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Induced Depressive behavioral studies in male and female rats through forced swimming test

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Abstract
Albino Wister rats provide a model for stress induced depressive behavior. This is depicted in the form of being immobile for several times in the forced swimming test session (FST). The present study examined the differences in immobile behavior of male and female mice in cylinder of two diameters (26 cm and 32 cm) used in forced swimming test. So FST in case of male rats of diameter 26 cm. 61 stops were observed in total 15 minutes, and in case of female were observed 36 stops in 15 minutes. The results obtain from the study exist on the influence of psychological variables on male and female ratio of induced depression because of age and sex specific population.

Keywords: Strain differences, sex difference in depressive behavior, induced depression

Introduction
Depression has been defined as a disturbance in normal behavior or simply it is a group of specific disturbances. There are two types of symptoms observed in depression: Emotional symptoms: sadness, lack of interest, guiltiness and ultimately suicidal thought. Physical symptoms: sleep disorders, pain, headache, gastrointestinal disorders, lack of energy (fatigue), and change in psychomotor functions. Physical conditions are often underestimated as compared to emotional conditions. Biological etiology of depression is hypothesized due to deficiency in *monoaminergic neurotransmitters* and more specifically deficiency in *nor-epinephrine (NE)* and *serotonin* (*5- hydroxytryptamine, 5HT).*The forced swimming test (FST) is a behavioral task used in rodents that predicts the efficiency of antidepressant treatments in humans. The FST was first of all described by Porsolt *et al.* (1977) [6] as a new behavioral method for inducing a depressed state in mice. This idea arose only by chance when they were doing some warning experiments with rats in water maize. Most rats were finding the exit within ten minutes. But they also noticed that some other rats stopped movements and remained immobile i.e. floating passively Porsolt *el al.* (1979) to describe this new behavioral model in mice, the following procedures was adopted.

Rodents are placed in a cylinder of water for an extended period of time, the length of time rodents remain immobile doing only those movements necessary to keep its head above the water, it measured. This immobilization is actually a state of induced depression in rodents which is a symptom seen in depression. Antidepressant drug treatment reduces this immobility time and thus results in increased activity. The purpose of our study is to compare the induced depressive behavior in male and female albino Wister rats and the effect of area of swimming (diameter of cylinders) through forced swimming test (FST) in cylinder of different diameters. The forced swimming test appears to be suitable for detecting antidepressant activity in rats not in mice. The obstacle in studying of predicting antidepressant activity of new compounds is represented by deficiencies in our understanding of human psychopathology which in turn preclude duplication of behavioral manifestations of such disorders in rodents. Actually, two approaches have been used to study antidepressants (Katz 1981): to find experimental conditions in which known antidepressants exert some pharmacological effects, and to reproduce in animals the human symptomatology of depression (theoretical models). Among the various theoretical models (see Willner 1984) [9], the forced swimming test (FST) (Porsolt *et al*. 1977b) [8] is widely used and much information is now available about it. The aim of present paper is to review the literature critically in order to understand its predictive value. On the other hand, it seems that rats are less fearful on subsequent immersion than on the previous one, suggesting that behavioral immobility is a consequence of adaptive response to a stressful situation more than "despair" (Hawkins *et al.* 1978) [11].

Material and Methods

The study was conducted in research laboratory of SHUATS for determining the induced depressive behavior of male and female Albino Wister rats and effects on the area of swimming. Rats are subjected to number of trials during which they are forced to swim in an acrylic/plastic/glass cylinder filled with water up to a height of 22 cm, from which they cannot escape. The tests sessions were performed for 15 minutes with intervals of 24 hrs.

In our study, 12 male and 12 female albino Wister rats weighing between 149-175gm, were housed in six polycarbonate cages in groups of four (2 males and 2 females). These rats were exposed to 12 hr cycle of light and dark (circadian rhythm).

Note: The experiments were performed according to the guide lines for care and use of laboratory animals and ethical committee of University of Allahabad, U.P., India for experiments on animals.

Forced swimming test (FST): This procedure, which we have used, has been previously described by Porsolt *et al.* (1977) [6] (Porsolt *et al*., Detke, Reneric). Each male and female rat was forced to swim inside two cylinder of different diameter (11cm, 26 cm and 32 cm) filled with water at 25 ºC without any possibility of escaping for 15 minutes with 24 hrs interval. The water was changed after every test session. The resulting anxiety produces vigorous swimming activity and attempt for escaping by diving or climbing the walls of cylinder. When the animal stops all the movements except those necessary for survival (keeping the head above the water), the behavior was consider to be immobile. This has been defined as induced depression. Immobility and number of stops was measured (in sec.). The test session were video taped for scoring the movements and studying behavior of rats (Scott *et al*., 1973) [5].

