Journal of Pharmacognosy and Phytochemistry

Available online at www.phytojournal.com

E-ISSN: 2278-4136
P-ISSN: 2349-8234
JPP 2020; 9(1): 474-480
Received: 16-11-2019
Accepted: 18-12-2019

AA Bhingare
S.N.D. College of Pharmacy, Yeola, Maharashtra, India

Rd Bhalke
Sanjivani College of Pharmaceutical Education and Research, Kopargaon, Maharashtra, India

K Vanitha Prakash
Shri Sai Jyoti College of pharmacy, Vattinagula Pally, Gandhi Peth, Hyderabad, India

Sanjay B Kasture
Pinnacle Biomedical Research Institute, Bhopal, Madhya Pradesh, India

MA Giri
Sanjivani College of Pharmaceutical Education and Research, Kopargaon, Maharashtra, India

Corresponding Author:
MA Giri
Sanjivani College of Pharmaceutical Education and Research, Kopargaon, Maharashtra, India

Antidepressant activity of ascorbic acid, caffeine and sertraline

AA Bhingare, Rd Bhalke, K Vanitha Prakash, Sanjay B Kasture And MA Giri

Abstract
Depression is major serious CNS disorder occurring in world which recorded about 450 million people suffer from mental or behavioural disorder according to world health report (Murugan, 2011). In the traditional systems of medicine, many plants have been used to treat depression for thousands of years. For screening of antidepressant activity tail suspension test, forced swim test and splash test have been used. During the study of Ascorbic acid (AA) (5 /10 mg/kg i. p.), Caffeine (5 mg/kg i.p.) sertraline (5 mg/kg i.p.) and fluoxetine (10 mg/kg p.o.) were administered individually and in combination. Ascorbic acid and caffeine combination at (5+5 mg/kg) has shown significant (P<0.01) increased in latency to immobility. A significant (P<0.01) decrease in the duration of immobility was seen in all treatment but marked result is shown in C + S (5+5 mg/kg) treated group. A significant (P<0.01) increase in time spent in climbing in Caffeine (5 mg/kg) treated group is observed. Sertraline and caffeine combination at (5+5 mg/kg) showed significant increase in time spent in swimming. Caffeine at 5 mg/kg has shown significant (P<0.01) increased in latency to immobility in acute tail suspension test as compare to sertraline whereas significant (P<0.01) decrease in the duration of immobility was seen in all treatment but marked result is shown in C + S (5+5 mg/kg) treated group. AA + C (10+5 mg/kg) has shown significant (P<0.01) decreased in latency to grooming whereas significant (P<0.01) increase in the duration of grooming was seen in AA (10mg/kg) and AA + C (10+5 mg/kg) treated groups. Present study was undertaken to evaluate the antidepressant potential ascorbic acid, caffeine, sertraline and their combinations.

Keywords: Ascorbic acid, sertraline, antidepressant, tail suspension test, forced swim test, splash test

Introduction
Depression is major serious CNS disorder occurring in world which recorded about 450 million people suffer from mental or behavioural disorder according to world health report [1]. The prevalence of depression is increasing day to day due to hectic life style. Women are more affected than men with a ratio of 2:1. In depression the level of neurotransmitter such as serotonin, norepinephrine, and dopamine are decreases [2] and also evidence suggest that increase oxidative stress [3, 5]. Now a day natural medicines are most preferable to treat depression rather than synthetic antidepressant drugs to conquer its side effect such as vomiting, nausea, irritation etc. Ascorbic acid is an essential micro-nutrient for human which obtained from plant origin. In various factors like age, smoking, diabetic, chronic diseases such as rheumatoid arthritis and cancer as well as in depression there is increase in oxidative stress. Ascorbic acid is essential for mental and emotional well-being. Depletion or deficiency of this vitamin may trigger depression, irritability, neurological, cardiac disorders, anxiety and fatigue. Anxiety has shown the increased rate of breakdown of ascorbic acid. It can be treated by increase dietary levels of ascorbic acid [6, 7]. Sertraline is selective serotonin reuptake inhibitor type of antidepressant agent; it acts like a functional antagonist of 5-HT3 receptors which metabotropic class of receptor for antidepressant effect [8]. Many studies have suggested that sertraline blocks the uptake of serotonin into human platelets. Animal study suggest that sertraline is selective inhibitor of neuronal serotonin reuptake but it has shown weak effects on norepinephrine and dopamine neuronal reuptake. Caffeine is xanthine derivative which psychomotor stimulant and antinociceptive activity [2]. Caffeine has ability to block adenosine A1 receptors by increasing extracellular level of serotonin [9, 10]. In animal study, Tail suspension test (TST), forced swim test (FST) and splash test are used for evaluation of antidepressant drugs. But till now no scientific works have been reported on antidepressant activity of caffeine and ascorbic acid combination. In light of above information, the present study has been undertaken to study the antidepressant and antioxidant effects of caffeine and ascorbic acid combination.

