

Prenatal desvenlafaxine induced behavioural alterations in Swiss albino mice

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KEY WORDS

Aluminum
Open field test
Elevated plus maze test
Norepinephrine
Serotonin

ABSTRACT

Background: Desvenlafaxine is used as an antidepressant and acts by inhibiting reuptake of serotonin and nor-adrenaline. **Purpose:** The safety profile of desvenlafaxine has not yet been established during pregnancy, so we planned this study to see the behavioral changes in pups of mice who received desvenlafaxine during gestational period. **Methods:** Swiss albino mice were used for the present study. The treated group was given desvenlafaxine orally in the dose of 80 mg/kg from 1st to 6th day of gestation and other group was given tap water by same route. **Results:** Desvenlafaxine treated mice in group 2, i.e. for the gestation period 1–6 showed increased activity and decrease anxiety in open field and elevated plus maze test as compared to control. However, after chronic exposure for the duration of 18 days the offspring showed increased anxiety and fearfulness as compared to controls. **Conclusion:** Above findings suggest that desvenlafaxine have a deleterious effect on brain development, thus resulting in abnormal anxiety states, possibly through altering uptake of serotonin and nor-epinephrine.

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doi : 10.5214/ans.0972.7531.210106

Introduction

Desvenlafaxine is a novel atypical antidepressant referred as 'serotonin and nor-adrenaline reuptake inhibitor' (SNRI) as it inhibits serotonin and nor-epinephrine reuptake in the presynaptic cleft.¹ But in contrast to older tricyclic antidepressants it does not interact with cholinergic, adrenergic or histaminergic receptors or have any sedative property.

It is one of the examples of chiral drugs composed of enantiomers which may differ in their pharmacokinetic and pharmacodynamic properties.²⁻⁴ It inhibits serotonin more than nor-adrenaline reuptake and slightly inhibits dopamine reuptake.⁵ Therefore it is used for the treatment of major depressive disorders, panic disorders, generalized anxiety disorders, and social anxiety disorders.⁶ It is used for the treatment of major depressive disorders, panic disorders, generalized anxiety disorders, and social anxiety disorders.⁶ Looking at very few and non-conclusive reports about the teratogenicity of desvenlafaxine and histopathological changes induced by it, the present project has been undertaken. The literature suggests that at the peak of cell differentiation for serotonergic neurons occurs on gestational days 15–16 and by gestational day 19 the distribution of serotonin neurons resembles that found in the adult. Therefore these drugs both inhibitors of serotonin reuptake might interfere with the developing serotonergic system at this critical time.^{7,8} Therefore for the above reason we have used this critical duration of drug exposure in our experiments.

Methods

The present study was conducted in the Department of Anatomy, Institute of Medical Sciences, Banaras Hindu University, Varanasi. Adult female swiss albino mice, weighing 20–25 gm, (average age of 80–100 days), were used after approval of institutional ethical committee. The animal room was maintained at an ambient temperature of 25 ± 2°C and 50–60% relative humidity with 12 hr light dark cycle each.

The animals were housed in polypropylene cages with rice husk bedding. They were fed on pelleted diet obtained from local Pashu Ahar kendra and tap water *ad libitum*. The female mice in their pre-estrous phase were transferred in the evening to the cages containing male mice in the ratio of 2:1. The presence of vaginal plug on the following morning indicated pregnancy and was designated as day zero (0) of gestation.

All pregnant mice were divided into following groups.

- Group-1 Treated with equivalent amount of tap water during same days of gestation.
- Group-2 Treated with desvenlafaxine from GD₁ to GD₆.
- Group-3 Treated with desvenlafaxine from GD₁ to GD₁₈.

Desvenlafaxine was obtained from Innova division of IPCA Industries (Mumbai). Each tablet contained 50 mg of the drug. Before treatment, one tablet was dissolved in 5 ml of tap water. Animals from both control and various treated groups were allowed to deliver. These delivered pups were subjected to various behavioral tests at the age of 8 weeks approximately.

Behavioral tests

1) Open field test

An open field apparatus made of plywood measuring 60.96 × 60.96 × 60.96 cms was used to test the open field exploratory behavior of mice. The floor of the apparatus was divided into 16 evenly spaced squares surrounded by opaque high walls of 60.96 cms. The entire apparatus was painted black except for the 6 mm wide white lines that divided the floor into 16 squares. The open field was illuminated by 100 w bulb focusing into field from a height of about 100 cms from the floor. The entire room, except the apparatus, was kept dark during experiment. Animals were kept in apparatus for 5 minutes to observe for following parameters:

- 1) Ambulation – The numbers of squares crossed by the mice.
- 2) Rearing – The number of times mice stood on its hind limbs.

- 3) Self grooming – The number of times the mice did action of licking, grooming, scratching.
- 4) Freezing time – Duration for which mice remains still without any movement.
- 5) Fecal pellets – The number of fecal pellets excreted by each individual mice during the trial of 5 minutes. Before each trial, the floor and the walls were cleaned with cotton soaked in 70% alcohol. This experiment was done to test the anxiety levels in mice under the effect of drug desvenlafaxine.