Results and Discussion

Albino Wistar Rat model was used for the present investigation with constants were made as temperature: 250C, height of water: 22 cm and time period of test session was15 minutes and diameter of cylinder was used as variable.

1. FST in cylinder of diameter 26 cm

12 young males were subjected to FST individually for 15 minutes and the findings were recorded in three periods of 5 minutes each. Average of 21 stops observed in first 5 minutes. in second period of 5 minutes 21 stops were observed. In third period of 5 minutes 19 stops were observed. So overall 61 stops were observed in total 15 minutes. In case of females, in first 5 minutes only 6 stops were observed, in second 5 minutes 13 stops and in third 5 minutes 17 stops were observed. Thus total 36 stops in females have been observed in 15 minutes as shown in Table 1.

After 24 hr. in male rats 10, 24, 30 stops were recorded in first, second and third 5 minutes of period respectively. In females, 5, 5and 20 stops were observed in first, second and third 5 minutes of period respectively. So total of 30 stops in 15 minutes as shown in Table 2.

Again after 24 hr. in males 20, 30, 37 stops were observed in first, second and third 5 minutes of period respectively. Total of 87 stops were observed in 15 minutes as shown in Table 3. In females. In females, 10, 17and 22 stops were observed in first, second and third 5 minutes of period respectively. So total of 49 stops in 15 minutes as shown in Table 4.9 figure 1 and 2).

Table 1: FST in cylinder of diameter 26 cm

18th December 16							
Male				**Female**			
Time considered ->	**1st 5 minutes**	**2nd 5 minutes**	**3rd 5 minutes**		**1st 5 minutes**	**2nd 5 minutes**	**3rd 5 minutes**
No. of stops	21	21	19	No. of stops	6	13	17
Total stops in 15 minutes	61			Total stops in 15 minutes	36		

Table 2: FST in cylinder of diameter 26 cm

22nd December 16							
Male				**Female**			
Time considered ->	**1st 5 minutes**	**2nd 5 minutes**	**3rd 5 minutes**		**1st 5 minutes**	**2nd 5 minutes**	**3rd 5 minutes**
No. of stops	10	24	30	No. of stops	5	5	20
Total stops in 15 minutes	64			Total stops in 15 minutes	30		

Table 3: FST in cylinder of diameter 26 cm

23rd December 16							
Male				**Female**			
Time considered ->	**1st 5 minutes**	**2nd 5 minutes**	**3rd 5 minutes**		**1st 5 minutes**	**2nd 5 minutes**	**3rd 5 minutes**
No. of stops	20	30	37	No. of stops	10	17	22
Total stops in 15 minutes	87			Total stops in 15 minutes	49		

Table 4: FST in cylinder of diameter 26 cm

6th January 17							
Male				**Female**			
Time considered ->	**1st 5 minutes**	**2nd 5 minutes**	**3rd 5 minutes**		**1st 5 minutes**	**2nd 5 minutes**	**3rd 5 minutes**
No. of stops	16	19	25	**No. of stops**	10	13	18
Total stops in 15 minutes	60			Total stops in 15 minutes	41		

2. FST in cylinder of 32 cm. of diameter

In male rats 28 stops in first 5 minutes, 45 stops in second 5 minutes and 48 stops in third 5 minutes were observed. So total of 121 stops were observed in 15 minutes. in female rats 13, 27, 32 stops were observed in first, second and third 5 minutes of period respectively. Thus total off 52 stops were observed in 15 minutes test session as shown in Table 5.

In second session i.e., after 24 hrs. In male rats, 25 stops in first 5 minutes period, 28 stops in second 5 min. period, and 39 stops in last 5 min. period were observed. Total 92 stops in 15 min. were recorded. Whereas in female rats 20 stops in first five mint period 21 stops in second five mints and 24 stops in last five mints were observed. Total 65 number of stops were observed in total 15 mints as shown in Table 6.

