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ORIGINAL ARTICLE **OPEN ACCESS**

Pharmacological and *In silico* Evaluation of Methanolic Flower Extract of *Tagetes patula* as Antidepressant and Anxiolytic

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ABSTRACT
Depression and Anxiety belong to neurobehavioral disorders which are considered by their imbalances in neurochemicals such as serotonin, noradrenaline, GABA and dopamine. The present study is envisaged on studying the antidepressant and anxiolytic activities of methanolic extract of flower heads of Tagetes patula. The extract was prepared by soxhlation technique and phytochemical analysis has revealed the presence of flavonoids, terpenoids, essential oils, thiophene derivatives, benzofunan derivatives, alkaloids and tannins. Two dosesof METP 200 and 400 mg/kg, bd.wt were used in the present study. The in vivo antidepressant activity was performed by using forced swim test & tail suspension test. METP has shown a significant reduction in duration of immobility inforced swim test & tail suspension test specifying antidepressant activity. Anxiolytic activity was performed by using elevated plus maze &rotarod test andsignificant anxiolytic effect was observed via increased open-arm exploration in the EPM and reduced muscle contraction in rota rodtest. This research aimed to analyze the biological activity and also to examine the possible mechanism of actions of phytoconstituents through molecular docking. The process of molecular docking involves study of different bonding modes of one ligand with active cavities of target receptors of monoamine oxidase inhibitor (PDB ID: 2NW8), and serotonin transport inhibitors (PDB ID: 3F3A), using MCULE database. Docking studies revealed score values on different receptors for antidepressant and anxiolytic activity and it is observed that constituents, namely terpenoids like β-caryophyllene, δ-elemene, β-ocimene, 1,8-cineole, piperitenone, β-ionone, limonene and flavonoids like quercetin and patuletin showed the best docking results on all the receptors, while the most significant results were observed by quercetin and patuletin on (PDB ID: 2NW8), and (PDB ID: 3F3A). The docking results have given better insights into the development of better monoamine oxidase and serotonin transport inhibitor so as to treat depression and anxiety related neurological disorders. From the above it is clear that METP possess antidepressant and anxiolytic activities.
***Key words:** Tagetes patula, molecular docking, MCULE database, antidepressant and anxiolytic activities.*

INTRODUCTION

Depression is a serious condition, common in adults of all ages worldwide. It is frequently treated with antidepressant drugs, with many countries reporting [1]. It is often accompanied by low self-esteem, loss of interest in normally pleasurable activities (anhedonia), low energy and feeling of worthlessness, sense of rejection, sense of guilt, loss of appetite, insomnia, unnecessary and excessive worry, suicidal thoughts, and pain without a clear cause [2]. There are different types of depressive disorders. Symptoms can range from relatively minor (but still disabling) through to very severe, so it's helpful to be aware of the range of conditions and their specific symptoms, majordepression, melancholia, psychotic depression, antenatal and postnataldepression [3].

Anxiety is accompanied by a characteristic set of behavioral and physiological responses including avoidance, vigilance and arousal, which evolved to protect the individual from danger. The American Psychological Association (APA) defines anxiety as "an emotion characterized by feelings of tension, worried thoughts and physical changes like increased blood pressure". The large numbers of neurotransmitters, peptides, hormones, and other neuromodulators have been implicated in fear and anxiety [4]. Pharmacological treatment of GAD usually employs benzodiazepines (e.g., diazepam and clonazepam), azaspirone (buspirone), and antidepressants (e.g., paroxetine) [5].

Tagetes patula, commonly called French marigold, is a speciesin the daisy familyAsteraceae[6]. The essential oil is being investigated for antifungal activity, including treatment of candidiasis and treating fungal infections inplants. Its root secretions are believed to kill nematodes in the soil and it is said to repel harmful insects, such as white flies on tomatoes [7]. It showed immunomodulatory, anti- arthritic effect [8] and larvicidal activity [9].

