Journal of Pharmaceutical and Scientific Innovation

www.jpsionline.com

Research Article

WHERE THERE IS A TIL, THERE IS A WILL

Parle Milind* and Dhamija Isha

Pharmacology Division, Dept. Pharm. Sciences, Guru Jambheshwar University of Science and Technology, Hisar, Haryana (India)

Received on: 15/12/12 Revised on: 20/02/13 Accepted on: 25/02/13

ABSTRACT

Til is a key ingredient in a variety of world cuisines, especially Asian, Middle Eastern and Mediterranean for its nutritional and medicinal value. Sesame seeds (Til) seem like tiny and simple pieces of nature's creation. The present study was undertaken to investigate the anxiolytic potential of *Sesamum indicum* in mice. A total of 150 Swiss mice, divided into 25 groups were employed in this study. *Sesamum indicum* (4%, 8% and 16% w/w) was admixed in diet of mice for a period of 14 days. Elevated plus maze, Light-dark model and Hole board test were used as behavioral models in this study. Effect of *Sesamum indicum* on MDA levels was also estimated. *Sesamum indicum* produced significant ($P<0.01$) increase in the time spent in the lit compartment in light-dark model. *Sesamum indicum* enhanced significantly ($P<0.05$) the number of entries and time spent in open arms, when tested using elevated plus maze model in mice. The hole-board test provides a simple method for measuring the response of an animal to an unfamiliar environment and is widely used to assess anxiety. *Sesamum indicum* significantly ($P<0.05$) increased head dip counts at different concentrations (4%, 8% and 16% w/w), indicating its anxiolytic effect. *Sesamum indicum* significantly ($P<0.01$) reduced MDA levels in the brains of mice, thereby revealing its property of bringing about reduction in free radical generation. These findings reflect the anxiolytic potential of *Sesamum indicum.*

KEYWORDS: Til, Anxiety, Hole Board, *Sesamum indicum*

INTRODUCTION

Anxiety is an emotion experienced as a part of everyday life and can be viewed as continuum. Mild anxiety can improve motivation and productivity, while intense anxiety with the 'fight or flight' response promotes survival in response to danger. Anxiety becomes pathological, when it occurs to an excessive degree and disrupts day-to-day functioning. Anxiety disorder is one of the most common psychiatric illnesses[1]. More than 20% of the adult population suffers from anxiety at some time during their life[2]. Anxiety is an aversive emotional state, in which the feeling of fear is disproportionate to the nature of the threat. It is characterized by feelings of apprehension, insecurity, uncertainty or fear[3]. It may interfere with intelligence, psychomotor function and memory. The principal regulatory systems involved in anxiety are GABA-ergic and serotoninergic systems. However, recent findings revealed that oxidative stress also acts as a plausible pathway for anxiety[4, 5]. Benzodiazepines are the most frequently prescribed synthetic drugs for anxiety. But these psycho-neural drugs have very serious side effects like deterioration of cognitive function, physical dependence and tolerance[6]. In this context, the focus has been recently shifted to developing medicines from natural sources in the hope, that drugs of plant origin will have significantly less side effects than synthetic drugs, while having comparable efficacy. Various types of herbs like *Ducrosia anethifolia,* leaves of *Ocimum sanctum*, *Azadirachta indica*, are known to have anxiolytic effects[7]. Sesame seeds have been used as a medicine since antiquity. They are considered to have antioxidant, anticancer, diuretic, antihyperlipidemic, and immunomodulator activity[8]. Niacin abundantly found in Til helps in reducing LDL in blood and enhances GABA-ergic activity inside the brain, which in turn might help in reducing aggression and neurosis. Therefore, this study was undertaken with an objective to investigate anti-anxiety potential of *Sesamum indicum*.

MATERIALS AND METHODS

Plant material

The *Sesamum indicum* seeds were purchased from local market of Hisar and got authenticated from Raw Materials Herbarium and Museum, National Institute of Science Communication and Information Resources (NISCAIR), New Delhi (Ref. NISCAIR/RHMD/Consult/-2011-12/1724/264).

Animals: A total of 150 Swiss mice weighing around 20-25g divided in 25 groups were employed in the present study. Each group comprised of six animals. Mice were procured from disease free animal house of Lala Lajpat Rai University of Veterinary and Animal Sciences, Hisar (Haryana, India). The animals had free access to food and water, and they were housed in a natural (12h each) light-dark cycle. The animals were acclimatized for at least 5 days to the laboratory conditions before behavioral experiments. The experimental protocol was approved by the Institutional Animals Ethics Committee and the care of laboratory animals was taken as per the guidelines of CPCSEA, Ministry of Forests and Environment, Government of India (Registration number - 0436).

