Open Access
Full Length Article

ORIGINAL ARTICLE

Determination of the Anxiolytic Potential of New Polyhedral Formulation

Saira Bano[1], Hina Mahmood[1], Kashifa Khanum[1], Khan Usmanghani[1,*], Samina Afzal[2], Muhammad Daniyal[3]

[1]Department of Research and Development, Herbion Pakistan (Pvt.) Limited, Karachi, Pakistan
[2]Faculty of Pharmacy, Bahauddin Zakriya University, Multan, Pakistan
[3]Department of Medical Affairs and Training, Herbion Pakistan (Pvt.) Limited, Karachi, Pakistan

Keywords: *Anxiolytic, polyherbal, Evolvulus alsinoids, Lavadula stoechas, Melissa officinalis, Rauwolfia serpentine, Valeriana officinalis.*

Author`s Contribution
All the authors contributed significantly to the research that resulted in the submitted manuscript.

Article info.
Received: October 5, 2016
Accepted: December 12, 2016

Funding Source: *Nil*
Conflict of Interest: *Nil*

Cite this article: *Bano S, Mahmood H, Khanum K, Usmanghani K, Afzal S, Daniyal M. Determination of the Anxiolytic Potential of New Polyhedral Formulation. J. Pharm. Pharm. Sci. 2017;5(1):14-17.*

Address of Correspondence:
ugk_2005@yahoo.com

ABSTRACT

Objective: The present study was aimed at investigating preclinical efficacy and toxicity of Anti-anxiety product. The different herbs have been used both traditionally and contemporaneously as anxiolytics. The polyherbal Anti-anxiety product consisting of Evolvulus alsinoides (Ariel part), Lavandula stoechas (flowers), Melissa officinalis (whole plant), Rauwolfia serpentine (roots) and Valeriana officinalis (roots) has been developed.

Results: The preclinical study on toxicity on animal model generated data which were analyzed statistically. The different tests were performed such as acute toxicity, elevated pulse maze, open field test, light-dark box test, locomotor activity. The results so obtained with polyherbal anti-anxiolytic drugs exhibited no toxicity and found to possess anxiolytic properties.

Conclusion: It was concluded that Polyherbal formulation has exhibited anti anxiolytic activity comparable to Diazepam.

INTRODUCTION

Anxiety is a psychosomatic and functional illness that categorized by somatic, emotional, cognitive, and behavioral constituents. It is the unpleasant perception of terror and worry. The source connotation of the term anxiety is 'to vex or trouble'; in either existence or absenteeism of psychological stress, anxiety can generate an emotional state of terror, worry, nervousness, and trepidation. But, anxiety must not be muddled with fear, it is more of an anxious about feeling about somewhat which seems threatening and can overwhelmed an individual. It can lead to difficulties such as the conquest of the immune system, digestive disorders, muscle tension, short-term memory loss, premature coronary artery disease, heart attack. If unwarranted disturbing and high anxiety go untreated, they can lead to depression and even hopeless thoughts.

Anti-anxiety product tested was composed of: Evolvulus alsinoides (aqueous extract of ariel part) was reported as Tranquilizer (Indian medicinal plant), Lavadula stoechas (aqueous extract of flowers) was reported to possess the effects of seductive activity (PDR), Melissa officinalis (aqueous extract of the whole plant) was reported to possess the significant sedative activity (Medicinal plants of the world, WHO 2, PDR), Rauwolfia serpentine (aqueous extract of roots), antihypertensive, tranquilizer (Medicinal plants of the world, WHO I, traditional medicine, PDR) Valerian a officinalis (aqueous extract roots) was reported as sedative (British herbal pharmacopeia, Traditional medicine, PDR).

METHODOLOGY

Animals

Male NMRI mice weighted 25g to 35g, where be gained from the Herbion Pak. Pvt. Ltd. Animal house facility and they were kept under the standard conservation circumstances and provided the Food and water were available ad libitum. The standard conservation circumstance is 25 ± 1 °C and 12 h dark / light cycle.

Chemicals

The polyherbal anti-anxiety formulation was provided by Herbion Pak. Pvt. Ltd. Sodium chloride and Diazepam was purchased from Merck Pvt. Ltd and EPLA pharmaceutical respectively. Distilled water was used during entire course of study.

