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Sex differences in forced-swim and open-field test behaviours after chronic administration of melatonin

Lori A. Brotto, Alasdair M. Barr, Boris B. Gorzalka*

Department of Psychology, The University of British Columbia, Vancouver, BC, Canada V6T 1Z4

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Abstract

The effects of melatonin administered chronically on forced-swim test and open-field test behaviours were examined in male and female rats. The forced-swim test has been shown to be sensitive to all major classes of antidepressants and evidence indicates that melatonin possesses putative antidepressive properties. Male and female Long-Evans rats received either a regimen of chronic administration of melatonin or the control condition for 14 days via the drinking water. On day 15, each animal was individually introduced into a swim chamber, and was scored for 15 min on the duration of swimming, struggling, and immobility. After 24 h, each animal was again tested in the forced-swim test for 10 min. On day 18, all animals were tested in the open-field test apparatus for 5 min. Results revealed that females consistently showed higher activity levels than males in the forced-swim and open-field tests. Melatonin significantly increased struggling in males on day 15, but failed to do so in females. Also, whereas melatonin-treated females showed higher levels of behavioural immobility during their first exposure to the forced-swim test, this effect was prevented upon a second exposure. In both males and females, melatonin decreased swimming in the forced-swim test while increasing open-field ambulatory behaviour. Therefore, it is unlikely that melatonin's mechanism of action is a general inhibitory effect on motor activity. Taken together, the results suggest that the effects of melatonin treatment on forced-swim test behaviours are sex- and test-dependent.

Keywords: Melatonin; Forced-swim test; Antidepressant; Sex difference; Depression

1. Introduction

Sex differences in the diagnosis of major depressive disorder in humans (Bracke, 1998) and in the relative efficacy of various antidepressants (Godfroid, 1999) have frequently been reported. The results of pharmacological investigations have revealed differences between men and women in, for example, binding capacities for paroxetine (Marazziti et al., 1998), hormonal responses to desipramine (Filip et al., 1989), and metabolism rates for nefazodone (Barbhaiya et al., 1996). The animal literature reveals parallel findings in that there emerge sex differences in animal models of depression and in responses to antidepressants. Although most investigations of behavioural responses to antidepressants in nonhuman species have employed males, the few existing studies to include female subjects demonstrate that males and females respond either differently, or even in opposite directions in animal models of depression (Alonso et al., 1991; Barros and Ferigolo, 1998). These apparent sex differences in response to antidepressants may be attributable, in part, to varying steroid levels. Estradiol can exert profound effects on mood by acting on neurotransmitter systems that are implicated in depression (Fink et al., 1996).

It has been suggested that the hormone, melatonin, possesses therapeutic benefits to individuals suffering from depression (Halbreich, 1997) although controlled, clinical trials have not been performed. However, empirical trials have demonstrated that in patients with major depressive disorder, either current or in remission, serum melatonin levels at nighttime are greatly reduced compared to non-psychiatric patients (Beck-Friis et al., 1984), and are reduced even further in patients who have undergone a

* Corresponding author. Tel.: +1-604-822-3095; fax: +1-604-922-6923.

E-mail address: bgorzalka@neuron3.psych.ubc.ca (B.B. Gorzalka).

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traumatic life event (Beck-Friis et al., 1985). In fact these authors have proposed that low nocturnal melatonin may be a trait-dependent marker for depression. Pharmacological investigations have also supported the notion of an antidepressive potential for melatonin (Datta and King, 1980; Eison et al., 1995).

Attempts have been made to model human depressive symptomatology in the rodent (Porsolt et al., 1978) and the efficacy of pharmacological treatments has been explored in this context. In particular, the forced-swim test is currently one of the most frequently used behavioural tests for investigating antidepressant potential (Lucki, 1997). The test measures the ability of antidepressants to reduce the occurrence of behavioural immobility, which is considered representative of passivity in response to stress, and to increase swimming and struggling behaviours, which may represent escape-directed, or coping behaviour (Lucki, 1997). The commonly employed paradigm involves two exposures to the swim chamber: a 15-min test followed 24 h later by a 10-min test. Effective antidepressants reduce the amount of behavioural immobility and increase the display of active behaviours (swimming and struggling) during the second test (Porsolt et al., 1978). It has been demonstrated that the forced-swim test is sensitive to all major classes of antidepressants (Porsolt et al., 1978), while still maintaining specificity to antidepressants with particular modes of action. For example, administration of selective serotonin reuptake inhibitors increases swimming behaviour and decreases immobility, whereas selective noradrenergic reuptake inhibitors increase struggling and decrease immobility (Detke et al., 1995). In order to rule out pharmacological effects on general motor activity that might account for behavioural patterns in the forced-swim test, the open-field test is often used in conjunction with the forced-swim test to assess locomotor activity. Typical behaviours measured in the open-field test include ambulatory behaviour (number of lines crossed) and rearing behaviour.