Molecular docking has become an increasingly important tool for drug discovery. The molecular docking approach can be used as a model to study the interaction between a small molecule and a protein at the atomic level, which allow us to characterize the behavior of small molecules in the binding site of target proteins as well as to elucidate fundamental biochemical processes. The present study is aimed evaluate the antidepressant and anxiolytic effect of methanolic extract of flower heads of *Tagetes patula.*

MATERIAL AND METHODS

The designing of methodology involves a series of steps taken in a systematic way in order to achieve the goal under prescribed guidelines and recommendation.

Plant collection and drying

Flowers of *Tagetes patula* were collected from Ranga Reddy District, Telangana state in the month of December and was identified and authenticated by botanist Rabiya Sultana, New Government Degree College, Kukatpally. The flowers were cleaned, dried under shade for about six days and coarsely powdered in a mixer grinder and taken up for extraction process.

Preparation of methanolic extract of *Tagetespatula*

The powdered material of flower heads of *Tagetes patula* extracted with methanol by soxhlation technique. Soxhlet extraction is the process of continuous extraction in which the same solvent can be circulated through the extractor for several times. This process involves extraction followed by evaporation of the solvent. The vapors of the solvent are taken to a condenser and the condensed liquid is returned to the drug for continuous extraction.

Preliminary phytochemical analysis of the extract

The extract was subjected to preliminary phytochemical investigations to identify various phytoconstituents present in the methanolic extract of flower heads of *Tagetes patula.*

Acute toxicity testing

Acute toxicity study was carried out in order to check the toxic effects for methanolic extract of flower heads of *Tagetes patula*. Acute toxicity studies were carried out as per the OECD 425 guidelines. The limit test is a sequential test that uses a maximum of 5 animals. The animals were fasted overnight, providing only water, after which the extract was administered to the respective groups orally at the dose level of 2000 mg/kg,bd.wt Observed continuously for 24 h for behavioral, neurological, autonomic profiles and for any lethality.

In vivo methods for evaluation of antidepressantactivity

In vivo evaluation of antidepressant activity of the methanolic extract of flower heads of *Tagetes patula* was carried out in following models Forced swim test and Tail suspension test.

Forced swim test(FST)

The test is based on the observation that Wistar albino rats when forced to swim in a restricted space from which they cannot escape will eventually cease apparent attempts to escape and become immobile apart from the small movements necessary to keep their heads above water. This identifiable behavioral of immobility reflects a state of despair in the rat and showed that immobility was reduced by a variety of agents which are therapeutically effective in depression[10]. Albino rats of either sex weighing about 150-180 gm were selected for this study. The FST was performed after 5 days of treatment using a modified form of the traditional method described by Porsolt. Rats were placed individually in water chamber/tank filled with water (22–25°C) to a depth of 30 cm [11]. The experimental session was of two trials. Conditioning trial on the 4^{th}day and on 5^{th} day of the treatment 2 h from the last dose, the rats were exposed to the cylinders again for 6 min (test session). The frequency and total duration of immobility was determined. Rat was considered immobile when it remained floating in the water, without struggling, making only very slight movements necessary to keep its head above the water [12]. In study design of forced swim test, group II and III receives METP at dose of 200mg/kg and 400mg/kg and group IV receives standard imipramine at 5mg/kg for 1-5 days.

Tail suspension test(TST)

Swiss Albino mice of either sex weighing about 25-30 gm were selected for this study. Immobility was induced by tail suspension according to the procedure of [13] and the animals were free to food and water. All treatments were administered once daily for 5 days. On the 5^{th}day, 2 h after the last dose, mice were suspended by tail individually through a paper adhesive tape, above the table top. Animals were allowed to suspend by their tail for 6 min and the duration of immobility was recorded. Mice were considered to be immobile only when suspended passively and completely motional[14]. Group I serves as control. Group II and III receive METP at 200 and 400 mg/kg dose for 1-5 days. Group IV receives Imipramine at dose of 5 mg/kg *i.p for* 1-5 days.

In vivo methods for evaluation of anxiolytic activity

In vivo evaluation of anxiolytic activity of the methanolic extract of flower heads of *Tagetes patula* was carried out in following models.

