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# Effect of Sub-Acute Administration of Celecoxib on Anxiolytic Activity of Fluoxetine in Albino Mice

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## ABSTRACT

Our objective was to evaluate the effect of sub-acute administration of celecoxib on the anxiolytic activity of fluoxetine in albino mice. After clearance from Institutional Animal Ethics Committee, 24 healthy albino mice (20 - 30g) of either sex were divided into four groups of six mice each and administered Distilled water 1ml/kg (control), Alprazolam 5mg/kg (standard), Fluoxetine 5mg/kg, and Fluoxetine 5mg/kg + Celecoxib 5mg/kg respectively by intraperitoneal route for 21 days. Anxiolytic effect was evaluated by Elevated Plus Maze (EPM) and Hole Board (HB) test in all groups at baseline (Day 0) and Day 21 after drug administration. Open arm exploration time in EPM and number of times of nose pokings in HB were measured in all groups and the results were expressed as Mean ± SD. Statistical analysis was done by one-way ANOVA followed by Unpaired 't' test with $P<0.05$ as the level of significance (95% confidence limits). In the fluoxetine + celecoxib group, open arm exploration time in EPM and number of nose pokings in HB were significantly decreased, compared to normal control. When we co-administered fluoxetine and celecoxib daily for a period of 21 days, we observed reversal of anxiolytic activity of fluoxetine in EPM, but anxiolytic effect was observed in HB test, although it was not significantly different from when fluoxetine alone was administered.

**Keywords:** Anxiolytic, Celecoxib, Elevated Plus maze, Fluoxetine, Hole Board, Subacute

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## INTRODUCTION

Selective Serotonin reuptake inhibitors (SSRIs) such as fluoxetine are widely used in anxiety states, especially when co-morbid depression also exists. It is also used for anxiety-related disorders such as obsessive-compulsive disorder, panic, social phobia and bulimia nervosa.[1] In humans, the anxiolytic effects of SSRIs emerge only after chronic treatment.[2] It is known that anxiety is one of the adverse effects of celecoxib administration.[3] In clinical practice, sometimes patients on fluoxetine may be required to take celecoxib for painful or inflammatory conditions. In such cases, there is a possibility of a drug interaction between them and this may lead to a decrease in the anxiolytic activity of fluoxetine.

We had earlier evaluated the effect of acute administration of celecoxib and fluoxetine in rodent models of anxiety.[4] Using the elevated plus maze and hole board tests in healthy adult albino mice of either sex, we observed both anxiogenic as well as anxiolytic activity of the combination in acute study. Hence, we wanted to observe whether there is any change in the anxiolytic effect of fluoxetine by sub-acute concurrent administration of celecoxib in rodent models of anxiety.

## MATERIALS AND METHODS

After getting clearance from the Institutional Animal Ethics Committee, we obtained 24 healthy adult albino mice of either sex (20-40g) from the Central Animal House of our institute and cared for them, as per the recommendations of the Committee for the purpose of control and supervision of experiments on animals (CPCSEA).[5]

We acquired the following drugs – fluoxetine, ibuprofen and celecoxib from Cadila Pharmaceuticals Ltd., J&K, Abbott India Ltd., Goa and Zydus Cadila, Zydus Healthcare, Sikkim respectively. We selected low doses of fluoxetine and celecoxib and standard dose of alprazolam from previous studies. [6-8] We suspended the drugs in Distilled water (D/W) (1ml/kg) and administered Fluoxetine 5mg/kg, Alprazolam 5mg/kg (standard anxiolytic) and Celecoxib 5mg/kg intra-peritoneally.

### Grouping and treatment scheduling:

We divided healthy albino mice of either sex (20-40g) into four arms containing six mice each for testing antidepressant activity (n=24). The treatment schedule was as follows:

Group A: D/W (1ml/kg)
Group B: Alprazolam (5mg/kg)
Group C: Fluoxetine (5mg/kg)
Group D: Fluoxetine (5mg/kg) + Celecoxib (5mg/kg) administered separately

### Experimental design:

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We performed the test for anxiolytic effect by Elevated plus maze (EPM) and Hole Board (HB). After taking baseline values of tests with EPM and HB on Day 0, the vehicle and the drugs were administered orally 30 minutes before subjecting them to EPM and HB tests on Day 21.

**Elevated Plus Maze (EPM):**

We performed the elevated plus maze test, similarly as described in our earlier study.[4] We recorded the Open arm exploratory time and compared the values of treated groups with controls.

**Hole board test:**

Similarly, we performed the Hole Board test to evaluate the exploratory behaviour of mice as we had done in our acute study.[4] Thirty minutes after intraperitoneal administration of the test/standard compound, the first animal was placed on the hole-board. We observed the number of times the mouse poked its nose into the hole during the 5 minute testing session.