The forced-swim paradigm has already been employed to test the effectiveness of melatonin. For example, the Flinders sensitive line, a genetically bred rat that shows high levels of immobility in the forced-swim test and exhibits other behavioural patterns that are analogous to those of depressed patients, has been used to screen for antidepressant potential (Overstreet et al., 1998). After chronic administration of the melatonin receptor agonist, S20304, immobility in the forced-swim test was reduced in Flinders sensitive line rats (Overstreet et al., 1998). In mice, a one-trial forced-swim paradigm is used, whereby animals are exposed to the swim chamber for a single 5- or 10-min exposure. Acute treatment with melatonin (10 mg/kg) significantly reduced immobility in mice (Shaji and Kulkarni, 1998) but higher doses of melatonin (20, 30 and 50 mg/kg) were ineffective (Shaji and Kulkarni, 1998; Dubocovich et al., 1990). Taken together, these studies suggest that melatonin might possess antidepressive potential. However, these findings are limited to male rodents since studies on female behaviour in the forced-swim test have yet to be performed.

A major purpose of the current study is to compare the effects of melatonin on forced-swim behaviour in male and female rats. Secondly, this will be the first study to examine the effectiveness of chronically administered, low doses of melatonin in rats. In previous studies, acute doses of melatonin as well as chronic doses of a melatonin agonist were both found to decrease behavioural immobility. However, the effects of chronic administration of melatonin on forced-swim behaviours have not been examined. It has been argued that a dosing regimen similar to that employed with antidepressants in humans may increase the external validity of forced-swim test findings (Borsini and Meli, 1988), and as such, a chronic mode of administration was chosen in the current study. Thirdly, this study will determine if melatonin exerts differential behavioural effects depending on whether animals had pre-exposure to the forced-swim test. This follows from recent evidence that the effect of the antidepressant fluoxetine in the forced-swim test depends on prior exposure to the test (Taghzouti et al., 1999). Finally, open-field behaviour will also be scored in order to assess any general effects on locomotor activity.

2. Materials and methods

2.1. Subjects

Sixteen female (300–400 g) and 20 male (600–800 g) Long-Evans rats, originally obtained from Charles River, Quebec, were used in this experiment. Naïve rats were 13 months of age, and were housed in groups of 3–4, by sex in standard triple-wire-mesh cages. Colony conditions were temperature-controlled (21 ± 1°C) and lights were set on a 12-h dark/12-h light reversed cycle with lights off at 0900 h. Animals were provided with Purina Rat Chow and tap water ad libitum.

2.2. Treatment

Animals were randomly assigned to receive either chronic treatment with melatonin or the vehicle, resulting in equal numbers of animals in each condition ($n = 8$ females and $n = 10$ males per condition). Male and female rats were administered melatonin and participated concurrently in behavioural testing. Melatonin was administered via the drinking water in a manner previously described (Oaknin-Bendahan et al., 1995). Melatonin (4 mg) was obtained from Sigma (Chicago, USA), and was dissolved in 0.1 cm^3 dimethylsulfoxide (DMSO). Once dissolved, the solution was mixed vigorously in 1-l water, resulting in a final melatonin concentration of 4 μg/ml. Rats in the control condition received 0.1 cm^3 DMSO dissolved in 1-l

water. Each cage was supplied with two 500-ml water bottles, and fresh solutions were prepared every 2 days for 18 days. The average fluid consumption was 35 ml/day for male rats and 30 ml/day for female rats, corresponding to a total melatonin intake of 2.48 and 2.12 mg for males and females, respectively. There were no significant differences in fluid consumption between animals in the experimental and control groups. All procedures were conducted in accordance with the Canadian Council on Animal Care guidelines for work with laboratory animals.

2.3. Forced-swim test procedure

The design of the forced-swim test was adapted from Hansen et al. (1997). On day 15 of melatonin administration, rats were placed in a cylindrical glass container (45-cm height, 28-cm diameter) which contained tap water (25 ± 1°C) to a depth adjusted for the weight of the individual animal, so that its hind paws could just touch the bottom of the container: 350–375 g (19.6 cm), 375–400 g (20.6 cm) and >400 g (21.6 cm). The water was changed after each subject was tested and the cylinder was thoroughly rinsed in order to remove the presence of any potential alarm substances (Abel and Hannigan, 1992). During test 1, rats were placed in the water for 15 min, and 24 h later they were retested in the water for 10 min (test 2). After testing, subjects were dried with a towel and returned to their home cages.