Materials and Methods

Animals

Healthy Swiss Albino female mice (22-25) g. each group contained 5 animals which had maintained at a temperature of 25 ˚C ± 1 ˚C and relative humidity of 45 to 55% and under standard environmental condition (12 hr. light and 12 hr. dark cycle). The animal had free access to standard pellet rodent diet and good quality water was provided throughout the study. The experimental protocol was approved by Institutional Animals Ethics Committee. The animal care was taken as per the guidelines of Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA).

Drugs and Chemicals

Ascorbic acid, sertraline, caffeine and fluoxetine were dissolved in distilled water and administered by intraperitonially route (i.p), only fluoxetine was administered orally (P.O.).

Antidepressant activity

Modified Forced swim test

The test was conducted using the method given by Porsolt et al. Principle of forced swim test is induced characteristics behaviour of immobility by forced the animal to swim in restricted space from which animal cannot escape that indicate depression in animal whereas it is similar to human depression. Mice were forced to swim inside a vertical Plexiglas cylinder (height: 40 cm; diameter: 18 cm, containing 15 cm of water maintained at 25 ˚C). Mice were placed individually in the cylinders and allowed to swim for 6 min. After 6 min in the water the mice were removed and allowed to dry in a heated enclosure (32 ^{O}C) before being returned to their home cages. After 24 h, test drugs, standard and vehicle were administered as per their body weight before half hour prior to testing. They were again placed in the cylinder for test and the total duration of immobility, latency to immobility and time spent in climbing was measured during a 6 min test [11, 13].

Tail suspension test

Tail suspension test was performed by Steru et al. in 1985. The principle of this test is that suspending mice from tail, suspended inverted mice produce characteristic behavior immobility which is similar to human depression. Mice were transported from the housing room to test sound-proof testing area. The animals were treated with the test compounds or the vehicle by intraperitoneally injection 30 min prior to testing. For the test mice were suspended on the edge of a shelf 58 cm above a tablet top by adhesive tape placed approximately 1 cm from the tip of the tail. The latency to immobility, duration of immobility was recorded for period of 6 min. Mice were considered immobile when they hung passively and completely motionless [14, 16].

Splash test

Splash test was described by Isingrini et al. in 2010. In this test variety of stressors applied randomly and at varying time of day for 14 days. Ascorbic acid and fluoxetine were dissolved in distilled water and administered orally (10 mg/kg) during the last 7 days of the chronic unpredictable mild stress (CUMS) procedure. Control and stressed animals were maintained in separate room due to stress odor. For this study animals were divided into ten groups. Animal was placed in clear Plexiglas boxes (9x 7x 11 cm) squirting of a 10% sucrose solution on dorsal coat of a mouse, animal initiate grooming behavior due to viscosity of solution. Time spend in grooming was manually recorded for a period 5 min as an index of self-care and motivational behavior [17].

Statistical analysis

The data were expressed as mean ± SEM, difference between three experimental groups were statistically analyzed by one-way analysis of variance (ANOVA) followed by Bonferroni test and Dunnett test.

Results

Forced swim test

Effects of various treatments on latency to immobility, duration of immobility, time spent in climbing and time spent in swimming in forced swim test acute and chronic were given in table 1 and table 2. Ascorbic acid and caffeine combination at (5+5 mg/kg) has shown significant (P<0.01) increased in latency to immobility as shown in Fig. 1. A significant (P<0.01) decrease in the duration of immobility was seen in all treatment but marked result is shown in C + S (5+5 mg/kg) treated group, results are depicted in Fig.2. A significant (P<0.01) increase in time spent in climbing in Caffeine (5 mg/kg) treated group is observed as shown in Fig.3. Sertraline and caffeine combination as shown in Fig.4 at (5+5 mg/kg) showed significant increase in time spent in swimming.

Table 1: Effect of Aa, Caffeine, Sertraline and Their Combination In Acute Forced Swim Test

Treatment	Latency to immobility	Duration of immobility	Time spent in climbing	Time spent in swimming
Vehicle (5 ml/kg)	26.4 ± 1.32	142.6 ± 1.58	82.4 ± 1.63	135 ± 1.93
AA (5 mg/kg)	74.3 ± 1.5*	3.6 ± 0.2*	188.4 ± 3.1*	168 ± 2.9*
AA (10 mg/kg)	59.8 ± 1.3*	7 ± 0.7*	181.6 ± 1.9*	171.4 ± 2.15*
C (5 mg/kg)	66.6 ± 1.63*	6.4±1	209.4 ± 2.47*	144.2 ± 1.77*
S (5 mg/kg)	73.8 ± 1.96*	3.4 ± 0.51*	140.6 ± 1.87*	216 ± 2.15*
AA + C (5+5 mg/kg)	78.6 ± 2.76*	3.2 ± 0.3*	181 ± 3.37*	175.8 ± 3.8*
AA + S (5+5 mg/kg)	75.6 ± 2.11*	3 ± 0.31*	143.6 ± 2.54*	213.4 ± 2.9*
AA + C (10+5 mg/kg)	65.4 ± 1.4*	4 ± 0.7*	186.8 ± 1.3*	169.2 ± 1.82*
AA + S (10+5 mg/kg)	72.6 ± 1.2*	3.8 ± 0.58*	136.6 ± 0.92*	219.6 ± 1.36*
C + S (5+5 mg/kg)	75.2 ± 1.5*	2.4 ± 0.5*	137.8 ± 0.9*	219.8 ± 0.5*

Where C-caffeine, S-Sertraline. Data presented as mean±SEM (n=5). *indicate significant (P<0.05) difference from respective control group. Data was analysed by one-way ANOVA followed by Dunnett test.

