Acta Neuropsychiatrica 2014

DOI: 10.1017/neu.2014.17

ACTA NEUROPSYCHIATRICA

Effects of single and combined gabapentin use in elevated plus maze and forced swimming tests

Kilic FS, Ismailoglu S, Kaygisiz B, Oner S, Effects of single and combined gabapentin use in elevated plus maze and forced swimming tests.

Background: Gabapentin, a third-generation antiepileptic drug, is a structural analogue of γ-aminobutyric acid, which is an important mediator of central nervous system. There is clinical data indicating its effectiveness in the treatment of psychiatric illnesses such as bipolar disorder and anxiety disorders.
Objectives: We aimed to investigate the antidepressant and anxiolytic-like effects and mechanisms of gabapentin in rats.
Material and Methods: Female Spraque–Dawley rats weighing 250 ± 20 g were used. A total of 13 groups were formed, each containing 8 rats: gabapentin (5, 10, 20, 40 mg/kg), amitriptyline (10 mg/kg), sertraline (5 mg/kg), diazepam (5 mg/kg), ketamine (10 mg/kg), gabapentin 20 mg/kg was also combined with amitriptyline (10 mg/kg), sertraline (5 mg/kg), diazepam (5 mg/kg) and ketamine (10 mg/kg). All the drugs were used intraperitoneally as single dose. Saline was administered to the control group. Elevated plus maze and forced swimming tests were used as experimental models of anxiety and depression, respectively.
Results: It was observed that gabapentin showed an anxiolytic-like and antidepressant-like effect in all doses in rats. Its antidepressant effect was found to be the same as the antidepressant effects of amitriptyline and sertraline. There was no change in the antidepressant effect when gabapentin was combined with amitriptyline and ketamine, but there was an increase when combined with sertraline and diazepam. Gabapentin and amitriptyline showed similar anxiolytic effect, whereas ketamine and diazepam had more potent anxiolytic effect compared with them.
Conclusions: These data suggest that gabapentin may possess antidepressant- and anxiolytic-like effects.

Fatma Sultan Kilic[1], Sule Ismailoglu[1], Bilgin Kaygisiz[1], Setenay Oner[2]

[1]Department of Pharmacology, Medical Faculty, Eskisehir Osmangazi University, Eskisehir, Turkey; and [2]Department of Biostatistics, Medical Faculty, Eskisehir Osmangazi University, Eskisehir, Turkey

Keywords: antidepressant; anxiolytic; elevated plus maze; forced swimming test; gabapentin

Prof. Dr. Fatma Sultan Kilic, Eskisehir Osmangazi University, Medical Faculty, Department of Pharmacology, Meselik, Eskisehir, Turkey.
Tel: +0 222 239 2979/4564;
Fax: +90 222 239 3772;
E-mail: fskilic@ogu.edu.tr

Accepted for publication June 14, 2014

First published online July 30, 2014

Significant Outcomes

- Gabapentin is an antiepileptic agent. Additional antidepressant- and anxiolytic-like effects of gabapentin will present an option in the treatment of epileptic patients with depression and anxiety or epilepsy-associated depression and anxiety.
- Gabapentin is used in the treatment of neuropathic pain. Anxiety and depression can be seen as comorbidities in patients with neuropathic pain. Therefore, to use gabapentin in the treatment of neuropathic pain may help to resolve depression and anxiety, which can be seen in neuropathic pain.
- Prefering gabapentin in the conditions mentioned above will prevent the use of multiple drugs and associated adverse drug interactions and will provide economic benefits.

Limitations

- Primary limitation is the lack of motor outcome to estimate the construct validity of the test outcome measures.
- We could not study the mechanism of action of gabapentin on receptor level and on neurotransmitters; however, we are planning to perform these in the future.
- We could not study the effects of gabapentin on different animal models of depression and anxiety.
- The second limitation is related to our limited financial support because we are financially supported for only two scientific projects per year.

