



SLEEP DEPRIVED BEHAVIOR OF MALE AND FEMALE RATS VIA SWIM TEST

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ABSTRACT

Key Words

Sleep deprivation,
Swimming test, *Elletaria
cadamomum*, and
Imipramine



In this article the depression is forcibly produced by sleep deprivation method and the analysis of the depression behavior is done by swimming behavior of rats. More on a recent paper suggested that females are more prone to depression than males. To resolve this *Elletaria Cardamomum* was used to analyze the effect of depression via sleep deprivation method. The aim of study was undertaken to evaluate the antidepressant effect of crude methanolic extract of seed of *Elletaria cardamom* at doses 200mg/kg and 400mg/kg using sleep deprivation test (imipramine (20mg/kg, body weight) was used as standard by taking the male and female rats for study. Effect of *Elletaria cardamom* dose (200mg/kg and 400mg/kg) were determined, the effect of dose were compared with control and standard group of animals. Data expressed as mean \pm SEM (n=6) $p < 0.05$, $*p < 0.01$, $*p < 0.03$, $*p < 0.04$. Significant dose dependent decline in immobility time was observed in doses by swim test and it was observed, that female are more prone to depression elicited and *Elletaria Cardamom* extract exhibited effectual results.

INTRODUCTION:

Depression is not just a form of extreme sadness. It is a disorder that affects both brain and body, including cognition, behavior, the immune system and peripheral nervous system. Unlike a passing sad mood, depression is considered a disorder because it interferes with ordinary functioning in work, school, or relationships. Depression is a heterogeneous mood disorder that has been classified and treated in variety of ways. As we know that the numbers of synthetic drugs are being used as standard treatment for clinically depressed patient. Synthetic drugs have adverse effects that can compromise the therapeutic treatment.

Sleep deprivation play important role in causing depression. Although it is not a widely used technique, it causes the depression at a very large scale.

Unlike normal grief, which comes in waves, it is constant and oppressive.¹ Depression is an affective (not natural) very common and serious disorder. It is also called mania depression syndrome, It can occur at any age, but the highest rate is in the age group of 25-45 years. The incidence of depression is about 2-3 times more common in females than in males.

Type-1. Manic Depression- It is characterized by conflicting behavior, like-enthusiasm, rapid thoughts, disturbed speech pattern and extreme self-confidence with impaired judgment. The manic

depression is further of two types- unipolar depression (up and down mood) and bipolar depression (cyclization of mood).

Type-2.Endogenous Depression- It can be classified into four categories- true depression (delusion, sleep disorder), neurotic (depression with anxiety and fear), obsessive (suicidal tendencies), involuntional depression (helplessness and unhappy emotional state).² Depression is a heterogeneous disorder that affects a person's mood, physical health and behavior. Anti-depressant drugs such as tricyclic anti-depressant and selective serotonin re-uptake inhibitor (SSRI) are used to treat depression showing various side effects and thus, the search for a new anti-depressant without side effects is deemed important.³ Major depressive disorder (MDD) is commonly referred as depression. That is characterized by sad mood, loss of interest, unhappiness, change of appetite, somatic complaints (e.g., aches and pains), psychomotor changes (e.g., agitation), decreased energy and tiredness, a sense of worthlessness or guilt. Impaired concentration, suicidal ideation and cognitive deficits Moreover, depression is the most common of the affective disorders; it may vary from very mild condition, bordering on normality, to severe psychotic depression accompanied by hallucinations and delusions. Depression is a state of squat mood and dislike to activity or apathy that can affect a person's thoughts, feelings, behavior and sense of well-being. It is a very serious and disabling psychiatric condition that occasionally leads to suicide or premature death due to unattended physical problems. The association between depression and increased risk of death and morbidity is an obvious indicator of the severity of the condition, which is usually clear when reflecting upon the WHO data on life expectancy and the causes of death. The World Health Organization estimated that by 2020 unipolar major depression were come the second largest cause of global disease problems in the world, only behind

ischemic heart disease.⁴ Because the mechanism of depression is quite complex, many currently available synthetic chemical antidepressants have low rates of response and remission and even severe adverse effects.⁵ The majority of patients are often reluctant to take synthetic antidepressants in their appropriate doses due to their anticipated side effects including inability to drive a car, dry mouth, constipation, sexual dysfunction and so on. Therefore, finding more effective and less toxic agents is a serious and urgent problem. Furthermore, natural plants may be some of the most attractive sources of new drugs with lower side effects than those of synthetic antidepressants.⁶