In third session (after 24 hrs.) in male rats, 27 stops in first 5 mints, 50 in second 5 mints and 58 in last 5 mints are recorded making the total 135 stops in 15 mints as shown in Table 7

After 24 hrs. in fourth session in males 30 stops in first five mints, 55 in second 5 mints and 60 in last five mints were observed making total of 145 no. of stops in 15 mints. Where as in case of females 24 stops in first 5 mints, 30 stops in second 5 mints. And 50 in last five mints were recorded making 104 total numbers of stops in 15 mints of FST as shown in Table 8. Figure 3 and 4

Table 5: FST in cylinder of diameter 32 cm

24th December16							
Male				**Female**			
Time considered ->	**1st 5 minutes**	**2nd 5 minutes**	**3rd 5 minutes**		**1st 5 minutes**	**2nd 5 minutes**	**3rd 5 minutes**
No. of stops	28	45	48	No. of stops	13	27	32
Total stops in 15 minutes	121			Total stops in 15 minutes	52		

Table 6: FST in cylinder of diameter 32 cm

29th Decembe16							
Male				**Female**			
Time considered ->	**1st 5 minutes**	**2nd 5 minutes**	**3rd 5 minutes**		**1st 5 minutes**	**2nd 5 minutes**	**3rd 5 minutes**
No. of stops	25	28	39	No. of stops	20	21	24
Total stops in 15 minutes	92			Total stops in 15 minutes	65		

Table 7: FST in cylinder of diameter 32 cm

31st December16							
Male				**Female**			
Time considered ->	**1st 5 minutes**	**2nd 5 minutes**	**3rd 5 minutes**		**1st 5 minutes**	**2nd 5 minutes**	**3rd 5 minutes**
No. of stops	27	50	58	No. of stops	25	30	45
Total stops in 15 minutes	135			Total stops in 15 minutes	100		

Table 8: FST in cylinder of diameter 32 cm

1st January 17							
Male				**Female**			
Time considered ->	**1st 5 minutes**	**2nd 5 minutes**	**3rd 5 minutes**		**1st 5 minutes**	**2nd 5 minutes**	**3rd 5 minutes**
No. of stops	30	55	60	No. of stops	24	30	50
Total stops in 15 minutes	145			Total stops in 15 minutes	104		

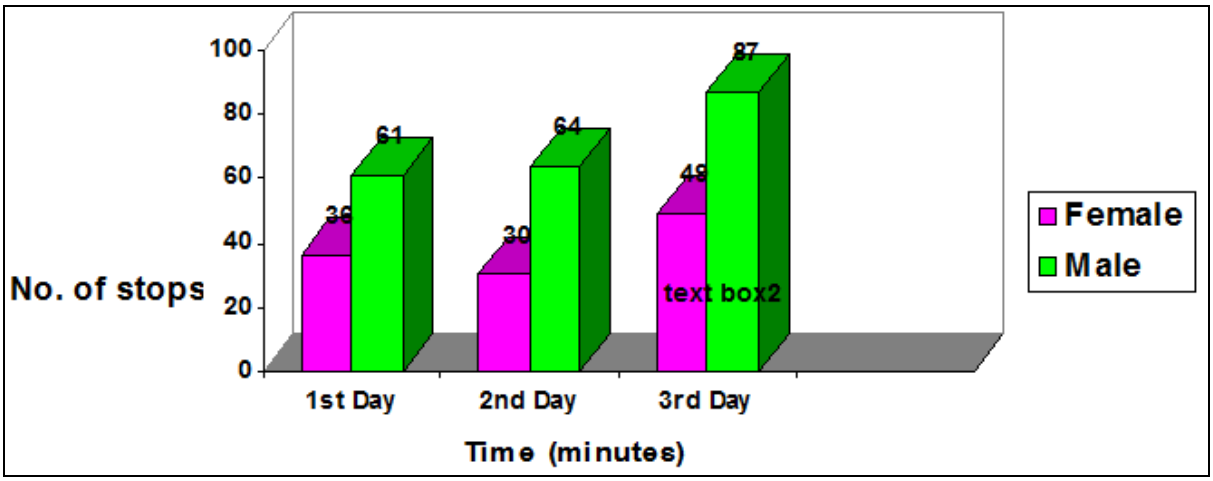

Fig 1: Showing comparative no. of stops between male and female in three consecutive days (Cylinder of 26 cm. Diameter)