The number of counts for nose-poking of treated animals was compared with those of control.

**Statistical analysis:**

We performed Statistical analysis, using SPSS statistical software Version 16.0. Duration of immobility was expressed as Mean ± SD. For demonstration of anxiolytic activity, we used one way ANOVA, followed by Unpaired 't' test for analysing the difference between groups (if any), with $P < 0.05$ as level of significance with 95% confidence interval.

## RESULTS

The results of EPM and HB expressed as Mean ± SD are shown in Table 1 and results of one way ANOVA are shown in Table 2. Inter-group comparisons on day of experiment (Day 1) using Unpaired 't' test are shown in Tables 3 and 4.

**Table 1: Cumulative duration of time (seconds) spent in the open arm in Elevated Plus Maze (EPM) and Number of times of nose poking in Hole Board (HB), expressed as Mean ± SD**

| GROUP | EPM (D0) | EPM (D21) | HB (D0) | HB (D21) |
|---|---|---|---|---|
| A | 21.17 ± 11.89 | 15.83 ± 3.19 | 34.83 ± 7.00 | 31.33 ± 4.23 |
| B | 16.50 ± 5.96 | 40.67 ± 8.66 | 40.33 ± 13.05 | 4.67 ± 1.75 |
| C | 17.50 ± 6.97 | 34.50 ± 11.02 | 31.83 ± 5.38 | 22.00 ± 5.80 |
| D | 17.50 ± 8.26 | 6.67 ± 3.01 | 50.00 ± 10.28 | 31.67 ± 10.41 |

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**Table 2: Results of One Way ANOVA**

| Test | Day 0 (*P* value) | Day 21 (*P* value) |
|---|---|---|
| EPM | 0.78 | 0.00** |
| HB | 0.07 | 0.00** |

*P* value < 0.05, not significant on Day 0, highly significant (**) on Day 1

**Table 3: Results of Unpaired 't' test between groups on day of experiment (Day21) for EPM**

| t-test | | A | B | C | D | E |
|---|---|---|---|---|---|---|
| A | t= | | | | | |
| | p= | | | | | |
| B | t= | 6.59 | | | | |
| | p= | 0.00** | | | | |
| C | t= | 3.98 | 1.08 | | | |
| | p= | 0.00** | 0.31 | | | |
| D | t= | 5.12 | 9.08 | 5.97 | | |
| | p= | 0.00* | 0.00** | 0.00** | | |

*indicates significant and ** indicates highly significant difference between groups

**Table 4: Results of Unpaired 't' test between groups on day of experiment (Day21) for HB**

| t-test | | A | B | C | D | E |
|---|---|---|---|---|---|---|
| A | t= | | | | | |
| | p= | | | | | |
| B | t= | 14.28 | | | | |
| | p= | 0.00** | | | | |
| C | t= | 3.19 | 7.01 | | | |
| | p= | 0.00** | 0.00** | | | |
| D | t= | 0.07 | 6.27 | 1.99 | | |
| | p= | 0.94 | 0.00** | 0.07 | | |

*indicates significant and ** indicates highly significant difference between groups

## DISCUSSION

We had earlier observed both anxiogenic as well as anxiolytic activity of the combination of fluoxetine and celecoxib in an acute study, using elevated plus maze and hole board tests.[4] Hence, we conducted a study of longer duration to observe whether the daily concurrent administration of celecoxib and fluoxetine produces anxiogenic or anxiolytic activity. Our objective was to evaluate the effect of sub-acute administration of celecoxib on the anxiolytic activity of fluoxetine in albino mice over a period of 21 days.

We found that in the alprazolam and fluoxetine groups, there was a mean increase in the time spent in the open arm in EPM after 21 days of administration of drugs. Thus, these groups demonstrate anxiolytic activity. However, in the combination group of fluoxetine with celecoxib, we observed a mean decrease in the time spent in the open arm, demonstrating a decrease in anxiolytic activity. But, in the HB test, mean number of nose pokings decreased in all groups compared to the control, demonstrating anxiolytic activity. Therefore, we observed significant anxiolytic activity in the combination group of fluoxetine and celecoxib.

Results of one way ANOVA showed that there was no significant difference between the performances of the groups in EPM and HB test on Day 0. Thus, the groups were comparable at baseline before drug administration. But, we observed a highly significant difference between the performances of the animals in EPM and HB test on Day 21 (after drug administration) compared to Day 0.