Introduction

Gabapentin, an analogue of the neurotransmitter γ-aminobutyric acid (GABA), is a third-generation antiepileptic drug. It is used not only in the treatment of partial epileptic seizures but also in the management of neuropathic pain. It is also reported that gabapentin is effective in the treatment of anxiety disorders and depression period of bipolar disorder (1,2). Case reports supported the use of gabapentin in the management of depressive symptoms in bipolar disorder (3). In accordance, bipolar disorder has been the first psychiatric disease in which gabapentin was assessed for its efficacy in psychiatric disorders. Furthermore, pregabalin, a third-generation anticonvulsant drug and an analogue of GABA similar to gabapentin, has been approved for the treatment of generalised anxiety (4–6). GABAergic inhibitory processes have been reported to be tightly related to anxiety (7). Previous studies have shown that drugs of the main benzodiazepine subunit (α) of the $GABA_A$ receptors are able to modify anxiolytic responses (8,9). It has also been suggested that gabapentin, which affects GABA metabolism without an effect on GABA receptors, may be effective in the treatment of anxiety disorders (10). Some researchers also reported that gabapentin showed antianxiety activity in the elevated plus maze test that is a behavioural animal test to study the anxiolytic-like effects of drugs (11,12). To the best of our knowledge, there are no sufficient studies investigating the antidepressant-like effect of gabapentin in animals. However, Valente et al. (13) suggested that α2δ ligands, gabapentin and pregabalin positively modulated adult hippocampal neurogenesis, which is thought to be deregulated in various neuropsychiatric disorders, including major depression, and especially pregabalin confirmed these effects by preventing depression-like behaviours induced by chronic restraint stress in mice.

In this study, we aimed to examine the antidepressant-like and anxiolytic-like effects of gabapentin at different doses. In addition, we aimed to compare the effects of gabapentin with amitriptyline, sertraline, diazepam and ketamine, and to investigate the effects when gabapentin was combined with amitriptyline, sertraline, diazepam and ketamine. Amitriptyline, a tricyclic antidepressant drug; sertraline, an antidepressant drug classified in selective serotonin reuptake inhibitors (SSRI); diazepam, a benzodiazepine derivative; and ketamine, an *N*-methyl-D-aspartate (NMDA) receptor antagonist were selected as they are accepted as reference drugs functioning in different mechanisms. Elevated plus maze test was chosen to evaluate the anxiolytic-like effect as a drug that may be suggested to have an anxiolytic-like effect if increased time spent in open arms of the maze is observed with the drug administration. To evaluate the antidepressant-like effect, forced swimming test was chosen, because a drug may be stated to have an antidepressant-like effect if it decreases the immobility time in the forced swimming test.

Material and methods

Animals

Female Sprague–Dawley rats (250 ± 20 g) were used for the study and sheltered in standard laboratory conditions of light with 12 h light/dark cycle and temperature of 22 ± 2°C. Food and water were available *ad libitum*. Experiments were conducted with the permission of the Local Ethical Committee for Animal Experimentation of Eskisehir Osmangazi University Medical School (19/2/2010-147). Animals were brought to the experiment room 1 h before the experiments for habituation.

Drugs

Gabapentin (Sigma, Germany), amitriptyline (Deva, Istanbul, Turkey), diazepam (Deva), sertraline (Pfizer, Istanbul, Turkey) and ketamine (Pfizer) were used in the study. All the drugs were dissolved in saline. Saline was administered to the rats in the control group. All solutions were prepared freshly on test days and administered intraperitoneally (i.p.) in a volume of 2 ml/kg body weight of rats.

Study design

All the experiments were conducted at the Animal Experiment Laboratories, Department of

https://doi.org/10.1017/neu.2014.17 Published online by Cambridge University Press

Pharmacology, Eskişehir Osmangazi University, Medical School, Eskisehir, Turkey, from February to May 2011.

Animals were randomly divided into 13 groups each containing 8 rats as below:

Group 1: control (saline)
Group 2: gabapentin 5 mg/kg
Group 3: gabapentin 10 mg/kg
Group 4: gabapentin 20 mg/kg
Group 5: gabapentin 40 mg/kg
Group 6: amitriptyline 10 mg/kg
Group 7: gabapentin 20 mg/kg + amitriptyline 10 mg/kg
Group 8: diazepam 5 mg/kg
Group 9: gabapentin 20 mg/kg + diazepam 5 mg/kg
Group 10: sertraline 5 mg/kg
Group 11: gabapentin 20 mg/kg + sertraline 5 mg/kg
Group 12: ketamine 10 mg/kg
Group 13: gabapentin 20 mg/kg + ketamine 10 mg/kg

Anxiety behaviour was assessed with elevated plus maze test, and depression was assessed with forced swimming test. Rats used for behavioural testing were used in only a single behavioural paradigm (elevated plus maze or forced swimming test). There were two experimenters who conducted the experiments, one experimenter who was blind to the drug treatment made the observations and scoring, and the other experimenter performed the drug treatments.