II) *Elevated plus maze test*

This apparatus consists of two opposite arms of 50 × 10 cms, connected with a central square (10 × 10 cm), giving it a shape of plus sign. One arm painted white was the open arm whereas other arm was enclosed with 40 cms high walls and was painted black together with the walls. The maze was kept in a dimly lit room and was elevated 50 cms above the floor. The experimental animals were placed individually in the centre of maze facing an enclosed arm and the time spent on the open and closed arms were recorded during the next 5 min for each mice. An arm entry was recorded when all four paws of the rat entered the arm. Like the open field apparatus, the floor and walls of open and enclosed arm were cleaned with 70% alcohol before each trial. This model was also used to study anxiety in the treated and control mice.

Statistical analysis

Data was analyzed by applying one way ANOVA followed by post-hoc tukey test using software SPSS. All data was expressed in mean ± SD.

Results

Open field test

This test was performed to test anxiety in the offspring of all the experimental mice groups after the age of 8 wks. The grown up pups of the control and treated mice were subjected to this test.

In group 2 the offspring showed significant increase in ambulation and rearing. They also showed increase in the frequency of self-grooming which was observed to be highly significant in comparison to group 1 (control) and group 3. These fetuses showed significantly less reduced immobility period. These findings support the effectiveness of desvenlafaxine as an antidepressant which by decreasing anxiety helped to enhance activity of the young mice. While comparing fetuses of group 1 (control) and group 2 the frequency of rearing and self-grooming was found to be increased significantly but the other parameters (ambulation, fecal pellets) did not show any significant changes. There was decrease in the parameters of ambulation rearing and self-grooming in group 3 which was significant in comparison to group 2 and non-significant in comparison to group 1. The parameters of immobility period and fecal pellets were significantly increased in comparison to group 2, but they were again not-significantly increased in comparison to group 1. (Table-1)

Elevated plus maze test

This is an important test for observing behavioral abnormalities like anxiety and depression in the experimental animals. During this offspring of group 2 mice were found to spend significantly more time in open arms as compared to offspring of group 1 (control) and group 3. Of all the groups offspring of group 3 spent least time in open arms. On the other hand group 3 offspring were found to spend significantly more time in enclosed arms as compared to group 1 and group 2. On comparing the number of entries in the open arms and enclosed arms in the offspring of mice of various groups, offspring of group 2 fetuses showed significantly more entries in the open arms as compared to group 1 and group 3. But their entries in closed arms were significantly reduced in comparison to other two groups. Offspring of group 3 showed significantly more entries in closed arms. Thus, from the above findings, we can conclude that offspring of mice of group 2 show anti-depressant effect of desvenlafaxine by reducing anxiety enhanced its activity.

Table 1: Effect of prenatal Desvenlafaxine exposure on open field exploratory behaviour in mice offspring.

| Mice | Ambulation | Rearing | Self Grooming | Immobility Period | Faecal Pellets |
|---------|----------------|--------------|---------------|-------------------|----------------|
| Group 1 | 96.17 ± 26.46 | 11.33 ± 7.94 | 2.67 ± 2.33 | 53.00 ± 23.23 | 1.83 ± 0.98 |
| Group 2 | 140.17 ± 44.60 | 42.17 ± 5.56 | 10.00 ± 3.74 | 23.50 ± 15.68 | 0.83 ± 0.40 |
| Group 3 | 81.67 ± 14.02 | 7.50 ± 4.76 | 2.33 ± 1.03 | 65.17 ± 38.03 | 2.50 ± 1.04 |
| p-value | 0.014* | <0.001*** | <0.001*** | 0.049* | 0.015* |

*p<0.05; **p<0.01; ***p<0.001.

Table 2: Group comparison.

| Parameters | Group 1 vs 2 | Group 1 vs 3 | Group 2 vs 3 |
|-------------------|--------------|--------------|--------------|
| Ambulation | 0.065 | 0.703 | 0.014* |
| Rearing | <0.001*** | 0.550 | <0.001*** |
| Self Grooming | 0.001** | 0.974 | 0.001** |
| Immobility Period | 0.181 | 0.725 | 0.046* |
| Faecal Pellets | 0.145 | 0.397 | 0.012* |

*p<0.05; **p<0.01; ***p<0.001.

Table 3: Effect of prenatal Desvenlafaxine exposure on elevated plus maze test behaviour in mice offspring.

| Mice | Close arm duration | Open arm duration | Entry close arm | Entry open arm |
|---------|--------------------|-------------------|-----------------|----------------|
| Group 1 | 3.14 ± 0.47 | 2.02 ± 0.15 | 10.67 ± 3.61 | 5.33 ± 2.25 |
| Group 2 | 1.90 ± 1.07 | 3.10 ± 1.08 | 5.00 ± 1.54 | 15.67 ± 2.58 |
| Group 3 | 4.27 ± 0.22 | 0.72 ± 0.22 | 11.50 ± 3.20 | 3.17 ± 2.13 |
| p-value | <0.001*** | <0.001*** | 0.003** | <0.001*** |

*p<0.05; **p<0.01; ***p<0.001.