On comparing the performance of the drug treated groups in EPM on Day 21, alprazolam, fluoxetine and the combination group of fluoxetine with celecoxib showed significant anxiolytic activity compared to the control group. There was no significant difference in anxiolytic activity between the fluoxetine and alprazolam groups. However, there was a significant decrease in anxiolytic activity in the combination group of fluoxetine and celecoxib compared to the alprazolam and to the fluoxetine group. Sub-acute administration of celecoxib with fluoxetine over 21 days probably reversed the significant anxiolytic activity which was seen after administration of fluoxetine alone.

On comparing the performance of the drug treated groups in HB on Day 21, the alprazolam and fluoxetine groups showed significant anxiolytic activity compared to the control. Alprazolam group demonstrated significantly greater anxiolytic activity compared to fluoxetine and the combination group of fluoxetine with celecoxib. However, the combination group of fluoxetine and celecoxib demonstrated no significant anxiolytic activity compared to the fluoxetine group. Therefore, sub-acute administration of celecoxib with fluoxetine over 21 days had no significant effect on the anxiolytic activity which was seen after administration of fluoxetine alone.

Thus, in our study, we observed consistent anxiolytic activity of fluoxetine when administered alone in adult albino mice of either sex for 21 days. But, when we evaluated the effect of daily administration of celecoxib with fluoxetine, we observed reversal of the anxiolytic activity of fluoxetine in EPM and anxiolytic activity in HB which was not significantly different from the anxiolytic activity of fluoxetine alone.

The data obtained in animal studies using SSRIs are contradictory.[9-11] Specifically, the effect of fluoxetine on animal models of anxiety is controversial, and studies on its chronic effects are scarce. Chronic administration of fluoxetine suggests an anxiolytic effect of the drug.[12,13] But, one study has reported the absence of either an anxiogenic or an anxiolytic effect of the drug when administered chronically.[14] Previous literature on the sub-acute or

chronic effect of NSAIDs on anxiolytic activity of fluoxetine or any other SSRI in animal models of anxiety is lacking, so we were not able to compare our study with similar studies.

## CONCLUSION

Our objective was to evaluate the effect of sub-acute administration of celecoxib on the anxiolytic activity of fluoxetine, using elevated plus maze and hole board tests in albino mice. In a previous study, we had observed both anxiogenic and anxiolytic activity of this combination when administered acutely. In this study, we observed that fluoxetine consistently produced anxiolytic effect, but concurrent administration of celecoxib decreased the anxiolytic activity of fluoxetine in elevated plus maze. The combination of fluoxetine and celecoxib produced anxiolytic effect which was not significantly different from that produced by fluoxetine alone in the hole board test. Hence, no consistent anxiolytic effect of fuoxetine or its reversal can be inferred when the combination is administered sub-acutely. So, it is not possible to conclude that there is a drug interaction between fluoxetine and celecoxib which decreases the anxiolytic activity of fluoxetine. Therefore, animal studies of longer duration (chronic use), with a wider battery of screening tests are required to confirm whether this drug interaction exists.

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## REFERENCES

[1] Tripathi KD. Antidepressant drugs. Essentials of Medical Pharmacology, 6th edition, Jaypee Brothers Medical Publishers, New Delhi, 2008, p. 446
[2] Celebrex side effects. <Available at http://www.drugs.com/sfx/celebrex-side-effects.html. Accessed Jul 9, 2013>
[3] Nutt DJ, Forshall S, Bell C, Rich A, Sandford J, Nash J, *et al*. Eur Neuropsychopharmacol 1999; 9(Suppl 3):S81–S86.
[4] Pandian J, Lahon K, Somu L. Res J Pharm Biol Chem Sci 2013;4(2):1259-66
[5] Committee for the purpose of control and supervision of experiments on animals. CPCSEA guidelines for laboratory animal facility. Indian J Pharmacol 2003;35:257-74
[6] Elhwuegi AS, Hassan KM. Libyan J Med 2012; 7:17251.
[7] Wang PH, Horng HC, Chen YJ, Hsieh SL, Chao HT, Yuan CC. J Chin Med Assoc 2007; 70(6):245-8
[8] Bukhari IA, Dar A, Khan RA. Pak J Pharm Sci 2004;17(2):13-9
[9] Handley SL and McBlane JW. Psychopharmacology 1993;112:13-20
[10] Griebel G. Pharmacology and Therapeutics 1995;65: 319-95.
[11] Sánchez C and Meier E. Psychopharmacology 1997;129:197-205
[12] Bodnoff SR, Suranyi-Cadotte B, Quirion R and Meaney MJ. Psychopharmacology 1989;97:277-9

ISSN: 0975-8585

[13] Griebel G, Blanchard CD, Agnes RS and Blanchard RJ. Psychopharmacology 1995;120:57-66

[14] Tsuchiya K, Inoue T and Koyama T. Pharmacology, Biochemistry and Behavior 1996;54:687-691.