Elevated plusmaze (EPM)

EPM is extensively used method to examine anxiolytic effect in rodents. The plus maze apparatus is based on the innate aversion of rodents to open and high space. The apparatus has a central platform of 5 cm connected to two open arms (15 cm × 5 cm) and two closed arms (15 cm × 5 cm × 12 cm), bisecting each other. The maze was raised to a height of 25 cm from the ground. Albino rats of either sex weighing about 150-180 gm were selected for this study. In this model group I serve as control, group II and III receives METP at a dose of 200 mg/kg and 400 mg/kg to for a period of 15 days and group IV serves as a standard Diazepam at dose of 1 mg/kg given for a period of 15 days. On 15^{th}day after 60 min of administration of diazepam (1 mg/kg, *i.p*) and METP (200 & 400 mg/kg, *p.o*) rats were placed at the center of the maze facing an open arm. Number of entries in to open arm and time spent in the open arms was recorded for 5 min. Entry in to an arm is considered if the animal place it's all four paws in to the arm. After each test the maze was carefully cleaned up with a wet tissue paper [15].

Rota rod test

Swiss albino mice of either sex weighing about 25-30 gm were selected for this study. The test consisted of placing the mice upon a cylinder rotating at a speed of 12 rpm. Initially, untreated animals were trained to walk on the cylinder on three consecutive sessions; the pharmacological treatments were tested by placing treated animals on the Rota rod after the training sessions. The number of falls and time of permanence were recorded during a period of 120 seconds were counted. The increase in the number of falls was indicative of coordination failures. After 60 minutes of administration of diazepam and METP animals were tested for their motor coordination [16]. In rota rod test model group I serve as control, group II and III receives METP at dose of 200 mg/kg, *p.o* and 400 mg/kg, *p.o* and group IV serves as a standard Diazepam at dose of 1 mg/kg,*i.p.*

Molecular docking studies

Molecular docking is a kind of bioinformatics modelling which involves the interaction of two or more molecules to give the stable adduct. Depending upon binding properties of ligand and target, it predicts the three-dimensional structure of any complex. Molecular docking generates different possible adduct structures that are ranked and grouped together using scoring function in the software [17]. MCULE is an online drug discovery platform offers a unique solution for pharma and biotech companies by providing molecular models. Docking is done by initially identifying protein from PDB homepage and ligand structures are drawn in MCULE. Then results are obtained and 3D and 2D structures are collected from autovina or discovery studio.

Statistical analysis

Values are expressed as Mean ± SEM, (n=6). All the groups were compared with control, disease control and standard by using Dunnett's test.

RESULTS ANDDISCUSSION

Methanolic extract of flower heads of *Tagetes patula* was explored for its *in vivo* antidepressant and anxiolytic activities using suitable rodent models.

Preparation of methanolic extract of flower heads of *Tagetes patula*

$$\% \text{ of yield obtained} = \frac{\text{Amount of extract obtained (gm)}}{\text{Total amount of powder used (gm)}} \times 100$$

The methanolic extract of flower heads of *Tagetes patula* was prepared by soxhlation technique. The percentage yield of methanolic extract was calculated by using the following formula.

% Yield of extract = 45/500 ×100 = 9% w/w

Preliminary phytochemical analysis

The preliminary phytochemical investigation of methanolic extract of flower heads of *Tagetes patula* revealed the presence of bioactive compounds of which phenolic acids, flavonoids, essential oils, thiophene derivatives, benzofunan derivatives, alkaloids, tannins were the mostprominent.

Table 1: Preliminary phytochemical analysis

Phytochemical constituents	**Results**
Phenolic acids	++
Flavonoids	++
Essential oils	++
Thiophene derivatives	++
Benzafunan derivatives	++
Carotenoids	+
Alkaloids	+
Tannins	+

Note: + indicates present.