The test sessions were recorded on videotape, and later scored by an observer who was blind to the condition of each animal. Test behaviours were subsequently scored according to the criteria of Armario et al. (1988) which include (1) struggling — characterized behaviourally as when the rat struggles to get out of the container, with its forepaws above the surface of the water, (2) swimming — defined by when the subject makes active swimming motions, and (3) immobility — described as when three paws are immobile and the fourth paw exhibits only minimal movement. The presence of depressive activity is inferred from a significant reduction in the amount of time spent in either of the two types of active behaviour, i.e. either struggling or swimming.

2.4. Open-field test procedure

Forty-eight hours after test 2, on day 18, all subjects were tested in the open-field apparatus. The apparatus consisted of a square arena (120×120 cm), with a 40-cm high, opaque, white wall. The floor was marked into 16 equal segments, and three fluorescent lights provided diffuse overhead illumination (20 lx at the level of the arena). The animals were tested in a quiet room, and locomotor activity over a 5-min period was recorded on a ceiling-mounted videocamera (Tracker VP200; HVS Image, Hampton, England). Videotapes were later scored by an experienced observer who was blind to the condition of the animals, and measured for the number of segments crossed by the animal (defined as at least three paws in a quadrant) and the number of rears (defined as the animal standing upright on its hind legs).

2.5. Statistical analyses

Separate two-way Analyses of Variance (ANOVA) were performed on data from each of tests 1 and 2 with drug condition and sex as independent, between-subjects factors. For open-field test behaviours, each of crossing and rearing were subjected to separate two-way ANOVAs with treatment and sex as independent factors. In all cases, a P-level < 0.05 was considered significant.

3. Results

3.1. Forced-swim test — test 1

3.1.1. Struggling behaviour

Statistical analyses revealed a significant interaction between melatonin and sex for struggling behaviour, $F(1,32) = 4.05$, $P = 0.05$, but no significant effects of melatonin, $F(1,32) = 0.55$, $P = 0.46$, or sex, $F(1,32) = 1.87$, $P = 0.18$. Fisher's LSD test for pairwise comparisons revealed that melatonin significantly facilitated struggling behaviour in males, $P = 0.047$, as shown in Fig. 1. Also, in the absence of melatonin, females displayed higher overall levels of struggling than males, $P = 0.023$.

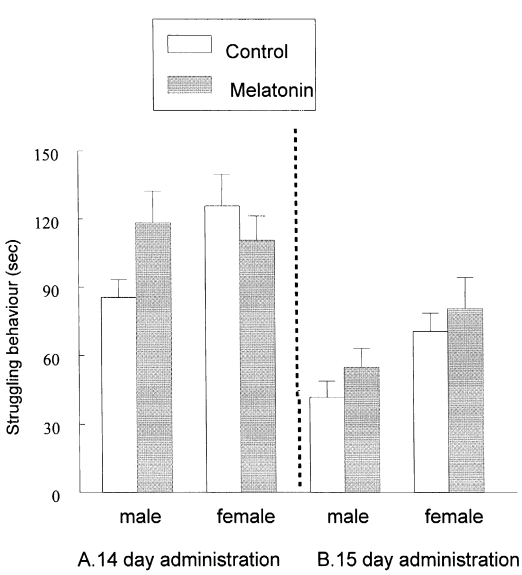

Fig. 1. Effects of melatonin administration on struggling behaviour (s) in male and female rats after 14 days of administration (panel A) and 15 days of administration (panel B) in the forced-swim test. Results demonstrate a significant interaction between melatonin and sex after 14 days of administration, $F(1,32) = 4.05$, $P = 0.05$, and a significant main effect of sex after 15 days of administration, $F(1,32) = 8.59$, $P = 0.006$. Data represent means $\pm$ S.E.M.

3.1.2. Swimming behaviour

On swimming behaviour, statistical analyses revealed that the interaction between melatonin and sex did not reach significance, $F(1,32) = 0.25$, $P = 0.62$, nor did the main effect of sex, $F(1,32) = 2.80$, $P = 0.104$. The main effect of melatonin, however, was statistically significant, $F(1,32) = 5.79$, $P = 0.02$. Inspection of Fig. 2 shows that all animals treated with melatonin, regardless of sex, spent significantly less time swimming than control animals.