Depression and Depressive Disorder-

Major depressive disorder is described as a mixture of symptoms (syndrome) that affect a person's capability to work, sleep, study, eat, and enjoy once pleasurable activities. Depressive disorder should not be confused with depression. Depression refers to suffering from depressive symptoms but it is not depressive disorder unless it meets the Diagnostic and Statistical Manual of Mental Disorders, 4th Edition text revision (DSM-IV-TR) criteria. According to DSM-IV-TR, there are two main depressive symptoms that is, depressed mood and anhedonia or loss of interest or pleasure in most or all behavior. Either of them must be present to make a diagnosis of major depressive episode. The other symptoms are sleeplessness, change in appetite or weight, psychomotor retardation or anxiety, low energy, poor concentration, thoughts of worthlessness or guilt and recurrent thoughts about death or suicide. Some people may experience depressive disorder only once within their lifetime, but sometimes a person may experience it more than once. Depression is a significant contributor to the global burden of disease and affects people in all communities across the world. Today, depression is estimated to affect 350

million people worldwide. The World Mental Health study conducted in 17 countries found that a standard of 1 in 20 people reported having an occurrence of depression in the previous year. Pursuant to this, depression is the leading cause of disability for both males and females, with the burden of depression being 50% higher for females than males, making it the leading cause of disease burden in women worldwide.⁷ Some of the neurological disorder such as epilepsy affects the 5% of the world population and more deteriorate the condition with the fact that 30% of the patients continue to have seizures with current antiepileptic drugs therapy more over recently with the advance lifestyle the behavioral disorders like anxiety and depression intensify by several folds.⁸ The clinical depression is going to become the leading cause of disability worldwide, therefore there is need to develop certain medicine to control and treat the depression. Herbal drugs are safer and economical that's by the present research work were designed and carried out on herbs in depressed model of rats.

Methodology:

Data were obtained from laboratory based studied by using experimental rats of male and female rats weighing between 100-150 g, maintained at room temperature having free access to food and tap water. All the study was performed in accordance with the institutional animal ethics committee as per the guidelines laid by CPCSEA.¹⁴ Chemicals, reagents and glass wares were procured from university lab. Extract of *Elettaria cardamomum* seed were carried out using standard procedure. *Sleep deprivation* were used for interactive study.

Plant material and extraction:

Dry *Elettaria cardamomum* seed were purchased from the market. The plant seed were identified by botanist at M.J.P. Rohilkhand University. A voucher specimen of the plant material was deposited at the herbarium of botany department. The dried seed of plant (100g)

were grinded into fine powder using an electric grinder and extracted by percolation method and through using methanol (80%) for 72 h at room temperature. The solvent were removed in a rotary evaporator, and after filtering, the extracts were concentrated to dryness.¹⁴

Apparatus:

Sleep has vital homeostatic functions, and sleep deprivation is a stressor that has consequences for the brain as well as for many body systems. Although sleep deprivation is not yet a well-established model of depression, many studies show that it alters important pathways related to stress. Increased levels of messenger RNA for interleukin-1b (a pro-inflammatory cytokine) and for cortisol have been shown in rodents after sleep deprivation. The method of this study consisted of handling the animals gently to prevent them from sleeping. Furthermore, 120 hours of sleep deprivation in mice was induced using the platform method, which is proficient by placing the animal on a platform submerged in water so that, when the animal falls asleep, it falls into the water and must then climb back onto the platform, thus forcing it to stay awake. Briefly platform technique consists of a small platform (3 cm) in a water maze (41 × 34 × 16.5 cm) filled with water. The height of water was kept 1 cm below the platform and bright light was provided whole the night. Animals were subjected to 5 days sleep deprivation protocol by keeping them on small platform. Animal are able to move from one platform to other by jumping. A 100-W light was used to light up the chamber during the sleep deprivation period. The presumption of this method is that sleep and drowsiness induce muscle relaxation and animal will fall in water, and after falling in water animals will wake up quickly. A glass jar is used to analyzing the depressive behavior which is induced by sleep deprivation.¹⁵

Experimental Protocol:

The animals were housed for 1 week in a laboratory room for acclimatization they were grouped into four, containing six animals each group. Rat were divided into 4 groups (n=6).

- **Group I** - served as control (vehicle only).
- **Group II** - animals treated orally with low dose of *Elettaria cardamomum* seed extract.
- **Group III** - animal treated orally with high dose of *Elettaria cardamomum* seed extract.
- **Group IV** - animals treated with imipramine at a dose of 20mg/kg p.o.

The rats were divided into four groups and named or tagged properly. The first group were controlled group were treated with normal saline as vehicle, the second group were test 1 group were treated with 200mg/kg of body weight. The third group was test 2 group were treating with 400mg/kg of body weight. The fourth group was the standard group was treated with imipramine 20mg/kg p.o.¹⁴

- **(day-0):** if animals are ordered from a supplier, they arrive and are re housed in single cages or into groups of 2-4; leave them for at least 7 days to habituate to the conditions.
- **(day-7):** perform the sleep deprivation test for 120 hours.
- **(days-12 - 19):** inject the dose to the animals for 7 days of different group.
- **(day-20):** code the depressive behavior of rats by swim test.

