

ANXIOLYTIC AND ANTIDEPRESSANT POTENTIAL OF HYDROALCOHOLIC
LEAVES EXTRACT (HLE) OF *HIPPEASTRUM VITTATUM* IN RATS.*¹Pragya Mishra, ²Pallavi Tiwari and ³Manoj Kumar Mishra¹Research Scholar, Shambhunath Institute of Pharmacy, Jhalwa, Prayagraj- 211012 (UP).²Assistant Professor, Shambhunath Institute of Pharmacy, Jhalwa, Prayagraj- 211012 (UP).³Professor, Shambhunath Institute of Pharmacy, Jhalwa, Prayagraj- 211012 (UP).

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ABSTRACT

Fear, dread, and unease are all symptoms of anxiety. It can make you sweat, feel restless and tight, and cause your heart to race. Women are more likely than men to acquire emotional anxiety during adolescence. Every year, around 700,000 people die by suicide. Suicide is the fourth highest cause of death among those aged 15 to 29. The present research focuses on pharmacological screening of anxiolytic and antidepressant effect of hydroalcoholic flower extract (HFE) of *Hippeastrum vittatum* in Wistar albino rats. The flowers of *Hippeastrum vittatum* was obtained from the NBRI, Lucknow region and extracted by maceration in hydroalcoholic solution- Ethanol + distilled water (1:1) for fifteen days with gradual stirrings. Parameters- Elevated Plus Maze (EPM), Light/dark arena test, FST and Actophotometer were utilized. In screening of anxiolytic and antidepressant potential, *Hippeastrum vittatum* demonstrated significant decrease in anxiety and depression in rats when compared with control. In conclusion, hydro-alcoholic flower extract (HFE) of *Hippeastrum vittatum* is significant anxiolytic and anti-depressant herbal drug. It can be effectively used in the treatment of depression, mental agitation and other neurological disorders after successfully evaluating mechanism of action of the plant- flower.

KEYWORDS: *Hippeastrum vittatum*, Lily flower, anxiolytic, antidepressant, animal model.

INTRODUCTION

Fear, dread, and unease are all symptoms of anxiety. It can make you sweat, feel restless and tight, and cause your heart to race. Women are more likely than men to acquire emotional anxiety during adolescence. Women are 1.5-02 times more likely than males to suffer from anxiety disorders (Thibaut, 2017). Panic disorder, GAD, and SAD had lower lifetime frequency in adolescents aged 13 to 17 years than in adults aged 18 to 64, although anxiety attacks, separation anxiety disorder, and agoraphobia without even a history of panic attacks were more common (Kessler et al. 2012). As a result, anxiety disorders in basic care are frequently misdiagnosed and undertreated (Witcher et al. 2002). Every year, around 700,000 people die by suicide. Suicide is the fourth highest cause of death among those aged 15 to 29 (Evans-Lacko et al. 2018).

Hippeastrum is a genus of plants that includes a lot of alkaloids and is becoming more popular as a treatment for neurological illnesses and neurodegenerative diseases (Silva et al. 2008). It is made up of more than 70 species that are found in South America's tropical and subtropical regions (Poggio et al. 2007). *Hippeastrum vittatum* possesses anticancer, antiviral, anti-

inflammatory, and other pharmaceutical properties. Malignant tumours have emerged as one of the most serious risks to human health and mortality (Chen et al. 2015; Wang et al. 2015; Liu et al. 2015; Chen et al. 2014).

The present research focuses on pharmacological screening of anxiolytic and antidepressant effect of hydroalcoholic flower extract (HFE) of *Hippeastrum vittatum* in Wistar albino rats.

MATERIALS AND METHODS

Requirements

Hippeastrum vittatum hydroalcoholic flower extract (HFE), Imipramine (API), Water bath, distilled water, Wistar albino rats (either sex), rotatory evaporator, weighing machine and ethanol.

Authentication & extraction of plant

The flowers of *Hippeastrum vittatum* was obtained from the NBRI, Lucknow region. It will be identified and authenticated by a botanist. The flowers are washed making dust-free and dried at room temperature or shade and extracted by soaking into hydroalcoholic solution-

Ethanol + distilled water (1:1) for fifteen days with gradual stirrings (Khan et al. 2020).

Preparation of animals

Albino rats of either sex weighing 150–200 g will be obtained from the Animal House, CDRI Lucknow. The animals are maintained in proper conditions, at room temperatures of $25 \pm 1^\circ\text{C}$ with 12-hour light/dark cycle. The relative humidity is maintained at 44-56%, and are fed with standard rodent diet and water ad libitum. Animals will keep on fasting but free access to water up to 1 h before the induction of ulcers (Bhajoni et al. 2016).