Acute toxicity studies

Methanolic extract of flower heads of *Tagetes patula* was tested on swiss albino mice up to a dose of 2000 mg/kg bd. wt. The animals did not elicit any signs of toxicity or mortality up to 2000 mg/kg bd. wt. Hence the extract was found to be safe up to 2000 mg/kg bd. wt.

***In vivo* antidepressant activity model.**

The methanolic extract of flower heads of *Tagetes patula* was screened for its antidepressant activity using the following models

Forced swimtest and tail suspension test.

Table 2: Effect of METP on duration of immobility in forced swim test and tail suspension test

Groups	Treatment	Force swim test	Tail suspension test
		Duration of immobility (sec)	Duration of immobility (sec)
I	Saline water	156.1±0.86	103.1±0.88
II	METP (200 mg/kg, *p.o)*	112.8±0.64^{**,a}	44.8±0.69^{**,a}
III	METP (400 mg/kg, *p.o)*	97.1±0.54^{**,a}	39±0.52^{**,a}
IV	Imipramine (5 mg/kg, *i.p)*	76.5±0.77**	35±0.72*

Values were expressed as mean ± SEM (n=6). Statistical analysis was performed by using ANOVA followed by Dunnett's t-test by comparing with control & standard. Significant values are expressed as control group (**p<0.01, *p<0.05) and standard(a=p<0.01).

From the above results, it is clear that the duration of immobility in control group was found to be higher but in groups treated with the METP and standard (imipramine 5 mg/kg, *i.p*) the duration of immobility was found to be reduced.

***In vivo* anxiolytic activity models**

Methanolic extract of flower heads of *Tagetes patula* was screened for its anxiolytic activity using the following models i.e., elevated plus maze and rota rod test.

Elevated plus maze

Elevated plus maze is ubiquitously used model to determine anxiolytic activity and constitutes a simple and routine rodent model for evaluation of behavioral exploratory activity in rodent models.

Table 3: Effect of METP on elevated plus maze

Groups	Treatment	No. of entries	Time spent (sec)
		Open arms	Open arms
I	Saline water	7.6±0.56	94±0.84
II	METP (200 mg/kg, *p.o)*	10.33±0.60^{**,a}	128.5±0.69^{**,a}
III	METP (400 mg/kg, *p.o)*	14.6±0.65^{**,a}	185.5±0.96^{**,a}
IV	Diazepam (1 mg/kg, *i.p)*	17.1±0.54**	224±0.78**

Values were expressed as mean ± SEM (n=6). Statistical analysis was performed by using ANOVA followed by Dunnett's t-test by comparing with control & standard, Significant values are expressed as control group (**p<0.01) and standard (a=p<0.01).

From the above results it is clear that in groups treated with the METP and standard (diazepam 1 mg/kg, *i.p*) the number of entries and time spent in open arms was found to be increased, but these were found to be less in control group.

Rota rod test

Table 4: Effect of METP on muscle rigidity using rota rod test

Groups	Treatment	No of falls	Time of permanence (sec)
I	Saline water	1.66±0.30	102.5±0.90
II	METP (200 mg/kg, *p.o)*	4±0.47^{*,a}	84.6±0.96^{**,a}
III	METP (400 mg/kg, *p.o)*	5.5±0.69^{**,b}	79.6±0.80^{**,a}
IV	Diazepam (1 mg/kg, *i.p)*	7.66 ±0.60**	74.3±0.93**

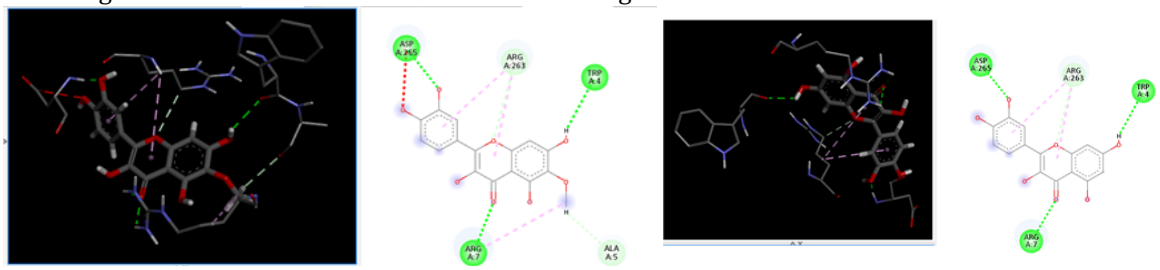
Values are expressed as Mean ± SEM, (n=6). Statistical analysis was performed by using ANOVA followed by Dunnett's t-test. Results were compared with control group (**p<0.01, *p<0.05) and standard (a = p< 0.01).