Table 2: Effect of AA, caffeine, sertraline and their combination in chronic Forced Swim Test

Treatment	Latency to immobility	Duration of immobility	Time spent in climbing	Time spent swimming
Vehicle (5 ml/kg)	23.6 ± 2.2	144 ± 3.7	88 ± 2.8	128 ± 4.5
AA (5 mg/kg)	76 ± 2.6*	3.2 ± 0.3*	180 ±2.5*	176.8 ± 2.8*
AA (10 mg/kg)	61.2 ± 1.5*	6.8 ± 0.58*	183.2 ± 1.71*	170 ± 1.5*
C (5 mg/kg)	67.4 ± 1.43*	6 ± 0.7*	210.4 ± 2.3*	143 ±1.9*
S (5 mg/kg)	75.2 ± 2.08*	3.8 ± 0.58*	129.2 ± 1.65*	227 ± 1.22*
AA + C (5+5 mg/kg)	79 ± 2.1*	3.1 ± 0.3*	180.9 ± 2.0*	176 ± 2.2*
AA + S (5+5 mg/kg)	76.8 ± 1.35*	3 ± 0.31*	139.2 ± 1.46*	217.8 ± 1.2*
AA + C (10+5 mg/kg)	67 ± 1.97*	3.8 ± 0.3*	191 ± 3.4*	168 ± 3.8*
AA + S (10+5 mg/kg)	73 ± 0.92*	2.4 ± 0.5*	141 ± 1.36*	215.6 ± 1.65*
C + S (5+5 mg/kg)	76.2 ± 1.15*	2.2 ± 0.3*	138.6 ± 1.03*	219.2 ± 0.58*

Where C-caffeine, S-Sertraline. Data presented as mean±SEM (n=5). *indicate significant (P<0.05) difference from respective control group. Data was analysed by one-way ANOVA followed by Dunnett test.

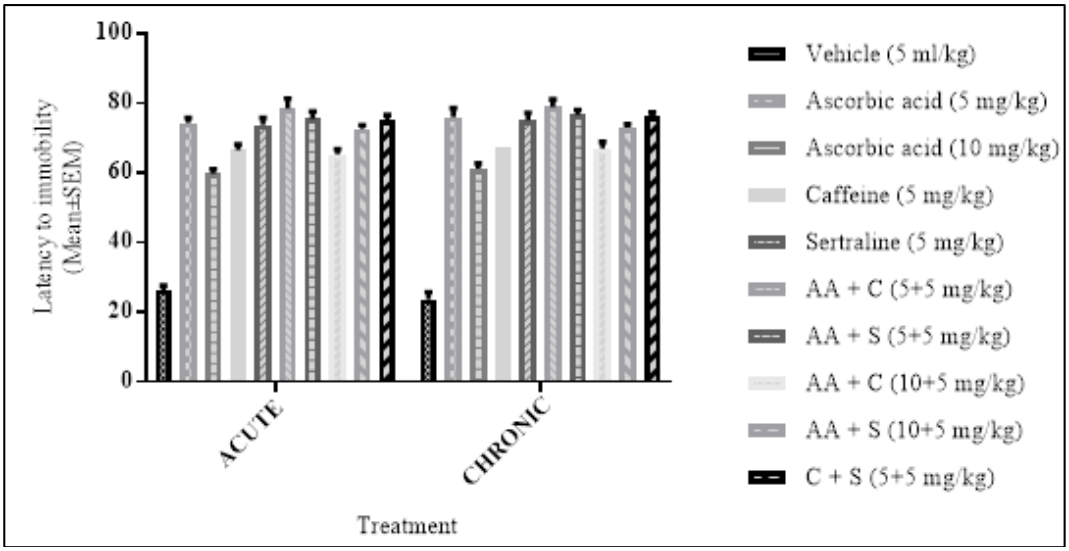

Fig 1: Effect of AA, caffeine, sertraline and their combination on latency to immobility in acute and chronic Forced Swim Test.