Elevated plus maze test

This model was performed as Pellow et al. (14) described. Experiments were conducted in a laboratory isolated from sound under low light conditions to encourage exploration between 09:00 and 13:00 h. After 1 h, the dosing rats were individually placed in the centre of the maze, a set up, that is 50 cm high and that has two open (50 × 10 cm) and two closed arms (50 × 10 × 40 cm). Entry into an arm was defined as all four paws crossing from the central region into the arm. The time spent in the open and closed arms was recorded for 5 min and manually scored by a trained experimenter blind to drug treatment. The maze was cleaned and dried between animals.

Forced swimming test

This test was executed in rats as Porsolt et al. (15) described. Rats were forced to swim in a cylinder with a water level of 25 cm and 25 ± 0.5°C. Rats were exposed to a trial session for 15 min, 24 h before the test procedure without any observation. In the test procedure, immobility and struggling times were recorded for 5 min and manually scored by an observer blind to drug treatment. Immobility is defined as the position that the rat gives up struggling and floats in the water with only small movements necessary to keep its head above water. The water in the cylinder was changed between each rat.

Statistical analysis

The data collected were evaluated with one-way ANOVA analysis by using PASW 18.0 packet program. Tukey test was used as the *post hoc* test. All the groups were statistically compared with each other. The results are given as mean ± SEM.

Results

Results of the single use of gabapentin on the elevated plus maze and forced swimming tests

We observed that gabapentin increased the time spent in the open arms of the elevated plus maze ($p < 0.05$) and decreased the immobility time in the forced swimming test at all doses ($p < 0.05$), as shown in Figs 1 and 2.

Results of the combined use of gabapentin on the elevated plus maze and forced swimming tests

Elevated plus maze test. A dose of 20 mg/kg of gabapentin was selected to use in the combination groups of gabapentin. We found that gabapentin 20 mg/kg dose alone increased the time spent in the open arms of the elevated plus maze comparably with amitriptyline 10 mg/kg dose alone. In the combined use of gabapentin and amitriptyline, there was a more increase in the time spent in the open arms, suggesting that gabapentin potentiated the effect of amitriptyline ($p < 0.05$; Fig. 3). Sertraline 5 mg/kg alone decreased the time spent in the open arms of the elevated plus maze comparably with the control group; however, it significantly decreased the time spent in the open arms compared with amitriptyline 10 mg/kg alone and gabapentin 20 mg/kg alone ($p < 0.05$; Fig. 3). In addition, the combination of gabapentin 20 mg/kg and sertraline 5 mg/kg decreased the time spent in the open arms compared with gabapentin 20 mg/kg alone, pointing out that sertraline diminished the effect of gabapentin ($p < 0.05$; Fig. 3). A dose of 5 mg/kg diazepam alone was found to increase the time spent in the open arms compared with gabapentin 20 mg/kg alone and control ($p < 0.05$). The increase in the time spent in the open arms was much more in the combination group of diazepam

https://doi.org/10.1017/neu.2014.17 Published online by Cambridge University Press

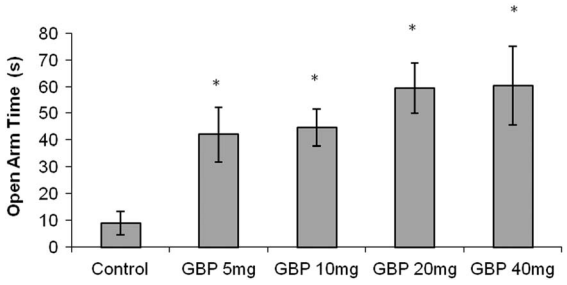

Fig. 1. The effects of gabapentin on the time spent in the open arms in elevated plus maze test. Values are given as mean ± SEM. *$p < 0.05$ compared with control. Statistical analysis was performed with one-way ANOVA, and Tukey test was used as the *post hoc* test. GBP, gabapentin.

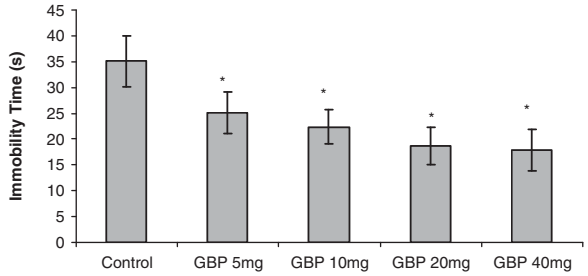

Fig. 2. The effects of gabapentin on immobility time in forced swimming test. Values are given as mean ± SEM. *$p < 0.05$ compared with control. Statistical analysis was performed using one-way ANOVA, and Tukey test was used as the *post hoc* test. GBP, gabapentin.