Molecular docking studies

To understand the interactions between the ligands and 3F3A and 2NW8 protein and to explore their binding mode, docking study was performed using MCULE database. The ligands were created and prepared for the docking procedure using ChemSketch and MCULE database.

Docking scores

The results revealed the docking scores of various active constituents found in the extract out of which the highest glide score were observed for Patuletin and quercetin given in table 5. Patuletinwith G-score -6.2 against (PDB ID: 3F3A) and with a G-score of -6.0 against (2NW8), Quercetin with G-score of -6.0against (PDB ID: 3F3A) and a G-score of –5.8against (PDB ID: 2NW8) respectively and the binding mode of ligands with 3F3A and 2NW8 were shown in Figure1.



Patuletin -6.2 in 3F3A **Quercetin -6.0 in 3F3A**

Quercetin -5.8 in 2NW8 **Patuletin -6.0 IN 2NW8**

Figure 1: The binding interactions of quercetin and patuletin against two proteins

Table 5: Glide scores of compounds from METP against PDB ID: 3F3A and PDB ID: 2NW8

S. No	Name of the Compound	G-Score	
		3F3A	2NW8
1	Quercetin	-6.0	-5.8
2	β-caryophyllene	-3.9	-5.4
3	δ-Elemene	-4.4	-4.9
4	Patuletin	-6.2	-6.0
5	Piperitenone	-4.2	-4.5
6	Tagetenone	-3.6	-4.4
7	B-Ocimene	-3.6	-4.2
8	B-Ionone	-3.9	-4.7
9	Limonene	-3.3	-4.0
10	2,6-Dimethylocta-5,7-dien-4-one	-	-4.2
11	Imipramine	-5.1	-5.8
12	Diazepam	-4.8	-6.1

DISCUSSION

The GC-MS studies of methanolic extract of flower heads of *Tagetes patula* showed the presence of various phytochemical constituents like flavonoids namely Quercetin, β-caryophyllene, δ-Elemene, Patuletin, Piperitenone, Tagetenone, β-Ocimene, β-Ionone, Limonene, 2,6-Dimethylocta-5,7-dien-4-one,phenolic acids, essential oils, thiophene derivatives, benzofunan derivatives, alkaloids, carotenoids and tannins [18].In the present study methanolic extract of flower heads of *Tagetes patula* was investigated against widely used animal models namely forced swim test (FST) and tail suspension test (TST) and their immobility time is measured for antidepressant activity. The decrease in duration of immobility is considered to be a good predictive value in the evaluation of potential antidepressant agents [19]. The treatment with the extract has produced a significant reduction in depression in forced swim test (FST) and tail suspension test (TST) models when compared with control group. The efficacies of the extract were comparable with standard imipramine.

The elevated plus maze (EPM) is considered to be an etiologically valid animal model for screening the anti-anxiety drugs [15]. An anxiolytic agent increases the frequency of entries into the open arms and increases the time spent in open arms of the EPM [20]. Anxiolytic activity was further evaluated in the rota rod test for evaluating the motor coordination in rodent models as impaired motor co-ordination is an indicative of anxiolytic activity [21].