3.1.3. Immobility behaviour

ANOVA tests determined no statistical interaction between melatonin and sex, $F(1,32) = 2.16$, $P = 0.15$ on behavioural immobility. However, main effects of melatonin, $F(1,32) = 4.46$, $P = 0.04$, and sex, $F(1,32) = 4.59$, $P = 0.04$, were statistically significant. As shown in Fig. 3, males showed higher levels of behavioural immobility than females, and treatment with melatonin induced higher levels of immobility, but apparently to a much greater degree in females.

3.2. Forced-swim test — test 2

3.2.1. Struggling behaviour

Statistical analyses of struggling behaviour revealed no significant interaction between melatonin and sex, $F(1,32) = 0.03$, $P = 0.87$, nor a significant main effect of melatonin, $F(1,32) = 1.53$, $P = 0.23$. The main effect of sex, however, was statistically significant, $F(1,32) = 8.59$, $P = 0.006$, with females showing higher levels of struggling behaviour than males (Fig. 1).

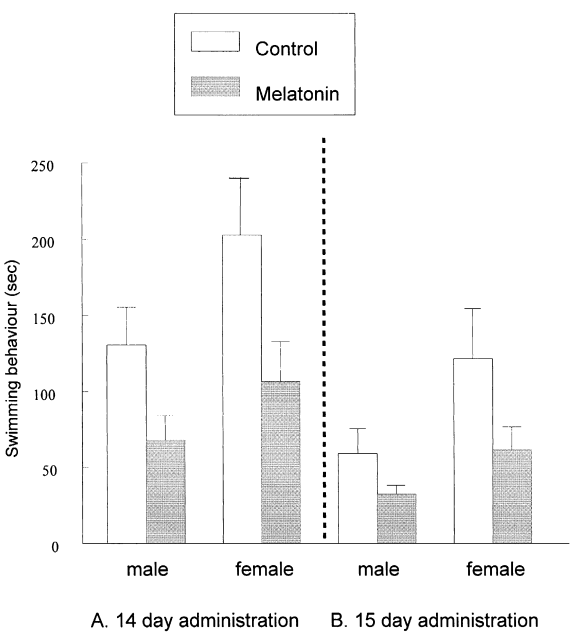

Fig. 2. Effects of melatonin administration on swimming behaviour (s) in male and female rats after 14 days of administration (panel A) and 15 days of administration (panel B) in the forced-swim test. Results demonstrate a significant main effect of melatonin after 14 days of administration, $F(1,32) = 5.79$, $P = 0.02$, and significant main effects of melatonin, $F(1,32) = 5.47$, $P = 0.03$, and sex, $F(1,32) = 6.09$, $P = 0.02$, after 15 days of administration. Data represent means ± S.E.M.

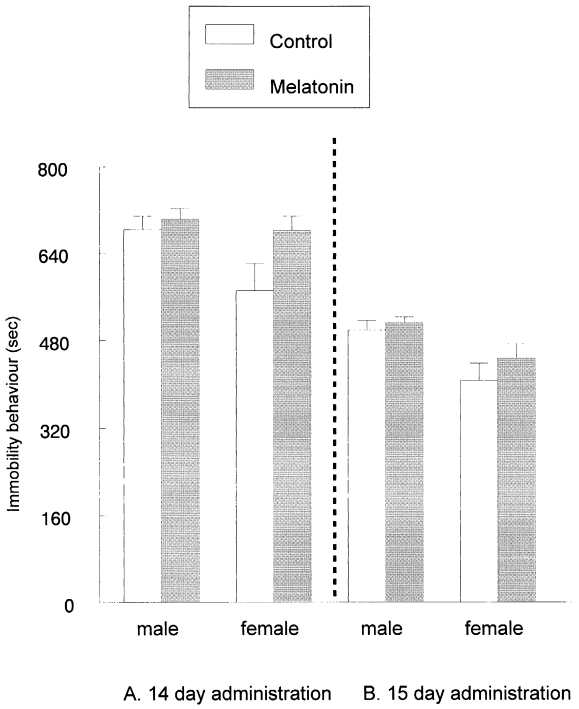

Fig. 3. Effects of melatonin administration on behavioural immobility (s) in male and female rats after 14 days of administration (panel A) and 15 days of administration (panel B) in the forced-swim test. Results demonstrate significant main effects of melatonin, $F(1,32) = 4.46$, $P = 0.04$, and sex, $F(1,32) = 4.59$, $P = 0.04$, after 14 days of administration, and a significant main effect of sex, $F(1,32) = 13.27$, $P = 0.001$, after 15 days of administration. Data represent means ± S.E.M.