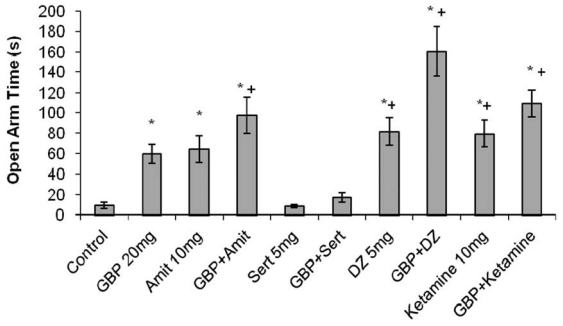

Fig. 3. The effects of single or combined use of gabapentin, amitriptyline, sertraline, ketamine and diazepam on the time spent in the open arms in the elevated plus maze test. Values are given as mean ± SEM. *$p < 0.05$ compared with control, +$p < 0.05$ compared with GBP 20 mg/kg. Statistical analysis was performed using one-way ANOVA, and Tukey test was used as the *post hoc* test. Amit, amitriptyline; DZ, diazepam; GBP, gabapentin, Sert, sertraline.

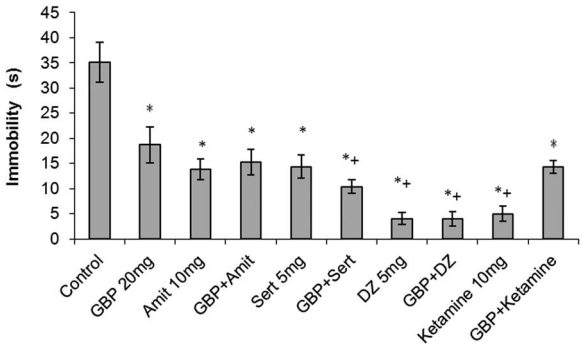

Fig. 4. The effects of single or combined use of gabapentin, amitriptyline, sertraline, ketamine and diazepam on immobility time in forced swimming test. Values are given as mean ± SEM. *$p < 0.05$ compared with control, +$p < 0.05$ compared with GBP 20 mg/kg. Statistical analysis was performed with one-way ANOVA, and Tukey test was used as the *post hoc* test. Amit, amitriptyline; DZ, diazepam; GBP, gabapentin, Sert, sertraline.

and gabapentin compared with gabapentin 20 mg/kg alone and diazepam 5 mg/kg alone, which evokes the thought that gabapentin potentiated the anxiolytic effect of diazepam (Fig. 3). Ketamine 10 mg/kg alone increased the time spent in the open arms compared with gabapentin 20 mg/kg alone and control ($p < 0.05$). More increase in the time spent in open arms was observed with the combination of ketamine and gabapentin compared with their single administrations ($p < 0.05$). The increase in the time spent in the open arms was similar in the 10 mg/kg ketamine and 5 mg/kg diazepam groups (Fig. 3) and in the gabapentin 20 mg/kg and amitriptyline 10 mg/kg groups (Fig. 3). The increase in the time spent in the open arms observed with ketamine alone and diazepam alone groups was more than observed in the gabapentin 20 mg/kg alone and amitriptyline 10 mg/kg alone groups ($p < 0.05$; Fig. 3). Combinations of gabapentin with amitriptyline, diazepam and ketamine exerted more increase in the time spent in open arms compared with single administration of 20 mg/kg gabapentin ($p < 0.05$) (Fig. 3). It was observed that there was no alteration in the time spent in the open arms when gabapentin 20 mg/kg and sertraline 5 mg/kg were used together compared with control ($p > 0.05$); however, a decrease in the time spent in the open arms was observed compared with gabapentin 20 mg/kg alone ($p < 0.05$; Fig. 3).

Forced swimming test. A dose of 20 mg/kg of gabapentin was selected to use in the combination groups of gabapentin. Single administrations of gabapentin 20 mg/kg, amitriptyline 10 mg/kg and sertraline 5 mg/kg similarly decreased the immobility time compared with control ($p < 0.05$; Fig. 4). There was no change in the immobility time when gabapentin 20 mg/kg and amitriptyline 10 mg/kg were used in combination compared with their single administrations ($p > 0.05$; Fig. 4). When gabapentin 20 mg/kg and sertraline 5 mg/kg were

https://doi.org/10.1017/neu.2014.17 Published online by Cambridge University Press