The results have shown that the methanolic extract of flower heads of *Tagetes patula* improved the number of entries as well as time spent in open arms in EPM and also increased the no of falls and time of permanence in rota rodtest. The literature reveals relative deficiency in GABA neurotransmission, is responsible for anxiogenic. Since METP also contains flavonoids like patuletin, quercetin, kampherol and phenolic compounds, alkaloids essential oils (linalool, β pinene) are known to possess antidepressant [22] and anxiolytic activity [23].thiophene derivatives, alkaloids, carotenoids, and benzafunan derivatives in *Tagetes patula* are known for its anti-anxiety activity and treating contracted skeletal muscles in anxiety conditions via GABAergic effect and by reducing the corticotrophin releasing hormone in the brain [5], whereas as β carboline alkaloids, were used as precursors for the synthesis of the trypataminederivates which are used in the prevention of depressive states.

Molecular docking continues to holds great promise in the field of computer based drug design which screens small molecules by orienting and scoring them in the binding site of a protein. The docking analysis of isolated compounds from *Tagetes patula* and standard drugs like Imipramine and diazepam were carried out using MCULE database. The various constituents identified in the plant extract are Quercetin, β-caryophyllene, δ-Elemene, Patuletin, Piperitenone, Tagetenone, β-Ocimene, β-Ionone, Limonene, 2,6-Dimethylocta-5,7-dien-4-one were subjected to docking against 3F3A AND 2NW8. The results revealed that patuletin and quercetin had shown highest glide scores among the various phytochemical constituents present in the extract. The glide score of standard drugs imipramine and diazepam were found to be -5.1and -4.8 against 3F3A, -5.8 and -6.1 against 2NW8**.** The glide scores of the patuletin and quercetin were found to be similar with the glide score of standard drugs stating that the compounds have same affinity to bind to the proteins [24]. These results clearly indicate that the chemical constituents patulet in and quercetin might have shown similar mechanism to that of the standard drugs imipramine and diazepam in reducing depression and anxiety

CONCLUSION

The methanolic extract of flower heads of *Tagetes patula* was screened for its antidepressant and anxiolytic activity. Further studies are needed to be carried out to isolate individual phytochemical constituents of the extract and to establish the exact mechanism for its antidepressant and anxiolytic activities.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

REFERENCES

1. Coupland, C., Hill, T., Morriss, R., Moore, M., Arthur, A., &Cox, H. (2018). Antidepressant use and risk of adverse outcomes in people aged 20-64 years: cohort study using a primary care database. BMC Med. 16(36):1-24.
2. Fasipe, O.J. (2018). Neuropharmacological classification of antidepressant agents based on their mechanisms of action. Arch. Med.Health.Sci.,6(1):88-94.

3. Stahl, S.M. (1996). Stahl's Essential Psychopharmacology: Neuroscientific Basis and Practical Applications. Cambridge: Cambridge University Press,

4. Bhandari, S.&Kabra, M. (2014). To evaluate anti-anxiety activity of thymol. Journal of Acute Disease., 3(2):136-140.

5. De, Sousa. D., Hocayen, P., Andrade, L.&Andreatini, R. (2015). A Systematic Review of the Anxiolytic-Like Effects of Essential Oils in Animal Models. Molecules.,20(10):18620-18660.

6. Hort.ifas.ufl.edu. (2018). Offline Available at provided link below https://hort.ifas.ufl.edu/ database/ documents/pdf/ shrub_fact_sheets/tagsppc.pdf.

7. Kashif, M., Bano, S. &Naqvi, S. (2015). Cytotoxic and antioxidant properties of phenolic compounds from *Tagetes patula* flower. Pharma. Biol., 53(5):672– 681.

8. Jabeen, A., Mesaik, M., Simjee, S., BanoL.S. &Faizi, S. (2016). Anti-TNF- α and anti-arthritic effect of patuletin: A rare flavonoid from *Tagetes patula*. Int. Immunopharmacol., 36:232-24.

9. Dharmagadda, V., Naik, S., Mittal, P. &Vasudevan, P. (2005). Larvicidal activity of *Tagetespatula*essential oil against three mosquito species. Bioresource Technol., 96(11):1235-1240.

10. Porsolt, R., Anton, G., Blavet, N. &Jalfre, M. (1978). Behavioural despair in rats: A new model sensitive to antidepressant treatments. Eur. J. Pharmacol., 47(4):379-391.