3.2.2. Swimming behaviour

For swimming behaviour, statistical analyses revealed that although the interaction between melatonin and sex did not reach significance, $F(1,32) = 0.80$, $P = 0.38$, there were significant main effects of melatonin, $F(1,32) = 5.47$, $P = 0.03$, and sex, $F(1,32) = 6.09$, $P = 0.02$. As shown in Fig. 2, melatonin decreased swimming behaviour in all animals, and females showed higher levels of swimming than males.

3.2.3. Immobility behaviour

ANOVA tests failed to demonstrate a statistically significant interaction between melatonin and sex, $F(1,32) = 0.39$, $P = 0.54$, or a main effect of melatonin, $F(1,32) = 1.58$, $P = 0.22$, on behavioural immobility. There was a statistically significant main effect of sex, $F(1,32) = 13.27$, $P = 0.001$, such that males showed higher overall levels of behavioural immobility than females (Fig. 3).

Table 1
Effects of 17-day melatonin administration on crossing and rearing behaviours in the open-field test in male ($n = 20$) and female ($n = 16$) rats

	Crossing	Rearing
Males		
Control	38.5 ± 4.1	9.6 ± 1.7
Melatonin	50.0 ± 4.8	12.5 ± 1.2
Females		
Control	54.3 ± 4.9	15.6 ± 1.0
Melatonin	66.0 ± 5.8	17.4 ± 2.0

There were significant main effects of sex, $F(1,32) = 10.68$, $P = 0.003$, and melatonin, $F(1,32) = 5.73$, $P = 0.023$, on crossing behaviour, as well as a significant main effect of sex, $F(1,32) = 12.71$, $P = 0.001$, on rearing behaviour. Values represent means ± S.E.M.

3.3. Open-field test

3.3.1. Crossing behaviour

Statistical analyses on crossing behaviour revealed significant main effects of sex, $F(1,32) = 10.68$, $P = 0.003$, and melatonin, $F(1,32) = 5.73$, $P = 0.023$, but no interaction between sex and melatonin, $F(1,32) = 0.001$, $P = 0.98$. Table 1 shows that females performed significantly more crossings than males, and that melatonin-treated animals exhibited a slight increase in crossing behaviour, regardless of sex.

3.3.2. Rearing behaviour

The interaction between melatonin and sex, $F(1,32) = 0.14$, $P = 0.71$, as well as the main effect of melatonin, $F(1,32) = 2.31$, $P = 0.14$, failed to reach statistical significance for rearing behaviour. However, statistical analyses did reveal a main effect of sex, $F(1,32) = 12.71$, $P = 0.001$. Inspection of Table 1 indicates that females showed more rearing behaviour than males, regardless of treatment.

4. Discussion

Overall, the results demonstrated significant effects of melatonin administration on components of the forced-swim test, but that these effects were sex- and test-dependent. As Fig. 1 shows, melatonin increased struggling behaviour in males during their first exposure to the forced-swim test, but it had no effect on struggling in females during either presentation. Whereas melatonin increased immobility in females during test 1, this effect disappeared by test 2, with melatonin-treated and control females showing equivalent rates of behavioural immobility. Swimming behaviour was decreased in melatonin-treated animals during both exposures to the forced-swim test. In naïve rats, treatment with melatonin increased struggling in males, and not females, but increased immobility in females, and not males. However, after one exposure to the forced-swim test, the behavioural effects of melatonin on struggling and immobility disappeared, whereas the inhibitory effect of melatonin on swimming was retained. This is consistent with the findings of Taghzouti et al. (1999) who found that the behavioural response to the antidepressant fluoxetine depends on having had prior exposure to the forced-swim test. Taken together, these findings suggest that the effects of melatonin depend on whether animals have had prior exposure to the forced-swim test, and whether males or females are being studied.

Across tests and treatments, a sex difference emerges in the forced-swim test, such that females consistently show a higher level of active behaviours (struggling, swimming) than males. The present results are consistent with previous reports of sex differences in immobile behaviour in the forced-swim test (Alonso et al., 1991; Walker et al., 1995; Barros and Ferigolo, 1998). As rats are particularly sensitive to water depth in the forced-swim test (Abel, 1994), water levels were adjusted for each individual animal in the current study. Therefore, the observed sex difference in activity is not simply an artifact of fluid level, rather, it more likely represents a sex-specific response in the forced-swim test. Moreover, females showed higher activity levels in the open-field test, which is supported by previous findings (Valle and Gorzalka, 1980), and is apparent regardless of treatment condition. The current study utilized animals much older than those typically used, and the findings suggest that sex differences in activity levels which have previously been reported for younger animals remain with age.