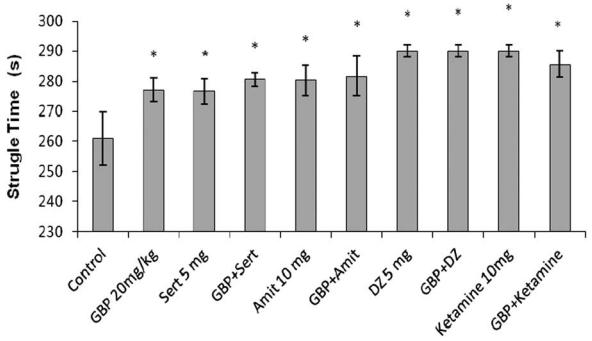

Fig. 5. The effects of single or combined use of gabapentin, amitriptyline, sertraline, ketamine and diazepam on struggle time. Values are given as mean ± SEM. *$p < 0.05$ compared with control. Statistical analysis was performed using one-way ANOVA, and Tukey test was used as the *post hoc* test. Amit, amitriptyline; DZ, diazepam; GBP, gabapentin, Sert, sertraline.

used in combination, a significant decrease in the immobility time was observed compared with their single administrations, suggesting a potentialisation in their effects ($p < 0.05$; Fig. 4).

Diazepam 5 mg/kg showed a significant decrease in the immobility time compared with gabapentin 20 mg/kg and control ($p < 0.05$). In addition, the decrease in the immobility time in the combined group of diazepam and gabapentin was comparable to the single use of diazepam but greater than single administration of gabapentin ($p < 0.05$; Fig. 4). Ketamine 10 mg/kg showed a significant decrease in the immobility time compared with gabapentin 20 mg/kg and control; however, combined use of ketamine and gabapentin increased the immobility time compared with single ketamine administration ($p < 0.05$); however, no change was observed compared with gabapentin 20 mg/kg alone ($p > 0.05$; Fig. 4).

There was no change in the immobility time when gabapentin was combined with amitriptyline and ketamine compared with gabapentin 20 mg/kg alone ($p > 0.05$); however, a decrease in the immobility time was observed compared with gabapentin 20 mg/kg alone when gabapentin was combined with sertraline and diazepam ($p < 0.05$; Fig. 4). There were significant increases in all groups compared with the control group in terms of struggle time ($p < 0.05$; Fig. 5). This finding supports the antidepressant-like effect of all experimental groups in the study.

Discussion

In this study, we observed that gabapentin increased the open arm time in the elevated plus maze test, suggesting an anxiolytic-like effect, and decreased immobility time in the forced swimming test, suggesting an antidepressant-like effect. When we used gabapentin in combination, a more increased open arm time was observed with the combinations of amitriptyline, diazepam and ketamine, except sertraline, and a more decreased immobility time was observed with the combinations of sertraline and diazepam, except amitriptyline and ketamine.

The clinical use of gabapentin in anxiety and its anxiolytic-like effect in preclinical studies were reported. Pollack et al. (16) reported that they observed a reduction in anxiety symptoms after adjuvant gabapentin use in two patients with generalised anxiety disorder who did not respond to or partially responded to anxiolytic medications. Kulkarni et al. (11) reported that gabapentin (10, 20 mg/kg) increased the number of open arm entries and open arm time in the elevated plus maze. Similarly, de-Paris et al. (12) observed that gabapentin (10, 30 mg/kg) increased the number of open arm entries and open arm time in the elevated plus maze in rats. These findings are consistent with our finding that gabapentin (5, 10, 20, 40 mg/kg) increased the open arm time in the elevated plus maze.

Gabapentin is known to have an inhibitory effect on the central nervous system with no direct effect on GABA receptors. Similarly, it was reported that some of the benzodiazepine derivatives evoke GABAmimetic effect by affecting GABA receptors indirectly (17,18). There are also studies reporting that diazepam, a benzodiazepine derivative, acts not by binding to GABA receptors but by interacting with clorur channels and GABA receptors coupled-central benzodiazepine receptors (18,19). Thus, it may be suggested that the combination of gabapentin with benzodiazepine derivatives may result in a potentiated effect. This is supported in our study. We observed a potentialisation in the anxiolytic-like effects of gabapentin and diazepam in the elevated plus maze test when they were administered in combination.

Some studies indicate that benzodiazepines also have antidepressant effect, besides their anxiolytic effect (20,21). It was suggested that benzodiazepines have no effect on biogenic amine reuptake and that does not affect their metabolism, but show antidepressant effects as a $GABA_A$ receptor agonist through the GABA theory of depression (20). In this study, our results supported these suggestions. We observed that diazepam displayed an antidepressant-like effect in the forced swimming test. This effect was more than the antidepressant-like effect observed with gabapentin in the forced swimming test, and there was no alteration in their antidepressant-like effects when administered in combination. On the

https://doi.org/10.1017/neu.2014.17 Published online by Cambridge University Press

contrary, it is not convenient to use benzodiazepines for long-term depression treatment, and hence they have addiction potential. It is suggested that these addiction rates are seen in 1/3 of the patients when benzodiazepines and antidepressants are used together (22). Consequently, gabapentin might be an alternative treatment option with regard to the addiction potential of benzodiazepines.