11. Detke, M., Rickels,M. &Lucki, I. (1995). Active behaviors in the rat forced swimming test differentially produced by serotonergic and noradrenergic antidepressants. Psychopharmacol., 121(1):66-72.

12. Sousa, F.C.F., Melo, C.T.V., Monteiro, A.P., Lima, V.T.M., Gutierrez, S.J.C, Pereira, B.A., Barbosa-Filho, J.M., Vasconcelos, S.M.M., Fonteles, M.F. &Viana, G.S.B. (2004). Antianxiety andantidepressant effects of Riparin III from *Anibariparia (Nees)* Mez (Lauraceae) inmice. Pharmacol.Biochem.Behav., 78:27–33.

13. Steru, L., Chermat, R., Thierry, B. and Simon, P. (1985). The tail suspension test: A new method for screening antidepressants in mice. Psychopharmacol.,85(3):367-370.

14. Ibrahim, A., Kelly,S., Adams, C. &Glazebrook, C. (2013). A systematic review of studies of depression prevalence in university students. J.Psychiatric. Res., 47(3):391-400.

15. Raju, G.M., Gouthami, K., Reddy, N.V.L.S. & Nicholas, M. (2020). Anti-Amnesic Effect of Methanolic Leaf Extract of *TecomaStans*: An Experimental Study in Rodents. J. Young. Pharm., 12(2)Suppl:s91-s6

16. Lopes, C.,Gonc,C.A.,deAlmeida, A.C.,Pereirada,J.C.,Costa, T.H.,Mendes, C., Barros, G. &Mendes, F. (2011). Sedative, anxiolytic and antidepressant activities of *Citrus limon*(Burn) essential oil in mice. Pharmazie.,66: 623–627.

17. Ayaz, M.D., Shafir M. Molecular docking: Approaches, types, applications and basic challenges. J. Analytical &Bioanalytical Tech., 2017; 8(2): 1-3.

18. Faizi,S., Fayyaz, S., Bano, S., Yawar,Iqbal, E., Siddiqi,H. &Naz,A. (2011). Isolation of Nematicidal Compounds from *Tagetespatula L*. Yellow Flowers: Structure–Activity Relationship Studies against Cyst Nematode Heteroderazeae Infective Stage Larvae. J. Agricultural. Food Chem., 59(17):9080-9093.

19. Vinod P, Akhade A, Baokar S, ShivakumarH. (2011). Antidepressant-like effects of *Acoruscalamus*in forced swimming and tail suspension test in mice. Asian. Pacific. J. Trop. Biomed.,(2012): 17-19.

20. Dawson, G., Rupniak, N., Iversen, S., Curnow, R., Tye, S., Stanhope, K. and Tricklebank, M. (1995). Lack of effect of CCKB receptor antagonists in ethological and conditioned animal screens for anxiolytic drugs. Psychopharmacol.,121(1):109-117*.*

21. Costa, J., de Oliveira, G., de Almeida, A., Islam,M., de Sousa,D. &de Freitas,R. (2014). Anxiolytic-like effects of phytol: Possible involvement of GABAergic transmission. Brain Res., 1547:34-42.

22. Kumar, P., Yogita, M. &Kuldeep, B. (2012). Reversal by quercetin of corticotrophin releasing factor induced anxiety- and depression-like effect in mice. Progress.NeuroPsychopharmacol. Biol. Psychiatry., 34(6): 955-960.

23. Joan,M. &Michiho,I.(2014). Sedative, anxiolytic and antidepressant-like effects of inhalation of the essential oil of *Ocimum gratissimum L*. from Cameroon in mice. J. Pharmacog. Phytochem.,2(5):1-9.

24. Raju,M.G., Srilakshmi,S.& ReddyN.V.L.S. (2019). Anti-Inflammatory, *In-Silico* Docking and ADME Analysis of some isolated compounds of *Tagetes erecta* Flower Heads. Int. J. Pharm. Sci. Res.,11(3): 1358-1366.

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