Drug protocol: Mice in group I were employed for pilot study carried out to determine the effective concentrations of *Sesamum indicum* (SI). Mice belonging to group II to VI were subjected to Light-Dark Model. Mice belonging to group VII to XI were subjected to Elevated Plus Maze Model, while group XII to XVI were employed in Hole Board test. Mice belonging to group XVII to XX were used in estimating MDA levels. Mice belonging to group XXI to XXV were exposed to photoactometer for assessing the locomotor activity. Diazepam (1 mg/kg) and *Sesamum indicum* (4%, 8% and 16% w/w) were admixed in diet for 14 successive days to mice. Biochemical studies were carried on 14th day after drugs/ SI administration. Effect on locomotor activity of mice was studied using photoactometer.

EXPERIMENTAL DESIGN: The Elevated Plus-Maze test has been widely validated for measuring anxiolytic and anxiogenic-activities in mice. The plus maze apparatus

consisted of two open arms, 16 x 5 cm^2, and two closed arms, 16 x 5 x 12 cm^3, connected to a central platform 5 x 5 cm^2. The maze was elevated to a height of 25 cm above the floor. Each mouse was placed individually at the centre of elevated plus maze with its head facing towards an open arm and was observed for 5 min to record the number of entries into open arm, closed arm and time spent in each arm[9].

Light-Dark model : Light-dark exploration test is commonly employed for evaluation of anxiolytic activity. The apparatus consisted of a rectangular box (45 x 27 x27 cm^3), partitioned into two compartments connected by a 7.5 x 7.5 cm^2 opening in the wall between compartments. An animal was placed into the centre of the lit compartment and was observed for 10 min for time spent in this compartment[10].

Hole Board Test : The Hole-board apparatus consisted of a wooden box (40 x 40 x 25 cm^3) with 16 holes (each of diameter 3 cm) evenly distributed on the floor. The numbers of head dippings were counted by placing the animal in the center of the apparatus[11] for a period of 5 min.

Locomotor activity: Locomotor activity was measured using photoactometer.

Collection of Brain samples

The animals were sacrificed by cervical decapitation under light anesthesia on the 14th day, 90 min. after administration of the diet admixed with SI or standard drugs. Immediately after decapitation, brain was carefully removed from the skull. For preparation of homogenate, the whole brain was weighed and transferred to a glass homogenizer and homogenized in an ice bath after adding 10 volumes of 0.9% sodium chloride solution. The homogenate was centrifuged at 3000 rpm for 10 min and the resultant cloudy supernatant liquid was used for estimation of brain Malondialdehyde (MDA) levels[12].

STATISTICAL ANALYSIS

All the results were expressed as mean ± standard error (S.E.M.). Data was analyzed using one-way ANOVA followed by Dunnett's *t*-test. *p*-values < 0.05 were considered as statistically significant.

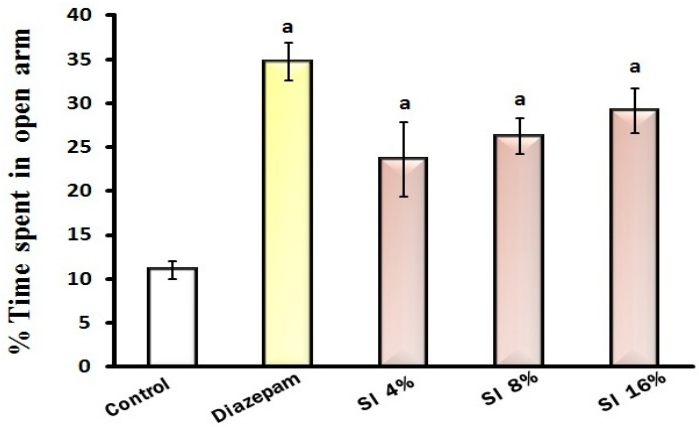

Fig. 1: Effect of *Sesamum indicum* on time spent in open arm by mice using Elevated plus maze model.
Values are expressed in mean ± S.E.M (n=6).
'a' denotes p<0.01 as compared to control group,
SI: *Sesamum indicum* (4, 8, 16% w/w)