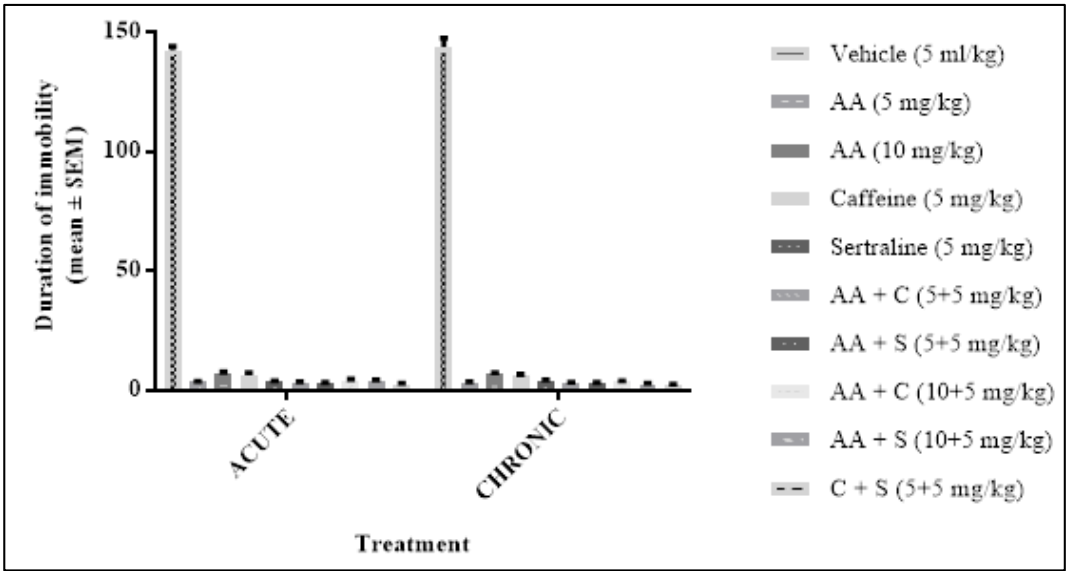

Fig 2: Effect of AA, caffeine, sertraline and their combination in duration of immobility in acute and chronic Forced Swim Test.

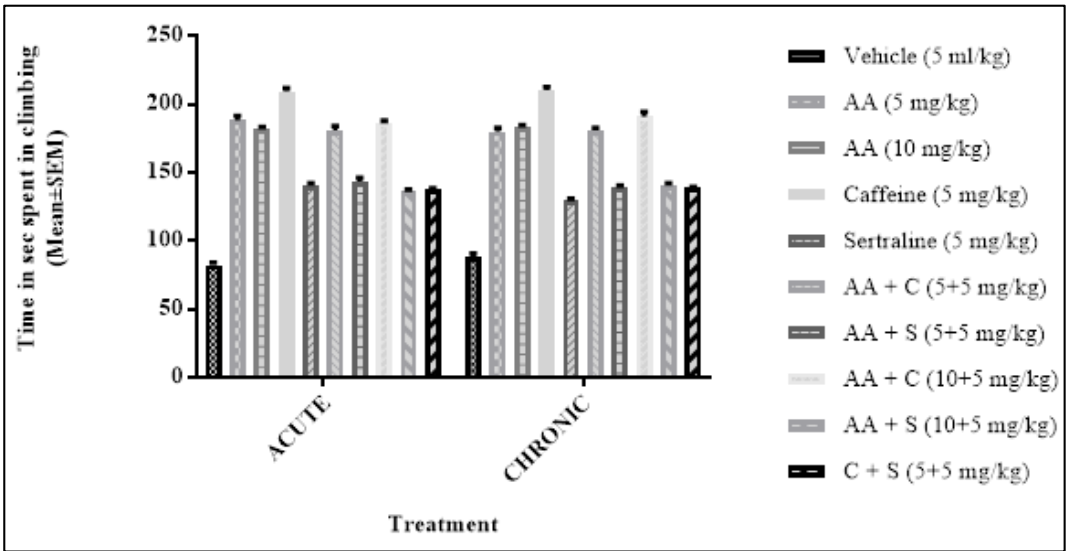

Fig 3: Effect of AA, caffeine, sertraline and their combination in time spent in climbing in acute and chronic Forced Swim Test.