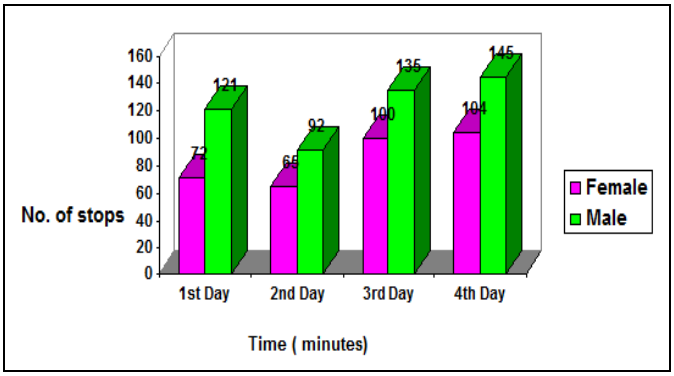

Fig 2: Showing comparative no. of stops between male and female in three consecutive days (Cylinder of 26 cm. Diameter)

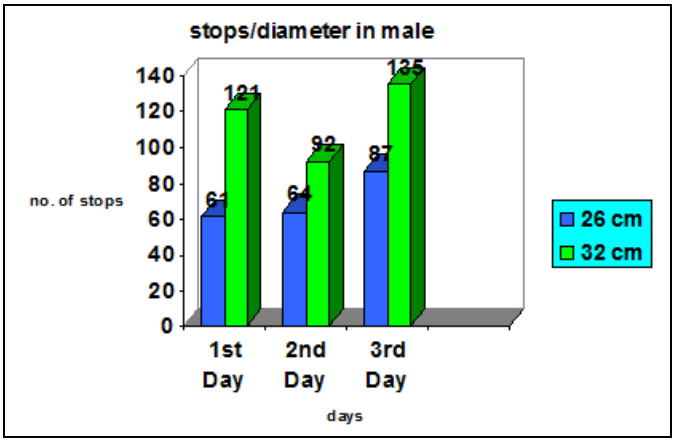

Fig 3: Showing comparative no. of stops between male and female in four consecutive days (Cylinder of 32 cm. Diameter)

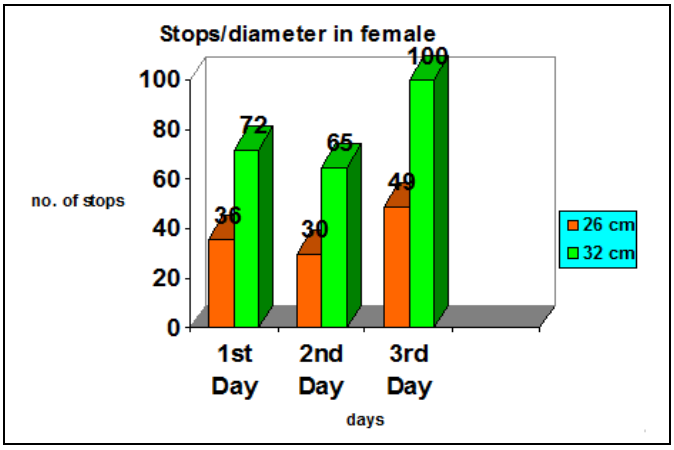

Fig 4: Chart showing no. of stops in two different diameters (26 cm & 32 cm) in male in three days.

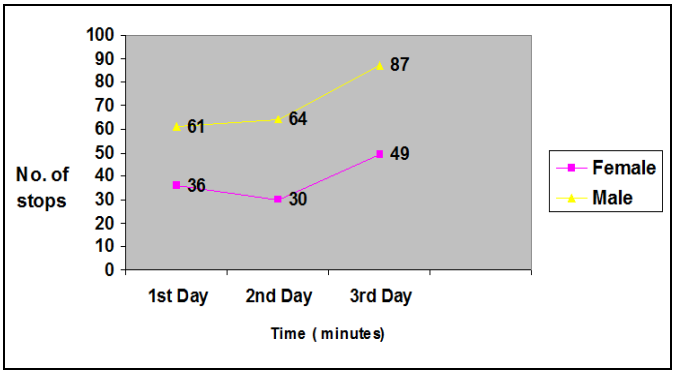

Fig 5: Chart showing no. of stops in two different diameters (26 cm & 32 cm) in female in three days.