The observation that melatonin increases struggling behaviour in naïve males provides support for an antidepressant-like effect of melatonin. The data are in agreement with the limited number of studies that have examined the effects of melatonin, or its analogues, in similar versions of the forced-swim test. While it appears that acute administration of a high dose of melatonin (30 mg/kg) does not alter the duration of immobility (Dubocovich et al., 1990), lower doses (10 mg/kg) reduce immobility times (Shaji and Kulkarni, 1998). The only existing study to employ a regimen of chronic administration demonstrated that the melatonin agonist, S20304, reduced immobility times in animals that were genetically predisposed to show high levels of immobility (Overstreet et al., 1998). In the present study, however, chronic melatonin at low doses had no effect on immobility in the male, and it increased immobility in females only during the initial exposure to the forced-swim test, after which its effect was not evident. A general inhibitory effect on motor activity does not seem likely since melatonin significantly increased line-crossing behaviour on the open-field test.

This is the first empirical study to examine the behavioural effects of melatonin on female rats. Consistent with the literature on sex-specific responses to antidepressants (Barros and Ferigolo, 1998) and to acute versus

repeated stressors (Kennett et al., 1986), the data suggest that chronic treatment with melatonin exerts antidepressive-like effects on males during a single exposure, but may do the opposite in females upon first exposure to the forced-swim test. Given that its immobility-enhancing effects were attenuated by the second test, it may be that if females were repeatedly exposed to the forced-swim test, antidepressive effects of melatonin would eventually become manifested as increased struggling and less immobility. In this way, melatonin may be acting somewhat as a pharmacological buffer against repeated stressors. This hypothesized method of action is not characteristic of antidepressants such as tricyclics and SSRIs, and may represent a lower grade of therapeutic benefit. Similarly, Kopp et al. (1999) discovered that chronic treatment with melatonin (1 mg/kg) in mice prevented the behavioural disturbances induced by a chronic mild stress regimen, in an animal model of human anhedonia. These authors suggested that melatonin might be involved in a homeostatic regulatory system which protects the animal from stress-induced injuries.

Divergent responses between males and females in a variety of other behavioural paradigms relevant to depression have been reported. For example, it has been shown that chronic exposure to a psychosocial stressor increases sexual activity of female rats, while decreasing that of males (Gorzalka et al., 1998). Similarly, Steenbergen et al. (1990) demonstrated that exposure to inescapable shock produced opposite effects in males and females on elevated plus-maze behaviour. One explanation for these behavioural discrepancies may be differences in hormonal responses between male and female subjects. For example, Galea et al. (1997) observed that male rats quickly habituated in their corticosterone response to a chronic stressor, whereas female rats showed little hormonal habituation, even after 21 days of restraint stress. Furthermore, it has been demonstrated that the anti-immobility effects of imipramine become more apparent during certain phases of the estrous cycle (Barros and Ferigolo, 1998). The current sex-dependent observations in the forced-swim test may be attributable to variations in hormonal responses, which may differentially interact with melatonin treatment.

It is currently unknown by what mechanism melatonin exerts its behavioural effects on components of the forced-swim test. Melatonin may be acting upon its own receptors given the observation that agonists highly selective for the melatonin receptor significantly decrease immobility times in the forced-swim test (Overstreet et al., 1998). Melatonin also readily interacts with other neurotransmitters and their receptors, including serotonergic, (Barros and Ferigolo, 1998; Eison et al., 1995; Gorzalka et al., 1999), noradrenergic (Datta and King, 1980), and GABAergic (Golombek et al., 1991) systems. Depending on melatonin's site of action, this may account for the observed selective decrease in swimming, yet increase in struggling behaviour, since agents which differentially interact with serotonergic and noradrenergic systems exert different effects on swimming and struggling (Detke et al., 1995). Nevertheless, it is changes in the struggling, and not swimming, behaviour that serve as a more valid predictor of antidepressant potential. On the other hand, it is possible that the effects of melatonin may be attributable to psychostimulant properties. Previous reports support a similarity between behavioural effects of melatonin and the psychostimulant amphetamine (Arushanian and Vodolazhskaia, 1997) and all dopaminergic substances that characteristically reduce behavioural immobility in the forced-swim test while concurrently enhancing motor activity in the open field (Borsini and Meli, 1988). However, given that only line crossing, but not rearing, was increased in melatonin-treated animals in the open field, a facilitatory effect of melatonin on locomotor activity cannot be stated conclusively. Therefore, antidepressant versus psychostimulant mechanisms of action for melatonin in the current study require additional study.