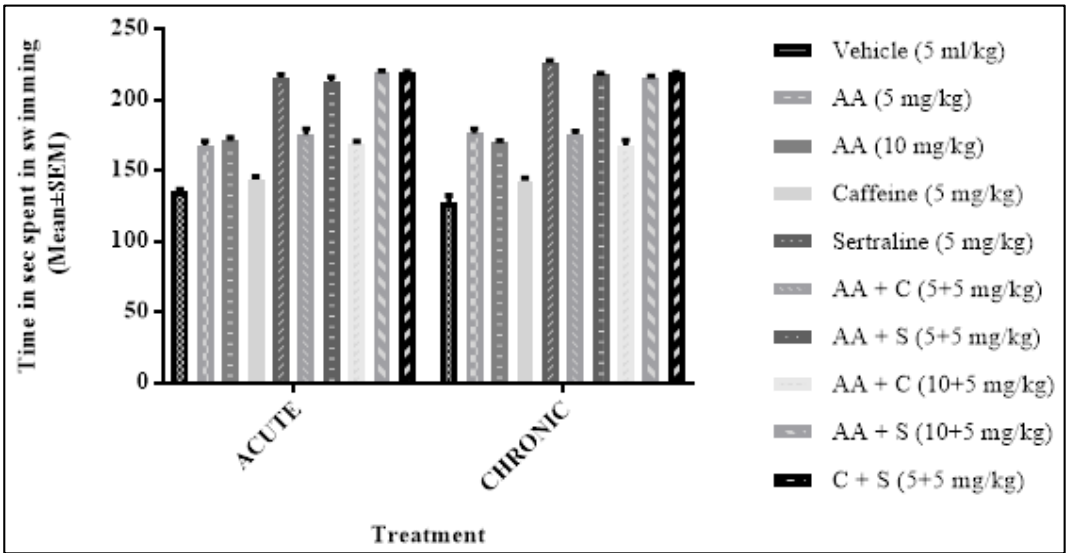

Fig 4: Effect of AA, caffeine, sertraline and their combination in time spent in swimming in acute and chronic Forced Swim Test.

Tail suspension test

Effects of various treatments on latency to immobility, duration of immobility in acute and chronic tail suspension test is given in table 3. Caffeine at 5 mg/kg has shown significant (P<0.01) increased in latency to immobility as shown in Fig. 5 in tail suspension test as compare to sertraline whereas significant (P<0.01) decrease in the duration of immobility was seen in all treatment but marked result is shown in C + S (5+5 mg/kg) treated group as shown in Fig.6.

Table 3: Effect of AA, caffeine, sertraline and their combination in acute and chronic tail suspension test

Treatment	Latency to immobility		Duration of immobility	
	Acute	Chronic	Acute	Chronic
Vehicle (5 ml/kg)	23.4 ± 1.2	19 ± 1.3	163.2 ± 1.06	166.4 ± 5.9
AA (5 mg/kg)	39.25 ± 1.1*	41 ± 1.2*	107 ± 4.3*	90.25 ± 4.6*
AA (10 mg/kg)	66.6 ± 1.37*	70.6 ± 1.07*	88.8 ± 0.86*	83.2 ± 1.28*
C (5 mg/kg)	91 ± 1.2*	94.2 ± 1.06*	83.8 ± 1.15*	81.6 ± 1.07*
S (5 mg/kg)	124.8 ± 1.4*	125 ± 1.14*	75 ± 1.5*	73 ± 1.07*
AA + C (5+5 mg/kg)	93.75 ± 2.2*	66.8 ± 2.2*	79.25 ± 6.9*	76.4 ± 1.8*
AA + S (5+5 mg/kg)	73.8 ± 2.1*	69.8 ± 2.8*	75.8 ± 2.5*	74.6 ± 1.3*
AA+C (10+5 mg/kg)	55 ± 1.58*	54 ± 1.22*	82.6 ± 1.83*	77.8 ± 1.06*
AA+S (10+5 mg/kg)	66.8 ± 0.86*	61 ± 1.16*	70.6 ± 0.8*	64.6 ± 1.72*
C + S (5+5 mg/kg)	104.8 ± 1.28*	106.6 ± 1.2*	23 ± 1.22*	22.2 ± 1.15*

Data presented as mean±SEM (n=5). *indicate significant (P<0.05) difference from respective control group. Data was analysed by one-way ANOVA) followed by Bonferroni test and Dunnett test.

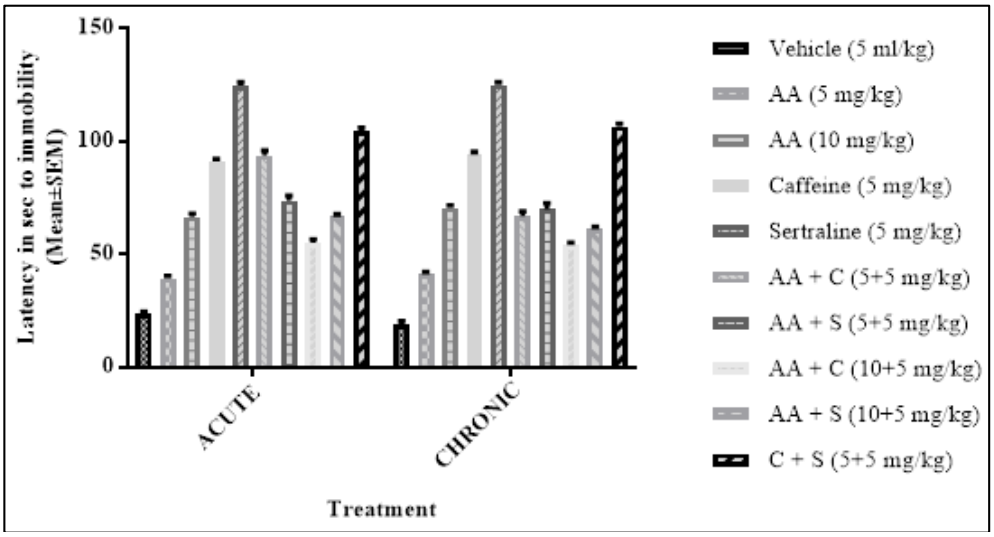

Fig 5: Effect of AA, caffeine, sertraline and their combination on latency to immobility in acute and chronic tail suspension test.