Table 4: Group comparison.

| Parameters | Group 1 vs 2 | Group 1 vs 3 | Group 2 vs 3 |
|-----------------|--------------|--------------|--------------|
| Close arm | 0.019* | 0.031* | <0.001*** |
| Open arm | 0.028* | 0.009** | <0.001*** |
| Entry close arm | 0.012* | 0.876 | 0.004** |
| Entry open arm | <0.001*** | 0.272 | <0.001*** |

*p<0.05; **p<0.01; ***p<0.001.

Discussion

In various behavioral tests, the drug caused alterations in the treated groups as compared to controls. During these experiments we noted that desvenlafaxine when given for short duration (1–6 days of gestation) caused increased ambulation, rearing and self-grooming in open field test as compared to controls. These findings corroborate with those of Fang et al⁹. This can be explained on the basis of the fact that this drug by selectively inhibiting serotonin and nor-epinephrine reuptake pumps tends to increase their concentration in the synaptic cleft which in turn elevates rat cortical and hypothalamic levels of both 5-HT and NE. The increased synaptic concentration of neurotransmitters is thought to induce anxiolytic effect and thus increase in activity. The extracellular 5-HT results directly from blockade of 5-HT transporter. It also increases synaptic level of serotonin leading to an increased activation of a multitude of specific post-synaptic 5-HT receptors. The increase in the various neurotransmitters in diencephalon and frontal cortex explains alteration in mood and cognitive deficit shown by the experimental mice.

Desvenlafaxine also demonstrates good brain-to-plasma ratio suggesting utility in CNS, peripheral nervous system and other changes associated with neurotransmitters. But in contrast to above reports chronic dosing of desvenlafaxine reduced the activities of offspring of treated mice in open field, elevated plus maze test. This may be explained by the fact that reduced uptake of serotonin and norepinephrine might render these neurotransmitters to their metabolic degradation which leads to their acute shortage in the presynaptic terminal buttons. Accumulation of the unchanged desvenlafaxine, desvenlafaxine-o-glucuronide conjugates and unconjugated N-desmethyl metabolite produced by various oxidative and conjugation processes may be the culprit behind the severe neuronal degeneration

caused by the drug. Excessive reduction of neurotransmitters in terminal buttons and toxic metabolites induced neuronal degeneration may be the factor responsible for behavioral impairment in offspring in the chronic treated mice dams.

The article complies with International Committee of Medical Journal Editor's uniform requirements for manuscript.

Conflict of Interests: None: Source of funding: None

Received Date : 31 August 2013; Revised Date : 29 November 2013;
Accepted Date : 28 January 2014

References

- Deecker DC, Beyer CE, Johnston G, et al. Desvenlafaxine succinate; a new serotonin and norepinephrine reuptake inhibitor. *J Pharmacol Exp Ther* 2006; 318(2): 657–65.
- Baker GB, Prior TI. Stereochemistry and drug efficacy and development: relevance of chirality to antidepressant and antipsychotic drugs. *Ann Med* 2002; 34: 537–543.
- Baumann P, Zullin DF, Eap CB. Enantiomers potential in psychopharmacology—a critical analysis with special emphasis on the antidepressant escitalopram. *Eur Neuropsychopharmacol* 2002; 12: 433–444.
- Howland RH. Clinical implication of chirality and stereochemistry in psychopharmacology. *J Psychosoc Nurs Ment Health Serv* 2009; 47: 17–21.
- Muth EA, Haskins JT, Moyer JA, et al. Antidepressant biochemical profile of the novel bicyclic compound Wy-45, 030, an ethyl cyclohexanol derivative. *Biochem Pharmacol* 1986; 35: 4493–4497.
- Stahl SM, Grady MM, Moret C, et al. SNRIs: their Pharmacology, clinical efficacy and tolerability in comparison with other classes of antidepressants. *CNS Spectr* 2005; 10: 732–747.
- Da-Silva VA, Moraes-Santos AR, Carvalho MS, et al. Prenatal and post-natal depression in low income Brazilian women. *Braz J Med Biol Res* 1998; 31: 799–804.
- Druse MJ, Kuo A, Tajuddin N. Effects of in utero ethanol exposure on the developing serotonergic system. *Alcohol Clin Exp Res* 1991; 15: 678–684.
- Fang S, Yan B, Wang D, et al. Chronic effects of venlafaxine on synaptophysin and neuronal cell adhesion molecule in the hippocampus of cerebral ischemic mice. *Biochem Cell Biol* 2010; 88(4): 655–663.