Behavioral Coding:

An animal were judged to be immobile whenever it remains floating passively in the water in a slightly hunched but upright position, its nose just above the surface, with no additional activity other than that necessary to keep its head above water. Swimming is defined as active movement throughout the swim chamber, which includes crossing into another quadrant. Climbing activity (also termed thrashing) consist of upward directed movements of the forepaws along the side

of the swim chamber. The data obtained were compared between control, standard and test.¹⁶

RESULT:

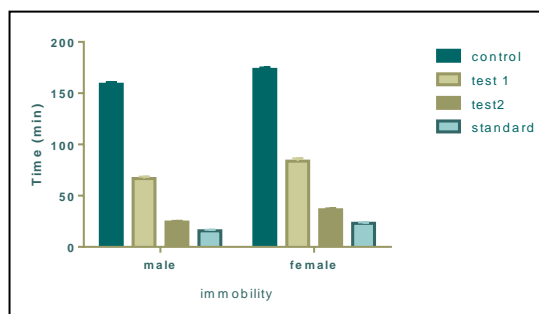
Elletaria cardamom (200 and 400mg/kg) significantly (p<0.03) and dose dependently decreased the immobility time as compared to control rats (table -1). The extent of the dose of 400mg/kg showed the almost same activity as imipramine, in decreasing immobility period. Thus, the activity of *Elletaria cardamom* could involve one of the mechanisms for the established agents as described above.

Result of anti-depressive activity of methanolic extract of *Elletaria cardamom* on sleep deprivation via swim test.

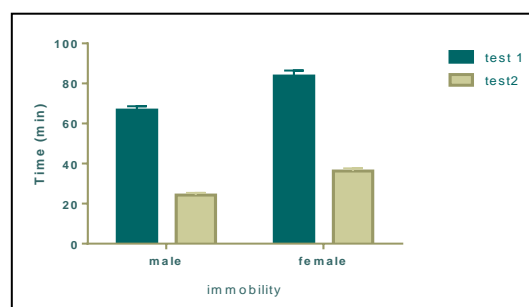
Treatment	mg/kg p.o. (n=6)	Immobility(time)	
		Male rats	Female rats
Vehicle	20	158.8±2.01	173.3±1.69
Extract Test-1	200	66.6±1.92	83.6±2.81
Extract Test - 2	400	24.33±1.01	36.3±1.32
Imipramine	20	15.6±1.09	23.0±0.88

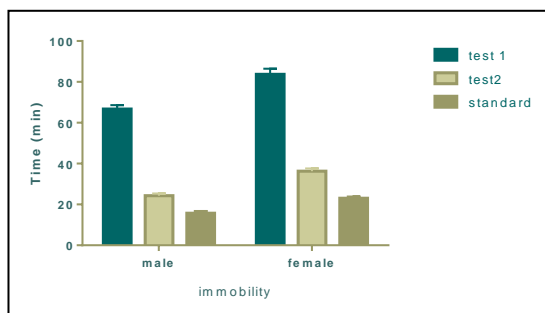
Values represent the group mean±SEM, (n=6), p<0.05vs.control

(A)



(B)





(C)

Fig.1: Effect of *Elletaria cardamom* dose (200mg/kg and 400mg/kg) were determined, the effect of dose were compared with control and standard group of animals. Data expressed as mean \pm SEM (n=6) $p < 0.05$, * $p < 0.01$, ** $p < 0.03$, * $p < 0.04$.

Future Aspect:

Although current anti depressant offers both short and long-term benefits, important problems persist, such as tolerance, delayed therapeutics onset, limited efficacy of common treatment in milder and resistant depression.¹⁷ Natural products such as plants crude extracts or their purified compounds provide unlimited opportunities for new drug discoveries because of the unmatched availability of chemical diversity.¹⁸ According to World Health Organization (WHO) more than 80% of the world population relies on the traditional medicine for their primary healthcare needs. Recently, study in the area of herbal psychopharmacology has demonstrated a variety of promising medicines that can provide benefit in the treatment of stress and anxiety disorders. Considering advantageous wide rang flavonoid effects such as cognitive, anxiolytic, anti-depressants and sedative effect and also presence of a large scale of flavonoids and linalool including quercetin in cardamom, we assessed *Elletaria cardamomum* methanolic extract effects on depression-like behavior in rat model.¹¹ for these reasons, patients need new therapeutics alternatives that could be provided by medicinal plant and natural products.

Discussion and Conclusion: As we know monoamines are neurotransmitters that include serotonin, dopamine, norepinephrine and epinephrine and monoamines are the chemicals which are responsible for depression whereas the cardamom contains many chemical constituents in which cineole and linalool are the main chemicals which are present in cardamom as an active constituents,

the linalool produce an antidepressant like effect through interaction with monoaminergic system including serotonergic and noradrenergic system. On the basis of this mechanism the rats were show the reduction in depression by reducing the immobility during the swim test and on the basis of this activity cardamom were knows as a good antidepressant spices. From the above valuable animal study, we conclude that plant extract *Elletaria cardamom* show a significant antidepressant activity in sleep deprivation method via swim test and it is also clear that female rats were more prone to depression rather than male rats.

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