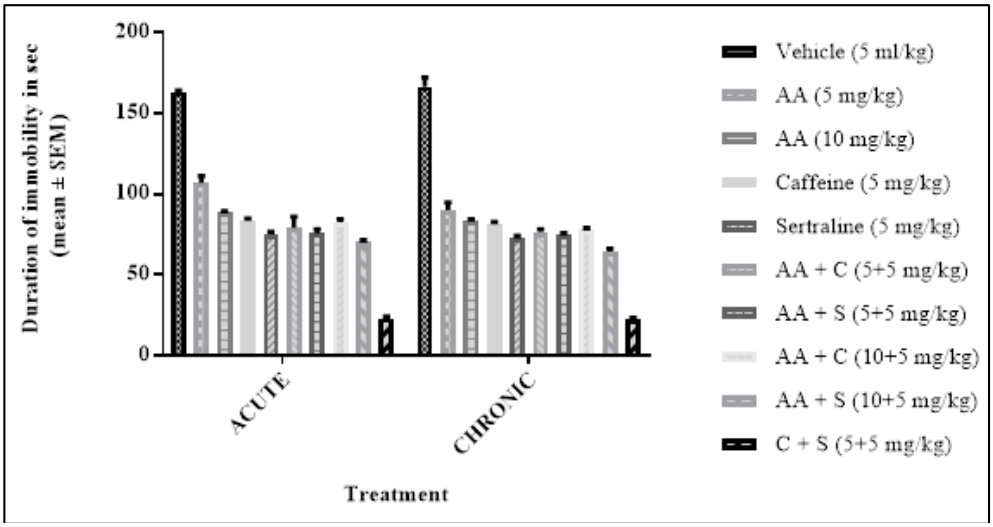

Fig 6: Effect of AA, caffeine, sertraline and their combination on duration of immobility in acute and chronic tail suspension test.

Splash test

Effects of various treatments on latency to grooming, duration of grooming in splash test is given in Fig. 7 and 8 where AA + C (10+5 mg/kg) has shown significant (P<0.01) decreased in latency to grooming whereas significant (P<0.01) increase in the duration of grooming was seen in AA (10mg/kg) and AA + C (10+5 mg/kg) treated groups.

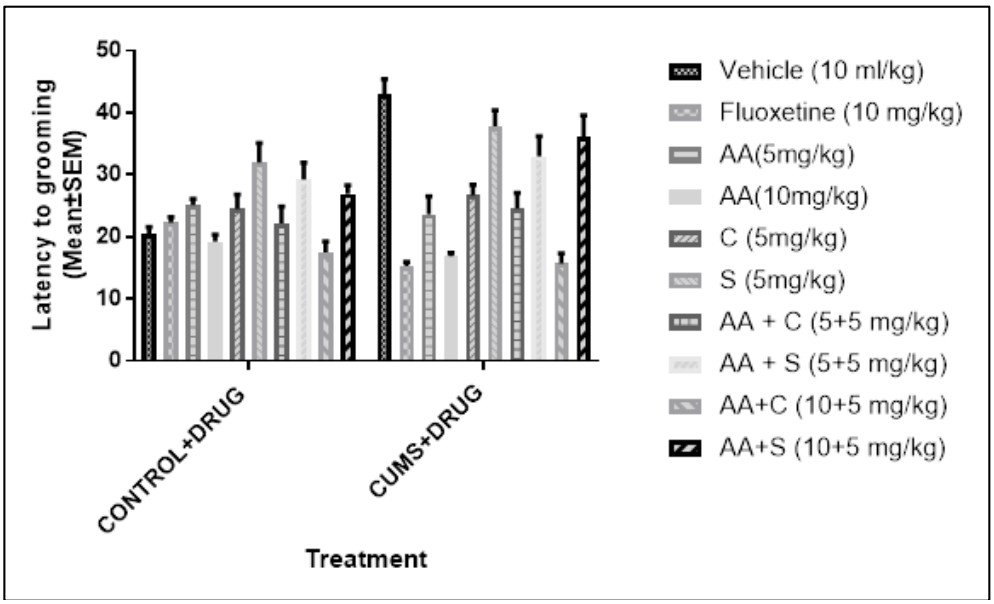

Fig 7: Effect of AA, fluoxetine and its combination on latency of grooming in splash test.