Given that the forced-swim test is considered a reliable animal model of depression, and that drugs which alter forced-swim test components have antidepressant potential, the current results suggest that melatonin may possess antidepressive properties. This would support clinical studies investigating melatonin correlates with mood and depressive symptomatology (Beck-Friis et al., 1984; Frazer et al., 1986; Miles and Philbrick, 1988) and the suggestion that these symptoms may be ameliorated by melatonin supplementation. Apart from correlational studies indicating low, or abnormal, levels of melatonin in individuals suffering from depression, empirical studies investigating the effects of melatonin in this domain are absent. Results from the current study suggest that melatonin may have implications for the treatment of behavioural despair in depression, but rigorous empirical research is clearly needed if melatonin is to be considered as an adjunct to treatment.

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References

Abel, E.L., 1994. Behavioral and physiological effects of different water depths in the forced swim test. Physiol. Behav. 56, 411–414.

Abel, E.L., Hannigan, J.H., 1992. Effects of chronic forced-swimming and exposure to alarm substance: physiological and behavioral consequences. Physiol. Behav. 52, 781–785.

Alonso, S.J., Castellano, M.A., Afonso, D., Rodriguez, M., 1991. Sex differences in behavioral despair: relationships between behavioral despair and open field activity. Physiol. Behav. 49, 69–72.

Armario, A., Gavalda, A., Marti, O., 1988. Forced swimming test in rats: effect of desipramine administration and the period of exposure to the test on struggling behavior, swimming, immobility and defecation rate. Eur. J. Pharmacol. 158, 207–212.

Arushanian, E.B., Vodolazhskaia, M.G., 1997. Influence of melatonin and amphetamine on formation of time-dependent conditional reflex in rats. Eksp. Klin. Farmakol. 60, 9–11.

Barbhaiya, R.H., Buch, A.B., Greene, D.S., 1996. A study of the effect of age and gender on the pharmacokinetics of nefazodone after single and multiple doses. J. Clin. Psychopharmacol. 16, 19–25.

Barros, H.M.T., Ferigolo, M., 1998. Ethopharmacology of imipramine in the forced-swimming test: gender differences. Neurosci. Biobehav. Rev. 23, 279–286.

Beck-Friis, J., von Rosen, D., Kjellman, B.F., Ljunggren, J.G., Wetterberg, L., 1984. Melatonin in relation to body measures, sex, age, season and the use of drugs in patients with major affective disorders and healthy subjects. Psychoneuroendocrinology 9, 261–277.

Beck-Friis, J., Kjellman, B.F., Aperia, B., Unden, F., von Rosen, D., Ljunggren, J.G., Wetterberg, L., 1985. Serum melatonin in relation to clinical variables in patients with major depressive disorder and a hypothesis of a low melatonin syndrome. Acta Psychiatr. Scand. 71, 319–330.

Borsini, F., Meli, A., 1988. Is the forced swimming test a suitable model for revealing antidepressant activity? Psychopharmacology 94, 147–160.

Bracke, P., 1998. Sex differences in the course of depression: evidence from a longitudinal study of a representative sample of the Belgian population. Soc. Psychiatry Psychiatr. Epidemiol. 33, 420–429.

Datta, P.C., King, M.G., 1980. Melatonin: effects on brain and behavior. Neurosci. Behav. Rev. 4, 451–458.

Detke, M.J., Rickels, M., Lucki, I., 1995. Active behaviors in the rat forced swimming test differentially produced by serotonergic and noradrenergic antidepressants. Psychopharmacology 121, 66–72.

Dubocovich, M.L., Mogilnicka, E., Areso, P.M., 1990. Antidepressant-like activity of the melatonin receptor antagonist, luzindole (N-0774), in the mouse behavioral despair test. Eur. J. Pharmacol. 182, 313–325.

Eison, A.S., Freeman, R.P., Guss, V.B., Mullins, U.L., Wright, R.N., 1995. Melatonin agonists modulate 5-HT_{2A} receptor-mediated neurotransmission: behavioral and biochemical studies in the rat. J. Pharmacol. Exp. Ther. 273, 304–308.

Filip, V., Alda, M., David, I., Topinka, J., Kristofikova, Z., Dvorakova, J., Sztaniszlav, D., Olajos, S., Albrecht, V., 1989. Neuroendocrine response to clomipramine and desipramine — the evidence of partial determination by heredity and sex. Neuropsychobiology 21, 111–116.

Fink, G., Sumner, B.E., Rosie, R., Grace, O., Quinn, J.P., 1996. Estrogen control of central neurotransmission: effect on mood, mental state, and memory. Cell. Mol. Neurobiol. 16, 325–344.

