la Reproduccion, Universidad Autonoma Metropolitana, Col. Vicentina. Del. Iztapalapa, Mexico City C.P. 09340, A. P. 55535 Mexico.

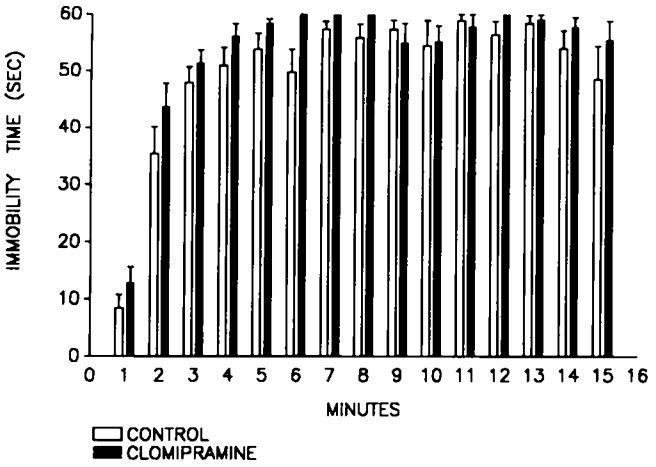

FIG. 1. Immobility time display during the forced swimming test by adult male rats neonatally treated with clomipramine or saline: 15-min test.

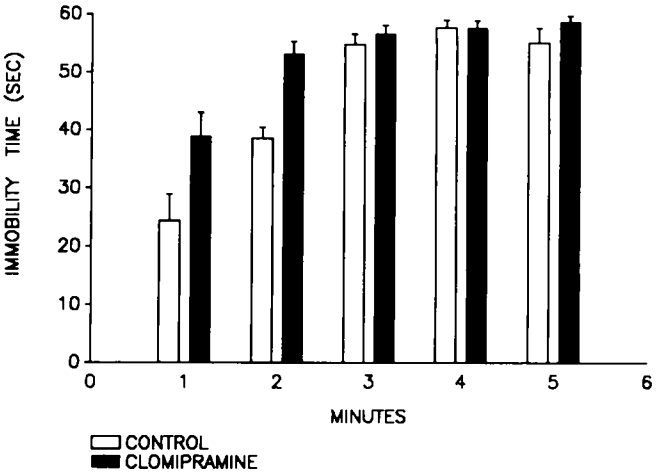

FIG. 2. Immobility time display during the forced swimming test by adult male rats neonatally treated with clomipramine or saline: 5-min test.

age, all Ss were submitted to the FST, which consists of dropping the rat into a glass cylinder containing water up to 15 cm high and maintained at 25°C. Animals remain in the water for 15 min on the first day and for 5 min 24 h later. According to the criteria previously reported (10), the animal was judged immobile when it was passively standing up, making only those small movements to keep its head above the water. The observer was blind to treatment conditions. Data were statistically analyzed using Student's *t*-test.

RESULTS

Figure 1 shows the results obtained from the 15-min test. As can be seen, almost all control values are lower than those displayed by the neonatally treated group. A similar effect was observed during the 5-min test 24 h later (Fig. 2). When the total amount of time between groups was compared, there were significant differences in the 15-min test, $t(23) = 2.176$, $p < 0.04$, as well as in the 5-min test, $t(23) = 3.5455$, $p < 0.001$.

Immobility time in FST is dependent upon several methodological factors, strain included (1). Nevertheless, the mean values of control rats reviewed in the literature by Borsini gives a value of 231 s (1). The average of our control values gives a mean of 230.7. However, when compared with the data originally reported by Porsolt (10) our control values are higher during the 15-min test (19%) and during the 5-min test (12%). Even with these high control values, FST detected an increase in immobility time of the experimental group.

Fecal boli were quantified during FST as an index of the stress impact of the test situation (4). There were no significant differences between groups in either the 15-min or 5-min test (Table 1).

DISCUSSION

It is well known that the immobility time observed during FST is decreased as a response to antidepressant treatments (1,10). Moreover, submissive mice display more immobility during the FST than that displayed by dominant mice (6). A similar immobility time increase was observed in rats exposed to an uncontrollable stressor (17). These data support the notion that when the immobility time observed in FST increases it is reflecting a state of behavioral depression in the rat.

Regarding the new animal model of endogenous depression proposed by Vogel et al. (14), this study adds another argument in agreement. There is a strong congruence between the present results and those obtained in rats neonatally treated with other different monoamine uptake inhibitors when they were submitted to the FST (3,5).

Thus, it seems that immobility time can be a valuable moni-

TABLE 1

FECAL BOLI AND TOTAL AMOUNT OF IMMOBILITY TIME DISPLAY BY ADULT RATS NEONATALLY TREATED WITH SALINE OR CLOMIPRAMINE SUBMITTED DURING 15 OR 5 MIN TO THE FORCED SWIMMING TEST

	Immobility Time (seconds)		Fecal Bolt (counts)	
	FST 15	FST 5	FST 15	FST5
Saline (n = 11)	747.45 ± 23.56	230.72 ± 6.64	5.63 ± 0.82	4.63 ± 0.6
Clomipramine (n = 14)	804.21 ± 13.98*	265.42 ± 6.92†	4.57 ± 0.54	4.21 ± 0.54

Values are mean ± SEM.
*$p < 0.05$ compared to saline with *t*-test.
†$p < 0.001$ compared to saline with *t*-test.

tor to assess mood state in the rat. Therefore, FST can be a suitable screening test for new antidepressant drugs and can also be used to further validate new proposals of animal models of human depression.

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