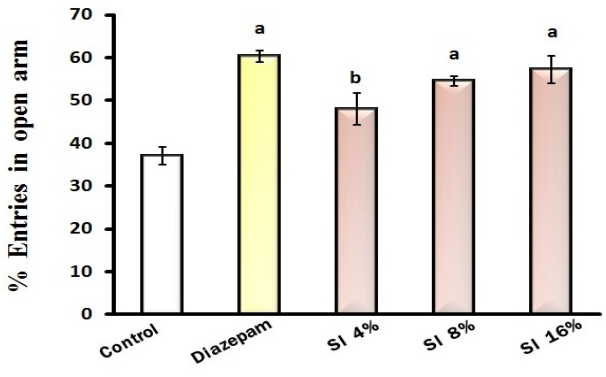

Fig. 2: Effect of *Sesamum indicum* on number of entries of mice in open arm using Elevated plus maze model.
'a' denotes p<0.01 as compared to control group
'b' denotes p<0.05 as compared to control group.
SI: *Sesamum indicum* (4, 8, 16% w/w)
Diazepam (1mg/kg, i.p.) was used as a standard drug

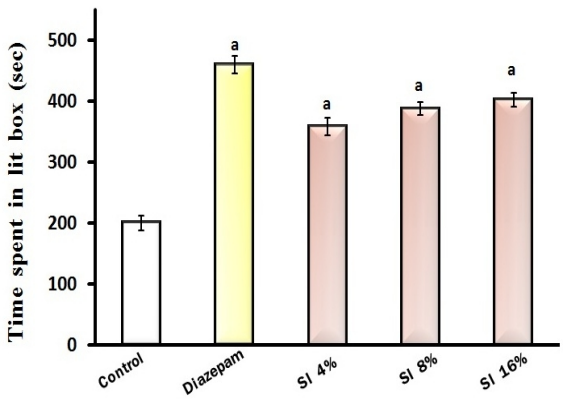

Fig. 3: Effect of *Sesamum indicum* on time spent in lit box of mice using Light-Dark model.
'a' denotes p<0.01 as compared to control group
SI: *Sesamum indicum* (4, 8, 16% w/w)

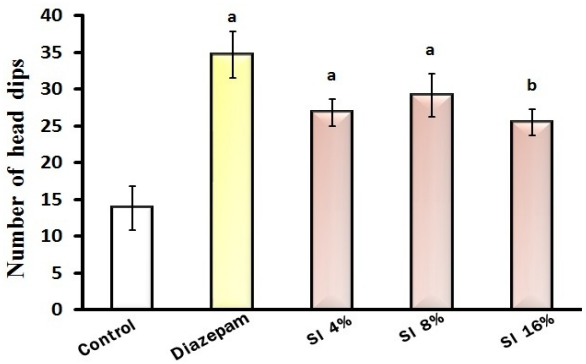

Fig. 4: Effect of *Sesamum indicum* on number of head dips by mice using Hole Board test.
'a' denotes p<0.01 as compared to control group
'b' denotes p<0.05 as compared to control group.
Diazepam (1mg/kg, i.p.) was used as a standard drug

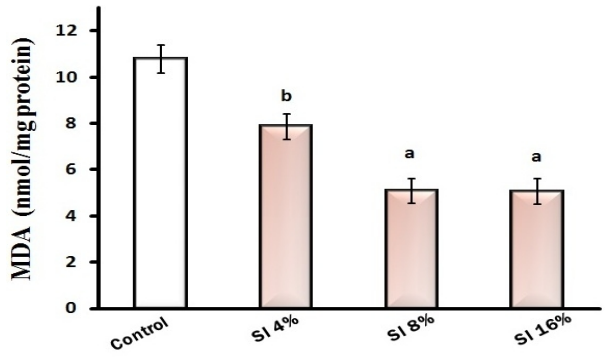

Fig. 5: Effect of *Sesamum indicum* on brain Malondialdehyde levels in mice.
'a' denotes p<0.01 as compared to control group
'b' denotes p<0.05 as compared to control group

Fig. 6: Sesame rolls

Fig. 7: Sesame brittle / Tilgul

RESULTS

Anxiolytic effect of *Sesamum indicum* using Elevated plus maze

Sesamum indicum (SI) at the concentrations of 4%, 8% and 16% w/w, when administered along with diet for 14 successive days, significantly ($p<0.01$) increased the time spent in open arm and enhanced the number of entries ($p<0.01$) in open arm of the Elevated plus maze. The effect of SI was found to be comparable to that of Diazepam (an established anxiolytic agent). (Fig. 1&2)

Anxiolytic effect of *Sesamum indi*cum using Light-Dark model

SI at the concentrations of 4%, 8% and 16% w/w increased the time spent in lit box significantly ($p< 0.01$), in Light-Dark model. The effect of SI was found to be comparable to that of Diazepam. (Fig. 3)