In experimental anxiety models, there are conflicting data on the effects of amitriptyline, a tricyclic antidepressant drug, which is in use for a long time (22). It was shown that both acute (23) and chronic (24) amitriptyline administrations had no anxiolytic-like effect in elevated plus maze. Even, chronic use of amitriptyline was shown to exhibit anxiogenic-like activity (25). Other studies reported that amitriptyline had non-anxiolytic (26), anxiogenic (27) or anxiolytic effect only after chronic administration (28). In our study, we observed that amitriptyline had an acute anxiolytic-like effect in the elevated plus maze test, and when combined with gabapentin, a potentiated effect was observed. We also observed that amitriptyline showed an antidepressant-like effect in the forced swimming test similar to sertraline and gabapentin, and no change in the effects in the forced swimming test was observed when amitriptyline and gabapentin were used in combination. However, combination of them with their half doses may be helpful to reduce their adverse effects.

Sertraline, classified in SSRI antidepressants, causes anxiety reaction in acute administration. This effect disappears in 15 days. It was observed that acute sertraline administration (5 mg/kg i.p.) increased anxiety-like symptoms in rats. In our study, we found that sertraline showed anxiogenic-like effect in the elevated plus maze test and anxiolytic-like effect of gabapentin disappeared in the elevated plus maze test when combined with sertraline. Sertraline has no sedative effect and does not affect psychomotor performance. Sertraline does not enhance catecholaminergic activity and has no affinity for dopamine, adrenaline, histamine, GABA and benzodiazepine receptors, which present a different mechanism of action for sertraline from gabapentin (29).

In our study, we observed that gabapentin had an antidepressant-like effect similar to sertraline, and combined use of sertraline and gabapentin showed an increased antidepressant-like effect in the forced swimming test. This increased effect may be as a consequence of the effects of gabapentin on neurotransmitter levels as reported in a study that gabapentin increased GABA turnover and serotonin levels in blood (30).

Evidence shows that ionotrophic glutamate NMDA receptors are involved in the ethiology of anxiety and major depressive disorders (31,32), and changes in seven transmembranal segment subunit of metabotrophic glutamate receptors are associated with anxiety and some other diseases such as epilepsy, Parkinson's disease, Huntington's disease and schizophrenia (33). Ketamine, a non-selective NMDA receptor antagonist and a dissociative anaesthetic agent, had anxiolytic-like effects in elevated plus maze (34). It was suggested that ketamine showed its anxiolytic effect by reducing theta waving in the hippocampus similar to diazepam, which is a common mechanism of action in all anxiolytic drugs. This feature supplies an evidence for anxiolytic effect of ketamine (35). In our study, we observed that subanaesthetic doses of ketamine showed an anxiolytic-like effect more than gabapentin and similar to diazepam in the elevated plus maze test. In addition, a potentialisation in the anxiolytic-like effect of gabapentin and ketamine was observed in case of their combined use.

There are studies reporting the antidepressant effect of ketamine at subanaesthetic doses in patients with depression without any side effects on memory (36). In addition, in a study, ketamine was shown to reduce behavioural despair in forced swimming test. It was reported that ketamine displays antidepressant effect through NMDA receptor inhibition (37) and also genetic factors are involved in its antidepressant effect (38). We also observed that ketamine had antidepressant-like effect in the forced swimming test and a slight decrease in the antidepressant-like effect was observed in the combination of gabapentin and ketamine compared with the single use of ketamine; however, this was not statistically significant.

As a conclusion, we suggest that gabapentin shows anxiolytic-like and antidepressant-like effects and its combinations with amitriptyline, diazepam and ketamine result in a potentiated anxiolytic-like effect and with sertraline and diazepam result in a potentiated antidepressant-like effect. In the light of these findings, gabapentin might be a choice in the treatment of depression and anxiety both alone or in a combined drug use. In addition, gabapentin use may be helpful in the treatment of anxiety and depression, which may be seen as comorbities in neuropathic pain.