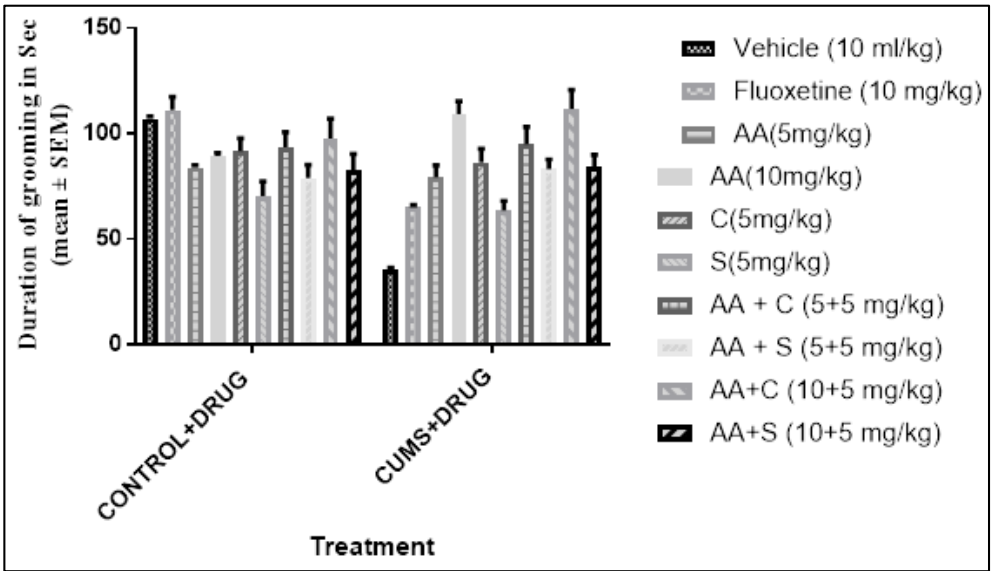

Fig 8: Effect of AA, fluoxetine and its combination on duration of grooming in splash test.

Discussion

Because of their relative simplicity and rapidity compared to other depression-associated procedures, the forced swim test (FST) and tail suspension test (TST) have become widely used in the screening of antidepressant drugs. The FST is also known as the Porsolt test, and involves placing an animal into a beaker of water from which it cannot escape. Both models predispose animals to the state of behavioral despair, which is comparable to human depression [18, 20]. Depressive symptoms manifest due to functional deficiency of noradrenalin, serotonin or dopamine neurotransmitters in the limbic system, prefrontal cortex, hippocampus and amygdale areas of the brain [21]. The major target of antidepressants is therefore to increase the level of these neurotransmitters in the brain and hence reverse the symptoms of depression. Clinically useful antidepressants such as imipramine and fluoxetine reduce the immobility time in both forced swim test and tail suspension test. In FST significant (P<0.01) decrease in the duration of immobility was seen in C + S (5+5 mg/kg) treated group. A significant (P<0.01) increase in time spent in climbing in Caffeine (5 mg/kg) treated group is observed. Sertraline and caffeine combination at (5+5 mg/kg) showed significant increase in time spent in swimming. In TST caffeine at 5 mg/kg has shown significant (P<0.01) increased in latency to immobility in acute tail suspension test which is comparable to sertraline whereas significant (P<0.01) decrease in the duration of immobility was seen in C + S (5+5 mg/kg) treated group. It is possible to demonstrate the usefulness of caffeine and its derivatives in the treatment of depression. It has been shown that caffeine can reverse the monoaminergic system changes observed in depression, for example, caffeine blocking the A1 adenosine receptor subunit may increase the levels of catecholamines and serotonin (5-HT) in the central nervous system (CNS). What seems to be important is the fact that caffeine contributes to an increased 5-HT release in the limbic areas and the release of dopamine (DA) in the prefrontal cortex, which is comparable to the effect obtained with the use of antidepressants [22, 27]. AA + C (10+5 mg/kg) has shown significant (P<0.01) decreased in latency to grooming whereas significant (P<0.01) increase in the duration of grooming was seen in AA (10mg/kg) and AA + C (10+5 mg/kg) treated groups. Chronic unpredictable mild stress (CUMS) has produces depression like behaviour in animal model so splash test model is used [28].

References

1. Murugan U, Suganthi J, Subakanmani S. Evaluation of antidepressant like activity of cucurbita lepo seed extracts in rats, international journal of current pharmaceutical research. 2011; 3(1):108-113.
2. Kale P, Addepalli V, Bafna P, Prabhaalkar K. Caffeine Induced Augmentation of Antidepressant Therapy, Journal of Experimental and Clinical Medicine. 2010; 2(6):282-286.
3. Khanzode SD *et al*. Oxidative damage and major depression: the potential antioxidant action of selective serotonin re-uptake inhibitors. Redox Rep. 2003; 8:365-370.
4. Ozcan ME *et al*. Antioxidant enzyme activities and Oxidative stress in affective disorders. International Clinical Psychopharmacology. 2004; 19:89-95.
5. Moretti M *et al*. Ascorbic acid treatment, similarly to fluoxetine, reverses depressive-like behavior and brain oxidative damage induced by chronic unpredictable stress, Journal of Psychiatric research. 2012; 46:331-340.
6. Griffiths H, Lunac J, Ascorbic acid in 21st century-more than a simple antioxidant, Environmental toxicology and pharmacology. 2001; 10:173-182.
7. Krone C, Ely J. Ascorbic acid, glycation, glycohemoglobin and aging, medical hypotheses. 2004; 62:275-279.
8. Kurhe Y, Radhakrishnan M, Gupta D, Thangaraj D. Effect of A Novel 5-HT_3 Receptor Antagonist 3-Methoxy-N-P-tolylquinoxalin-2- carboxamide (QCM-4) on the acute and chronic rodent models of depression, International Journal of Pharmacy and Pharmaceutical Sciences. 2013; 5(3):567-573.
9. Gan S, Ramli N. Dual effects of low and high dose caffeine, American journal of pharmacy. 2009; 28(3):465-469.
10. Chen S *et al*. Antidepressant-like effect of asiaticoside in mice, Pharmacology, biochemistry and behaviour. 2009; 89:444-449.
11. Porsolt R, Bertin A, Jalfre M. "Behavioural despair" in rats and mice: strain differences and the effects of imipramine, European Journal of Pharmacology. 1978; 51:291-294.
12. Detke MJ *et al*. Acute and chronic antidepressant drug treatment in the rat forced swimming test model of