Experimental protocols

All the rats are divided into four groups (n=6) as followings-

Group 1: Rats are given only normal saline each day for 21 days.

Group 2: Rats are given Imipramine (10mg/kg/day, p.o.) for 21 days.

Group 3: Rats are given HFE of *Hippeastrum vittatum* (200mg/kg/day, p.o.) for 21 days.

Group 4: Rats are given HFE of *Hippeastrum vittatum* (400mg/kg/day, p.o.) up to 21 days.

Protocols

1. Elevated Plus Maze (EPM) Test

The Elevated Plus Maze (EM) consists a 5 cm wide circular pathway; elevated 27 cm from the floor and diameter of maze kept 65 cm. The circular pathway is divided into 4 quadrants in which 2 are open and 2 are closed quadrants- where wall is 27 cm in height. Rats are placed facing towards anyone of the closed quadrants during each trial. Rats are allowed to explore the apparatus for 5 minutes only. No. of entries and time spent in open quadrants are recorded till 5 minutes (Kulkarni, 1999).

2. Light-Dark Arena Model

In light-dark arena model, a 100Watt bulb is being placed 30 cm above to base of box. Rats are kept in centre of light arena (box) and have to expose for 5 minutes. No. of entries and time spent in light arena segment are recorded till 5 minutes. It is cleansed every time before keeping a new rat (Khan et al. 2020).

3. Forced Swimming Test

Rats are dropped in glass (30×20cm) filled with water at depth of 15 cm and temperature maintained at approx. 30°C . Rats are allowed to forcefully swim for the period of 5 min. The total mobility time is recorded each time in seconds in 5 min using stopwatch (Kishore, 2015).

4. Locomotion Activity

Actophotometer is tuned on to check and make sure that all the photocells are working properly for accurate readings. Rats are placed once at a time in the activity cage for 10 min. Activity score is recorded for each rat till 10 min. Finally, motor activity is observed and compared with standard drug- Imipramine (Kulkarni, 1999).

RESULTS AND DISCUSSION

1. Elevated Plus Maze (EPM) Test

In EPM test, control group showed no. of entries, time spent and % of time spent in open arm as $4.42 \pm 0.27^*$, 67.32 ± 0.32 and $22.10 \pm 0.14^{**}$ respectively. Whereas, HFE of *Hippeastrum vittatum* treated group (200mg/kg) showed no. of entries and % of time spent as $9.23 \pm 0.15^{**}$ and $48.46 \pm 0.28^*$ respectively. In contrast, HFE of *Hippeastrum vittatum* (400mg/kg) administered group exhibited no. of entries and % of time spent as $8.36 \pm 0.30^{***}$ and $41.37 \pm 0.32^{**}$ respectively. It can easily be seen difference between test and control group. This model clearly confirms that the plant *Hippeastrum vittatum* flower are effective in treatment of anxiety and depression in animal model.

Table 4.1: No. of entries, time spent & % of time spent of *Hippeastrum vittatum*.

Treatment	In open arm (sec)		
	No. of entries	Time spent	% of time spent
Normal saline	$4.42 \pm 0.27^*$	67.32 ± 0.32	$22.10 \pm 0.14^{**}$
Imipramine (10mg/kg)	$9.23 \pm 0.15^{**}$	149.20 ± 0.43	$48.46 \pm 0.28^*$
HFE of <i>Hippeastrum vittatum</i> (200mg/kg)	$5.79 \pm 0.10^*$	94.29 ± 0.41	$32.73 \pm 0.23^{**}$
HFE of <i>Hippeastrum vittatum</i> (400mg/kg)	$8.36 \pm 0.30^{***}$	125.11 ± 0.26	$41.37 \pm 0.32^{**}$

Significance Level= *

Values were given in Mean \pm S.E.M. and found statistically significant at $P < 0.05$, compared to control (n=6)

2. Light/dark arena test

In light/dark arena test, no. of entries, time spent and % of time spent in light arena were recorded for 5 min. In Imipramine treated rats, no. of entries in light arena was recorded as $8.81 \pm 0.41^*$ and time spent 152.40 ± 0.61 sec and thus % of time spent as $48.61 \pm 0.25^{**}$ which was highest among all. Hydroalcoholic flowers extract (HFE)

of *Hippeastrum vittatum* also showed increased no. of entries and % of time spent in light arena as $6.38 \pm 0.24^{**}$ and $34.43 \pm 0.19^{**}$ respectively at the dose of 200mg/kg. Whereas, HFE of *Hippeastrum vittatum* exhibited $7.89 \pm 0.59^{**}$ (no. of entries) and 126.36 ± 0.42 (time spent) in light arena at the dose of 400mg/kg.