Anxiolytic effect of *Sesamum indicum* using Hole Board test

SI when administered with diet in different concentrations (4%, 8% and 16% w/w), produced a significant ($p<0.05$) increase in head dipping behavior of mice, when tested using hole board test. (Fig. 4)

Effect of *Sesamum indicum* on brain MDA level

The administration of SI for 14 days in mice produced a significant ($p<0.05$) fall in MDA levels, when compared to control group at all the concentrations (4%, 8% and 16% w/w). (Fig. 5)

DISCUSSION

Anxiety may be regarded as a particular form of behavioral inhibition that occurs as a result of exposure to a new situation. Anxiety is a mental disorder resulting from alterations in the levels of certain neuro-chemicals such as GABA, Serotonin, and Dopamine. The common targets for the treatment of anxiety are GABA and Serotonin. Though several medicines are available for the management of anxiety disorder, most of them are associated with some or the other limitation. Benzodiazepines are the most commonly prescribed medicines for the management of anxiety disorder, which act through GABAergic system. However, their consumption is associated with problems of sedation and dependence, while SSRIs like prozac evoke adverse effects like nervousness, sedation, sexual dysfunction, stomach upset and dizziness. In the present study, anti-anxiety potential of Til (*Sesamum indicum)* was tested in mice employing three experimental models viz. Light-Dark model, Elevated plus maze and Hole Board test. Chronic administration of *Sesamum indicum* (SI) for 14 successive days showed anti-anxiety potential in mice, as observed by increased time spent in the lit box using Light-Dark model; enhanced duration and increased entries in the open arm of Elevated plus maze and increased number of head dips in hole board test. Increased head dipping behavior of mice reflects anxiolytic activity, when tested using hole-board test[13]. The light-dark test is based on the innate aversion of rodents to brightly illuminated areas and on the spontaneous exploratory behavior in response to mild stressors. The EPM is considered to be an etiologically valid animal model of anxiety because it uses natural stimuli, such as a fear of height and brightly-lit open space[14]. The present study showed that Sesame seeds possessed potent anxiolytic activity, which was revealed using three different

experimental models of anxiety.

Plants containing lignans and antioxidants are reported to have anxiolytic effect. Plants such as *Fructus schisandrae* are found to have anti-anxiety activity due to the presence of lignans and antioxidants schisandra[15]. Sesame is reported to contain lignans, such as sesamol, sesamin, sesamolin, and sesaminol. Sesamol, is a constituent of Til (sesame seeds) and exhibits antioxidant property. Thus, antioxidant effect of sesamol may help in protecting the experimental animals against oxidative damage. Along with GABAergic and serotoninergic systems, oxidative stress also plays a principal role in pathogenesis of anxiety. When the production of reactive oxygen species (ROS) prevails over the brain defense systems, the lipid-rich constitution of brain may favor lipid peroxidation that may result in decreased membrane fluidity and damage to membrane proteins inactivating receptors, enzymes and ion channels, often to the extent of disrupting membrane integrity and eventually in cell death. Furthermore, oxidation of other sensitive components such as nucleic acids and neurotransmitters can also occur. Thus, oxidative stress can alter neurotransmission, neuronal function and overall brain activity[16]. Oxidative stress (OS) represents a loss of balance in oxidation-reduction reactions (redox state). It is characterized by the reduced ability of the anti-oxidant defense system, eliciting the toxicity of free radicals and its detrimental effects. Recent studies have demonstrated a close relationship between oxidative stress and anxiety in both human patients suffering from anxiety disorder and animals displaying high trait anxiety[16, 17]. At physiologic conditions, antioxidants play a crucial role in maintaining redox homeostasis by regulating the level of ROS, so as not to interfere with the optimal cellular functioning. Thus, the excess of ROS is neutralized by anti-oxidants avoiding the oxidation of cellular components and consequently their damage. Exogenous antioxidants supplement the anti-oxidative action of endogenous antioxidants by acting synergistically. The principal source of exogenous anti-oxidants is our diet. Vitamin E, vitamin C, carotenoids, zinc, selenium, and polyphenols constitute the principal dietary anti-oxidants present in food. Til serves as a good source of vitamin E, zinc, selenium and polyphenol, when supplemented with diet. The effect of dietary antioxidants on the central nervous system has gained interest in the last decade. Antioxidants may constitute a potential treatment when oxidative stress is the causal factor of anxiety. Phytochemically, sesame seeds are a good source of manganese, calcium, iron, phosphorous, zinc, vitamin B_1, B_6, niacin, trypthophan and dietary fibres. Sesame seeds contain a high amount of the antioxidant phytic acid, and lignans pinoresinol and lariciresinol[8]. Essential micronutrients like Calcium, niacin, pantothenic acid and non essential micronutrients like GABA, phytic acid or inositol and taurine are useful in treating anxiety. The presence of calcium, niacin and inositol in *Sesamum indicum* seeds might be contributing favorably to the anti-anxiety activity of Sesame observed in the present study. However the involvement of serotonergic or GABAergic pathway in the mechanism of action of Sesame seeds need to be explored. MDA is an important marker for lipid peroxidation. Sesame seeds decreased brain MDA levels in the present study suggesting that the free radical generation had been diminished, thereby protecting the brains of mice. These results uniformly suggest that Til (sesame seeds) may be looked upon as a promising anti-anxiety agent.