Acute toxicity

The mice (n=10 / group) was treated (100, 500 or 1000 mg/kg) orally with poly herbal formulation. The mortality and physical signs of toxicity (weight changes and hair loss) were monitored for 1 week.

Elevated plus maze

The mice were treated (oral) polyherbal formulation (10, 50, 100, 500 or 1000 mg/kg), diazepam (2.5 mg/kg) or water (10 ml/kg) of the. After 30 minutes, the animals were subjected to elevate plus maze (plus shape maze with a dimension of 25 x 5 cm of each arm at the height of 50 cm. The close arm is surrounded with 15 cm high walls). The quantity of entrances and time used up in the open arms was noted for 5 minutes and matched with control.

Open field test

After 30 minutes of treatment (10, 50, 100, 500 or 1000 mg/kg) with the polyherbal formulation, the mice were individually placed in the open field (2 x 2 feet box with 1 x 1 feet square in the center). The quantity of entrances and time used up in the center was recorded for 5 minutes and matched with control. Diazepam (2.5 mg/kg) was used as a +ve control.

Light dark box test

After 30 minutes of treatment with polyherbal formulation (10, 50, 100, 500 or 1000 mg/kg), diazepam (2.5 mg/kg) or water (10 ml/kg), mice were subjected to light dark box (1x1 feet light box connected via hole to 1x1 feet box covered from top to make it dark). The mice were positioned in the dark box and the quantity of entrances and time used up in the light box was noted for 5 minutes.

Locomotor activity test

The mice were treated with 50, 100, 500 or 1000 mg/kg of polyherbal formulation. After 30 minutes, the animals were kept in the activity cage for 5 minutes. The cage is 1.5 by 1 feet box having 3 x 3-inch markings on the floor. The quantity of boxes crossed by the animals was noted and matched with control.

RESULTS

Effect of poly herbal formulation on toxicity in mice

At the given doses of (100, 500 or 1000 mg/kg), no mortality and apparent signs of toxicity was observed.

Effect of polyherbal formulation on anxiolytic potential using elevated plus maze

The test formulation significantly increased the time consumed in the open arm at the doses of 100 mg/kg ($p < 0.005$) and 500 mg/kg ($p < 0.005$) as matched to control. The quantity of entrances in open arm was also increased at the similar doses ($p < 0.05$ and $p < 0.005$ respectively). Diazepam treated mice also presented significant upsurge ($p < 0.05$) in the time spent and the quantity of entrances in the open arm as matched to control (Table **1**).

Table 1. Effects of polyherbal on plus maze behavior in mice.

Treatment	Dose (mg/kg)	Time spent in open arm (sec)	Entries in open arm (count)
Herbal	10	29 ± 17	1.6 ± 0.7
	50	32 ± 18	1.6 ± 0.7
	100	111 ± 24***	4.6 ± 1.2*
	500	122 ± 16***	5.6 ± 0.5***
	1000	37 ± 13	2.6 ± 1.0
Diazepam	2.5	92 ± 27*	5.2 ± 1.0*
Control		32 ± 16	1.6 ± 0.9

Effect of polyherbal formulation on anxiolytic action using light-dark box test

The polyherbal formulation significantly ($p < 0.05$) improved the time consumed in the light box at the light box at the given dose of 100 mg/kg. The standard diazepam also showed significant enhancement in the time consumed in light box ($p < 0.01$). However, both failed to increase in the quantity of entries in the light box at given doses (Table **2**).

Table 2. Effects of polyherbal on light-dark box test behavior in mice.

Treatment	Dose (mg/kg)	Time spent in light box (sec)	Entries in light box (count)
Herbal	10	125 ± 28	5 ± 1
	50	90 ± 18	6 ± 1
	100	171 ± 48*	4 ± 1
	500	122 ± 31	5 ± 1
	1000	96 ± 16	5 ± 1
Diazepam	2.5	174 ± 29**	5 ± 2
Control	67 ± 13	6 ± 1	

Effect of poly herbal formulation on the locomotor activity test in mice

At the given doses of 50, 100, 500 or 1000 mg/kg, the poly herbal formulation failed to significantly alter the locomotors activity of mice as matched to control (Table **3**).

Table 3. Effects of polyherbal on locomotor activity in mice.