Acknowledgements

Prof. Dr. Fatma Sultan Kilic contributed to the conception, design, analysis and interpretation of data, and drafting and revising the article for important intellectual content and final approval of the version to be published. Sule Ismailoglu, MSc, contributed to the acquisition and analysis of data and

https://doi.org/10.1017/neu.2014.17 Published online by Cambridge University Press

drafting and final approval of the version to be published. Dr. Bilgin Kaygisiz, MD, contributed to revising the article for important intellectual content and final approval of the version to be published. Assoc. Prof. Dr. Setenay Oner contributed to the statistical analysis and interpretation of data; final approval of the version to be published.

Financial Support

This research received no specific grant from any funding agency, commercial or not-for-profit sectors.

Conflicts of Interest

None.

Ethical Standards

The authors assert that all procedures contributing to this work comply with the ethical standards of the relevant national and institutional guides on the care and use of laboratory animals.

References

1. Karataş G, Tamam L, Ozpoyraz N. Gabapentin in the treatment of anxiety disorders. Bull Clin Psychopharmacol 2003;**13**:37–42.
2. Young LT, Robb JC, Hasey GM et al. Gabapentin as an adjunctive treatment in bipolar disorder. J Affect Disord 1999;**55**:73–77.
3. Egashira T, Inoue T, Shirai Y, Iwata K, Honma J, Koyama T. Adjunctive gabapentin for treatment-resistant insomnia of bipolar disorder: a case report. Clin Neuropharmacol 2011;**34**:129–130.
4. Wettermark B, Brandt L, Kieler H, Bodén R. Pregabalin is increasingly prescribed for neuropathic pain, generalised anxiety disorder and epilepsy but many patients discontinue treatment. Int J Clin Pract 2014;**68**:104–110.
5. Darbà J, Kaskens L, Pérez C, Álvarez E, Navarro-Artieda R, Sicras-Mainar A. Pharmacoeconomic outcomes for pregabalin: a systematic review in neuropathic pain, generalized anxiety disorder, and epilepsy from a Spanish perspective. Adv Ther 2014;**31**:1–29.
6. Tassone DM, Boyce E, Guyer J, Nuzum D. Pregabalin: a novel gamma-aminobutyric acid analogue in the treatment of neuropathic pain, partial-onset seizures, and anxiety disorders. Clin Ther 2007;**29**:26–48.
7. Calza A, Sogliani C, Santoru F et al. Neonatal exposure to estradiol in rats influences neuroactive steroid concentrations, $GABA_A$ receptor expression, and behavioral sensitivity to anxiolytic drugs. J Neurochem 2010;**113**:1285–1295.
8. Smith SS. α4βδ $GABA_A$ receptors and tonic inhibitory current during adolescence: effects on mood and synaptic plasticity. Front Neural Circuits 2013;**7**:135.
9. Gulinello M, Gong QH, Smith SS. Progesterone withdrawal increases the anxiolytic actions of gaboxadol: role of α4βδ $GABA_A$ receptors. Neuroreport 2003;**14**:43–46.
10. de-Paris F, Sant'Anna MK, Vianna MR et al. Effects of gabapentin on anxiety induced by simulated public speaking. J Psychopharmacol 2003;**17**:184–188.
11. Kulkarni SK, Singh K, Bishnoi M. Comparative behavioural profile of newer antianxiety drugs on different mazes. Indian J Exp Biol 2008;**46**:633–638.
12. de-Paris F, Busnello JV, Vianna MR et al. The anticonvulsant compound gabapentin possesses anxiolytic but not amnesic effects in rats. Behav Pharmacol 2000;**11**:169–173.
13. Valente MM, Bortolotto V, Cuccurazzu B et al. α2δ ligands act as positive modulators of adult hippocampal neurogenesis and prevent depression-like behavior induced by chronic restraint stress. Mol Pharmacol 2012;**82**:271–280.
14. Pellow S, Chopin P, File SE, Briley M. Validation of open: closed arm entries in an elevated plus-maze as a measure of anxiety in the rat. J Neurosci Methods 1985;**14**:149–167.
15. Porsolt RD, Anton G, Blavet N, Jalfre M. Behavioural despair in rats: a new model sensitive to antidepressant treatments. Eur J Pharmacol 1978;**47**:379–391.
16. Pollack MH, Matthews J, Scott EL. Gabapentin as a potential treatment for anxiety disorders. Am J Psychiatry 1998;**155**:992–993.
17. Foye W. Central nervous system depressants: sedative-hypnotics, benzodiazepines. In: William O. Foye, editor. Principles of Medicinal Chemistry, 3rd edn. Pennsylvania, PA: Lea and Febiger, 1989. p. 165–166.
18. Kayaalp O. Antiepileptik İlaçlar. In: Rasyonel Tedavi Yönünden Tıbbi Farmakoloji, vol. 2. Ankara, 2009. p. 884–901.
19. Gilman GA, Goodman S, Rail T, Mured F. Goodman and Gilman's The Pharmacological Basis of Therapeutics. USA: McGraw-Hill, 2006; p. 339–351.
20. Patty F, Trivedi MH, Fulton M, Rush AJ. Benzodiazepines as antidepressant: does GABA play a role in depression? Biol Psychiatry 1995;**38**:578–591.
21. Furukawa TA, Streiner D, Young LT, Kinosta Y. Antidepressant plus benzodiazepine for major depression. Cochrane Database Syst Rev 2000;**4**:CD001026.
22. Feighner JP. Overview of antidepressants currently used to treat anxiety disorders. J Clin Psychiatry 1999;**60**:18–22.
23. Parra A, Everss E, Monleon S, Vinader-Caerols C, Arenas MC. Effects of acute amitriptyline administration on memory, anxiety and activity in male and female mice. Neurosci Res Commun 2002;**31**:135–144.
24. Everss E, Arenas MC, Vinader-Caerols C, Monleon S, Parra A. Piracetam counteracts the effects of amitriptyline on inhibitory avoidance in CD1 mice. Behav Brain Res 2005;**159**:235–242.
25. Weinstock M, Poltyrev V, Bejar C, Youdim MB. Effect of TV3326, a novel monoamine-oxidase cholinesterase inhibitor, in rats models of anxiety and depression. Psychopharmacology 2002;**160**:318–324.
26. Bilkei-Gorzo A, Gyertyan I, Levay G. mCPP-induced anxiety in the light-dark box in rats: a new method for screening anxiolytic activity. Psychopharmacology (Berl) 1998;**136**:291–298.
27. Bodnoff SR, Suranyi-Cadotte B, Quirion R, Meaney MJ. A comparison of the effects of diazepam versus several typical and atypical antidepressant drugs in an animal model of anxiety. Psychopharmacology (Berl) 1989;**97**:277–279.
28. Bodnoff SR, Suranyi-Cadotte B, Aitken DH, Meaney MJ. The effects of chronic antidepressant treatment in an animal model of anxiety. Psychopharmacology (Berl) 1998;**95**:298–302.