CONCLUSION

Thus, it can be concluded that *Sesamum indicum* possesses useful anti-anxiety activity, which was manifested when consumed along with diet, in the present study. In nutshell, where there is a Til, there is a will.

ACKNOWLEDGEMENT

Ms Isha Dhamija is a UGC-BSR fellow

REFERENCES

1. Pillay NS, Stein DJ. Emerging anxiolytics. Expert Opinion on Emerging Drugs 2007; 12: 541-554.
2. Eisenberg DM, Davis RB, Ettner SL, Appel S, Wilkey S, Rompay V et al. Trends in alternative medicine used in United States. Results of a follow up national survey. Journal of the American Medical Association 1998; 280: 1569-1575.
3. Parle M, Devi S, Verma S. Animal models for screening anxiolytic agents. Annals of Pharmacy and Pharmaceutical Sciences 2010; 1: 116-128.
4. Kuloglu M, Atmaca M, Tezcan E, Gecici O, Tunckol H, Ustundag B. Antioxidant enzyme activities and malondialdehyde levels in patients with obsessive-compulsive disorder. Neuropsychobiology 2002; 46: 27-32.
5. Kuloglu M, Atmaca M, Tezcan E, Ustundag B, Bulut S. Antioxidant enzyme and malondialdehyde levels in patients with panic disorder. Neuropsychobiology 2002; 46: 186-189.
6. Parle M, Chaturvedi D. Eat an orange to keep anxiety at long range. International Research Journal of Pharmacy 2012; 3: 149-151.
7. Thaxter KA, Young LE, Young RE, Prashad O, Addae J. An extract of neem leaves reduces anxiety without causing motor side effects in an experimental model. West Indian Medical Journal 2010; 59: 245-248.
8. Parle M, Dhamija I. Eat til and protect dil. International Research Journal of Pharmacy 2012; 3: 54-57.
9. Lister RG. The use of a plus-maze to measure anxiety in the mouse. Psychopharmacology 1987; 92: 180-185.
10. Bourin M, Hascoët M. The mouse light/dark box test. European Journal of Pharmacology 2003; 463: 55-65.
11. Yadav AV, Kawale LA, Nade VS. Effect of Morus alba L. (mulberry) leaves on anxiety in mice. Indian Journal of Pharmacology 2008; 40: 32-36.
12. Okhawa H, Ohishi N, Yagi K. Assay of lipid peroxides in animals tissue by thiobarbituraic acid reaction. Analytical Biochemistry 1979; 95: 351-358.
13. Takeda T, Tsuji M, Matsumiya T. Changes in head-dipping behavior in the hole-board test reflect the anxiogenic and/or anxiolytic state in mice. European Journal of Pharmacology 1998; 350: 21-29.
14. Radhakrishna A, Kumar H, Chamundeeswari D et al. Evaluation of anti-anxiety activity of Ricinus communis. Pharmanest 2011; 2: 362-368.
15. Chen WW, He RR, Li YF, Li SB, Kurihara H. Pharmacological studies on the anxiolytic effect of standardized Schisandra lignans extract on restraint-stressed mice. Phytomedicine 2011; 18: 1144-1147.
16. Bouayed J, Rammal H, Soulimani R. Oxidative stress and anxiety: relationship and cellular pathways. Oxidative Medicine and Cellular Longevity 2009; 2: 63-67.
17. Bouayed J. Polyphenols: A Potential New Strategy for the Prevention and Treatment of Anxiety and Depression Current nutrition and food science 2010; 6: 13-18.

QUICK RESPONSE CODE	ISSN (Online) : 2277 –4572
	Website http://www.jpsionline.com

How to cite this article:

Parle Milind and Dhamija Isha. Where there is a Til, there is a will. *J Pharm Sci Innov*. 2013; 2(1): 37-40.