Frazer, A., Brown, R., Kocsis, J., Caroff, S., Amsterdam, J., Winokur, A., Sweeney, J., Stokes, P., 1986. Patterns of melatonin rhythms in depression. J. Neural Transm., Suppl. 21, 269–290.

Galea, L.A., McEwen, B.S., Tanapat, P., Deak, T., Spencer, R.L., Dhabhar, F.S., 1997. Sex differences in dendritic atrophy of CA3 pyramidal neurons in response to chronic restraint stress. Neuroscience 81, 689–697.

Godfroid, I.O., 1999. Sex differences relating to psychiatric treatment. Can. J. Psychiatry 44, 362–367.

Golombek, D.A., Escolar, E., Cardinali, D.P., 1991. Melatonin-induced depression of locomotor activity in hamsters: time dependency and inhibition by the central type benzodiazepine antagonist RO15-1788. Physiol. Behav. 49, 1091–1098.

Gorzalka, B.B., Hanson, L.A., Brotto, L.A., 1998. Chronic stress effects on sexual behavior in male and female rats: mediation by 5-HT_{2A} receptors. Pharmacol. Biochem. Behav. 61, 405–412.

Gorzalka, B.B., Brotto, L.A., Hong, J.J., 1999. Corticosterone regulation of 5-HT_{2A} receptor-mediated behaviors: attenuation by melatonin. Physiol. Behav. 67, 439–442.

Halbreich, U., 1997. Hormonal interventions with psychopharmacological potential: an overview. Psychopharmacol. Bull. 33, 281–286.

Hansen, H.H., Sanchez, C., Meier, E., 1997. Neonatal administration of the selective serotonin reuptake inhibitor Lu 10-134-C increases forced swimming-induced immobility in adult rats: a putative animal model of depression. J. Pharmacol. Exp. Ther. 283, 1333–1341.

Kennett, G.A., Chaouloff, F., Marcou, M., Curzon, G., 1986. Female rats are more vulnerable than males in an animal model of depression: the possible role of serotonin. Brain Res. 382, 416–421.

Kopp, C., Vogel, E., Rettori, M.C., Delagrange, P., Misslin, R., 1999. The effects of melatonin on the behavioural disturbances induced by chronic mild stress in C_3H/He mice. Behav. Pharmacol. 10, 73–83.

Lucki, I., 1997. The forced swimming test as a model for core and component behavioral effects of antidepressant drugs. Behav. Pharmacol. 8, 523–532.

Marazziti, D., Rossi, A., Palego, L., Barsanti, A., Carrai, M., Giannaccini, G., Serra, P., Lucacchini, A., Cassano, G.B., 1998. Effect of aging and sex on the [3H]-paroxetine binding to human platelets. J. Affective Disord. 50, 11–15.

Miles, A., Philbrick, D.R.S., 1988. Melatonin and psychiatry. Biol. Psychiatry 23, 405–425.

Oaknin-Bendahan, S., Anis, Y., Nir, I., Zisapel, N., 1995. Effects of long-term administration of melatonin and a putative antagonist on the ageing rat. Neuroreport 6, 785–788.

Overstreet, D.H., Pucilowski, O., Retton, M.C., Delagrange, P., Guardiola-Lemaitre, B., 1998. Effects of melatonin receptor ligands on swim test immobility. Neuroreport 9, 249–253.

Porsolt, R.D., Anton, G., Blavet, N., Jalfre, M., 1978. Behavioral despair in rats: a new model sensitive to antidepressant treatments. Eur. J. Pharmacol. 47, 379–391.

Shaji, A.V., Kulkarni, S.K., 1998. Central nervous system depressant activities of melatonin in rats and mice. Ind. J. Exp. Biol. 36, 257–263.

Steenbergen, H.L., Heinsbroek, R.P., Van Hest, A., Van de Poll, N.E., 1990. Sex-dependent effects of inescapable shock administration on shuttlebox-escape performance and elevated plus-maze behavior. Physiol. Behav. 48, 571–576.

Taghzouti, K., Lamarque, S., Kharouby, M., Simon, H., 1999. Interindividual differences in active and passive behaviors in the forced-swimming test: implications for animal models of psychopathology. Biol. Psychiatry 45, 750–758.

Valle, F.P., Gorzalka, B.B., 1980. Open-field sex differences prior to puberty in rats. Bull. Psychon. Soc. 16, 429–431.

Walker, C.D., Trottier, G., Rochford, J., Lavallee, D., 1995. Dissociation between behavioral and hormonal responses to the forced swim stress in lactating rats. J. Neuroendocrinol. 7, 615–622.