https://doi.org/10.1017/neu.2014.17 Published online by Cambridge University Press

29. Askin R, Turan M, Cilli AS, Kaya N. Clomipramine versus sertraline in the treatment of obsessive compulsive disorder. Eur Neuropsychopharmacol 1997;**7**:228–229.
30. Rao D, Tovar M, Oseroff SB et al. Crystal-field and exchange interactions of dilute Gd + 3 ions in Eu_2CuO_4. Phys Rev B Condens Matter 1988;**38**:8920–8922.
31. Javitt DC. Glutamate as a therapeutic target in psychiatric disorders. Mol Psychiatry 2004;**9**:984–997.
32. Krystal JH, D'Souza DC, Petrakis IL et al. NMDA agonists and antagonists as probes of glutamatergic dsyfunction and pharmacotherapies in neuropsychiatric disorders. Harv Rev Psychiatry 1999;**7**:125–143.
33. Vizi ES. Role of high-affinity receptors and membrane transporters in nonsynaptic communication and drug action in central nervous system. Pharmacol Rev 2000;**52**:63–90.
34. Engin E, Treit D, Dickson CT. Anxiolytic and antidepressant-like properties of ketamine in behavioral and neurophysiological animal models. Neuroscience 2009;**161**:359–369.
35. Engin E, Treit D, Dickson CT. Corrigendum to 'anxiolytic- and antidepressant-like properties of ketamine in behavioral and neurophysiological animal models'. Neuroscience 2009;**162**:1438–1439.
36. Maeng S, Carlos A, Zarate JR et al. Cellular mechanisms underlying the antidepressant effects of ketamine: role of α-amino-3hydroxy-5-methylisoxazole-4-propionic acid receptors. Biol Psychiatry 2008;**63**:349–352.
37. Pirrenger C, Sanacora G, Krystal JH. The NMDA receptor as a therapeutic target in major depressive disorders. CNS Neurol Disord Drug Targets 2007;**6**: 101–115.
38. Phelps LE, Brutsche N, Moral JR. Family history of alcohol dependence and initial antidepressant response to an NMDA antagonist. Biol Psychiatry 2008;**65**:181–184.

https://doi.org/10.1017/neu.2014.17 Published online by Cambridge University Press