depression. Experimental and Clinical Psychopharmacology. 1997; 5:107-112.
13. Oriaifo S, omogbai E. Effect of nifidipine, imipramine and sertraline on the antidepressant-like actions of furosemide in forced swim (FST) and tail suspension (TST) tests models of depression in mice, African journal of biotechnology. 2012; 11(52):11524-11529.
14. Steru L, Chermat R, Thierry B, Simon P. The tail suspension test: A new method for screening antidepressants in mice, Psychopharmacology. 1985; 85(3):367-370.
15. Isingrini E, Camus V, Guisquet AM, Pingaud M, Devers S, Belzung C *et al*. Association between Repeated Unpredictable Chronic Mild Stress (UCMS) Procedures with a High Fat Diet: A Model of Fluoxetine Resistance in Mice, PLoS ONE. 2010; 5(4):10404.
16. Bernadeta Szewczyka, Bartłomiej Pochwata, Bożena Muszyńskab, Włodzimierz Opokac, Agata Krakowskac, Anna Rafało-Ulińskaa *et al*. Antidepressant-like activity of hyperforin and changes in BDNF and zinc levels in mice exposed to chronic unpredictable mild stress. Behavioural Brain Research. 2019; 372:112045.
17. Isingrini E, Camus V, Guisquet A M, Pingaud M, Devers S, Belzung C *et al*. Association between Repeated Unpredictable Chronic Mild Stress (UCMS) Procedures with a High Fat Diet: A Model of Fluoxetine Resistance in Mice, PLoS ONE. 2010; 5(4):10404.
18. Dhingra D, Valecha R. Evidence for involvement of the monoaminergic system in antidepressant-like activity of an ethanol extract of Boerhaavia diffusa and its isolated constituent, punarnavine, in mice. Pharm Biol. 2014; 52:767-774, doi: 10.3109/13880209.2013.870583.
19. Teixeira CP, de Melo CT, de Araujo FL, de Carvalho AM, Silva MI, Barbosa-Filho JM *et al*. Antidepressant-like effect of riparin II from *Aniba riparia* in mice: evidence for the involvement of the monoaminergic system. Fundam Clin Pharmacol. 2013; 27:129-137. doi: 10.1111/j.1472-8206.2011.00973.x.
20. Bhattacharya SK, Bhattacharya A, Sairam K, Ghosal S. Anxiolytic-antidepressant activity of Withania somnifera glycowithanolides: an experimental study. Phytomedicine. 2000; 7:463-469. doi: 10.1016/S0944-7113(00)80030-6.
21. Chen PJ, Hsieh CL, Su KP, Hou YC, Chiang HM, Sheen LY *et al*. Rhizomes of *Gastrodia elata* B(L) possess antidepressant like effect via monoamine modulation in subchronic animal model. Am J Chin Med. 2009; 37:1113-1124, doi: 10.1142/ S0192415X09007533.
22. Acquas E, Tanda G, Di CG. Differential effects of caffeine on dopamine and acetylcholine transmission in brain areas of drug-naive and caffeine-pretreated rats. Neuropsychopharmacology. 2002; 27:182-193.
23. Barone JJ, Roberts HR. Caffeine consumption. Food Chem Toxicol. 1996; 34:119-129.
24. Benowitz NL. Clinical pharmacology of caffeine. Annu Rev Med. 1990; 41:277-288.
25. Fernstrom MH, Bazil CW, Fernstrom JD. Caffeine injection raises brain tryptophan level, but does not stimulate the rate of serotonin synthesis in rat brain. Life Sci. 1984; 35:1241-1247.
26. Fisone G, Borgkvist A, Usiello A. Caffeine as a psychomotor stimulant: mechanism of action. Cell Mol Life Sci. 2004; 61:857-872.
27. Fredholm BB, Bättig K, Holmén J, Nehlig A, Zvartau EE. Actions of caffeine in the brain with special reference to factors that contribute to its widespread use. Pharmacol Rev. 1999; 51:83-133.
28. Moretti M, Freitas AE, Budni J, Fernandes SC, Balen Gde O, Rodrigues AL *et al*. Involvement of nitric oxide-cGMP pathway in the antidepressant-like effect of ascorbic acid in the tail suspension test. Behav Brain Res. 2011; 225:328-33.