Treatment	Dose (mg/kg)	Number of boxes crossed
Herbal	50	115 ± 27
	100	103 ± 14
	500	81 ± 6
	1000	86 ± 7
Control		107 ± 24

Effect of poly herbal formulation on anxiogenic potential in open field test

At the given doses of 50, 100, 500 or 1000mg/kg, the poly herbal formulation failed to significantly alter the locomotors activity of mice as matched to control (Table **4**).

Table 4. Effects of polyherbal on open field test in mice.

Treatment	Dose (mg/kg)	Time spent in center area(sec)	Entries in center area (count)
Herbal	10	13 ± 6	4 ± 1
	50	18 ± 5	6 ± 1
	100	17 ± 1	4 ± 1
	500	8 ± 3	5 ± 1
	1000	6 ± 2	4 ± 1
Diazepam	2.5	10 ± 2	4 ± 1
Control		13 ± 1	6 ± 1

Statistical Analysis

In elevated plus maze, the quantity of entries and time consumed in open arm are presented as mean ± SEM of counts and seconds respectively. The number of boxes crossed in activity cage is shown as mean ± SEM of counts. Variances between various means were evaluated by one-way ANOVA followed by LSD. Asterisks represent * ($p < 0.05$), ** ($p < 0.01$) and *** ($p < 0.005$) as matched to control.

DISCUSSION

As polyherbal formulation contains, the different chemical constituents have so far been reported and out of the Evolvulus alsinoides C-E have shown in normalizing stress activity [1].

Shamsi et al. have conducted clinical studies on the management anxiety neurosis on patients with Evolvulus alsinoides and compared with placebo, the statistically it was proved that the former relieved anxiety symptoms without producing any side effects [2]. The use of extracts of Melissa officinalis have been cited to be effective for anxiolytic action in dropping stress and physiological instabilities due to its straight interface with the CNS and the cholinergic and GABAergic systems [3]. Lavender, Lavadula stoechas was subjected for the management of anxiety illnesses and associated situations and clinical trials of anxiolytic consequence of lavender was superior to placebo in 221 subjects suffering from anxiety illness [4]. This suggests that Lavandula stoechas exhibit superior anxiolytic activity in human subjects. The citation on efficacy of Rauwolfia serpentina drugs in the control of unconcealed anxiety in ambulatory psychiatric patients is equivalent to that of a conservative treatment of anxiety, as shown in a study of corresponding

patient's groups. The uses of Rauwolfia drugs were well known and effortlessly controlled the depressive symptoms are not altered by Rauwolfia drugs given at the amount and duration levels of clinical trials. This study provides genuine in-house generated information reflecting preclinical toxicity and efficacy of Anti-anxiety product, which will increase our confidence in the product and will facilitate the registration process.

If these considerations are taken into account, then all the parameters such as acute toxicity, elevated plus maze, open field test, light-dark box test, locomotor activity test, the data so generated clearly exhibit that the polyherbal formulation in combination exert antianxiolytic effects. This can be explained that the different chemical constituents to gather act in unison like fashion to exert pronounce antianxiolytic effects and this can be explained that all the chemicals synergistically brought up beneficial effects to relieve anxiety.

CONCLUSION

The product seems to have a typical "U-shape" response with the effective dose lies at around 100 mg/Kg. The anxiolytic activity was found to be pronounced with polyherbal product. The results are to resolve that the different medicinal plants have already been cited as anxiolytic relief both in experimental and clinical evidences. However, if these plants are put together in polyherbal products, then such types could still be valid to be utilized for anxiety. These ingredients in this experimental study have verified the claims to give the antianxiety effects and function.

REFERENCES

1. Gupta P, Sharma U, Gupta P, Siripurapu KB, Maurya R. Evolvosides C-E, flavonol-4'O-triglycosides from Evolvulus alsinoides and their anti-stress activity. Bioorg Med Chem. 2013; 21(8):1116-22.
2. Shamsi Y, Ahmad J, Khan AA. A clinical study on the management of anxiety neurosis with Sankhaholi. Indian J Traditional Knowledge. 2007; 6(4):668-77.
3. Fermino BL, Khalil NM, Bonini JS, Pereira RP, da Rocha JB, da Silva WC. Anxiolytic properties of Melissa officinalis and associated mechanisms of action: A review of the literature. Afr J Pharm Pharmacol. 2015; 9(3): 53-9.
4. Koulivand PH, Khaleghi Ghadiri M, Gorji A. Lavender and the nervous system. Evid Based Complement Alternat Med. 2013; 2013:681304.