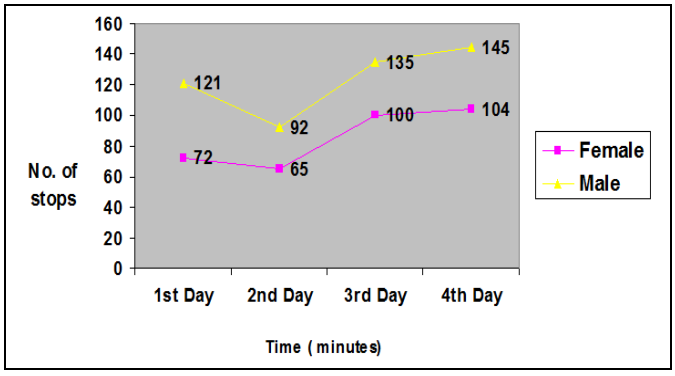

Fig 6: Line Chart showing comparative no. of stops between male and female in three consecutive days (Cylinder of 26 cm. Diameter)

2) Line chart showing comparative no. of stops between male and female in three consecutive days (Cylinder of **32 cm**. Diameter)

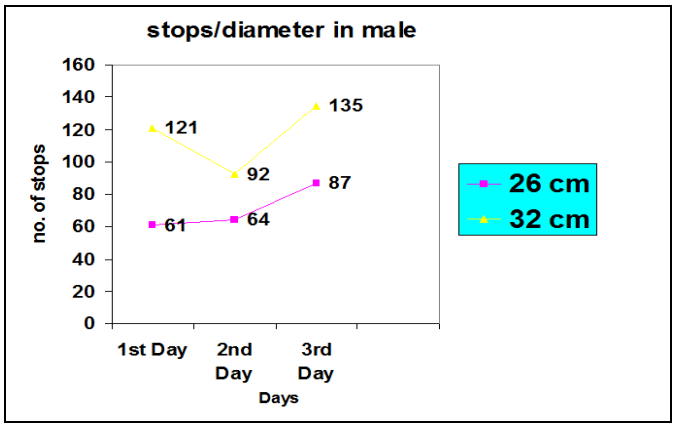

Fig 7: Line Chart showing comparative no. of stops in two different diameters (26 cm & 32 cm) in male in three days.

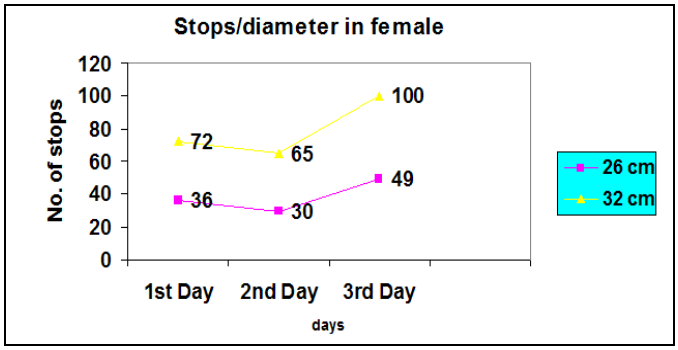

Fig 8: Line Chart showing comparative no. of stops in two different diameters (26 cm & 32 cm) in Female in three days

Exit data on the psychosocial variables on the female/male ratio of induced depression because of the small number of cases and Age and sex specific population based depression rates were calculated. In reviewing published studies on age specific sex differences in depression, Jorn [1] concluded that the broad claim that depression is more common in women than in men is an oversimplification. He found that reflecting the finding that the difference between male and female rates are rather small in young age, increase towards mid-life and decrease thereafter.

Thus, while among the married, rates were usually lower than among the not married, the gender gap was larger at a much earlier age than in the total population. This favors a psychosocial interpretation (marriage being more stressful for

women than for men), but still, a biological interpretation is not ruled out, since married women may rather have children -and child bearing is related to hormonal changes (there was no possibility in our analyses to find out how frequently post-partum depression occurred).

Among the not married (taken as a group, including the never married, the divorced and the widowed) the inverted U-shape curve was found only in a rudimentary form, with a decline after the age of 40 (from around 2.8:1 to 1:1 at the age of 50 and later). It is difficult to interpret this curve, also if considering that the subgroup of the never married corresponded to the total group of the not married (for divorced and widowed persons the analysis could not be carried out due to the small numbers of cases in these subgroups if divided into single age groups.) Employment status also changed the general pattern of an inverted U-shape found for the total population to a substantial degree. Among the employed the female/male ratio was constant (below 2:1) from the age of 18 to 50, and dropped thereafter to 1:1. The inverted U-shape virtually disappeared in this analysis. It seems that up to the age of fifty the raised risk for employed women was not age-dependent on age; the female preponderance tended to disappear beyond the age of 50. The not employed showed a pattern which was the opposite to that observed in the total population – there is a U-curve – but not an inverted one, but an upright U, i.e. in midlife not employed men had a clearly higher risk than not employed women.

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