Table 4.2: Light/dark arena test of control, standard and *Hippeastrum vittatum* treated groups.

Treatment	In light arena (sec)		
	No. of entries	Time spent	% of time spent
Normal saline	5.35±0.33**	66.23± 0.71	21.07±0.32**
Imipramine (10mg/kg)	8.81±0.41*	152.40± 0.61	48.61±0.25**
HFE of <i>Hippeastrum vittatum</i> (200mg/kg)	6.38±0.24**	95.29±0.53	34.43±0.19**
HFE of <i>Hippeastrum vittatum</i> (400mg/kg)	7.89±0.59**	126.36±0.42	42.12±0.40**

Significance Level= *

Values were given in Mean ± S.E.M. and found statistically significant at P<0.05, compared to control (n=6)

3. Forced swimming test

In FST, mobility time was observed lowest in the case of control and highest in control which indicates for their anti-depressant accordingly. HFE of *Hippeastrum vittatum* exhibited increase in mobility time as 243.26±0.70** at dose 200mg/kg and 267.13±0.43*** at

400mg/kg. In both the doses, it significantly proved for its anxiolytic and anti-depressant potential by facilitating the mood of animals. At higher dose, its effect was similar about same to standard group. It might be effective in relieving the depression, anxiety and low mood in human too.

Table 4.3: Mobility time in FST of control, standard and *Hippeastrum vittatum* treated rats.

Treatment	Mobility time (sec) Mean± SEM
Normal saline	234.19±0.70*
Imipramine (10mg/kg)	283.36±0.52***
HFE of <i>Hippeastrum vittatum</i> (200mg/kg)	243.26±0.70**
HFE of <i>Hippeastrum vittatum</i> (400mg/kg)	267.13±0.43***

Significance Level= *

Values were given in Mean ± S.E.M. and found statistically significant at P<0.05, compared to control (n=6)

4. Locomotion activity

In locomotor activity score test, highest activity was achieved in control as 154.31±0.39* whereas lowest

activity score as found in Imipramine treated group as 95.12±0.53** in 10 min. Activity score was exhibited as 132.40±0.67** and 103.29±0.65** at the dose of 200mg/kg and 400mg/kg respectively, of HLE of *Clitoria ternatea*. It significantly decreased locomotion activity score at both the doses- proving itself a better anti-depressant and anxiolytic moiety.

Table 4.4: Locomotor activity score.

Treatment	Locomotor activity score (Sec ± SEM)
Normal saline	152.61±0.30*
Imipramine (10mg/kg)	97.15±0.56***
HFE of <i>Hippeastrum vittatum</i> (200mg/kg)	134.41±0.47*
HFE of <i>Hippeastrum vittatum</i> (400mg/kg)	102.37±0.91***

Significance Level= *

Values were given in Mean ± S.E.M. and found statistically significant at P<0.05, compared to control (n=6)

In results, in all the models, hydroalcoholic flower extract of *Hippeastrum vittatum* significantly demonstrated anxiolytic and antidepressant potential in both the doses when compared to control.

Its effect was about similar and near to standard drug treated group. It indicates that actions might be similar to Imipramine, Nitrazepam etc. It exhibits antidepressant action probably by facilitating the release of neurotransmitters i.e., serotonin, dopamine. It also increases the release of GABA (Gamma Amino Butyric Acid) and chloride ions influx that leads to hyperpolarization. The effect was determined in dose-

dependent manner. When compared to the control, the hydroalcoholic flower extract (HFE) showed a considerable reduction in immobility period. The plant's successful antidepressant action prompted a futuristic attempt to bypass the blood-brain barrier by blocking P-gp. In contrast, this study confirmed anxiolytic and antidepressant activity in both the doses without targeting the significant and selective constituent produced potential.

CONCLUSION

In conclusion, hydro-alcoholic flower extract (HFE) of *Hippeastrum vittatum* is significant anxiolytic and antidepressant herbal drug. It can be effectively used in the treatment of depression, mental agitation and other

neurological disorders after successfully evaluating mechanism of action of the plant- leaf.

As anxiety and depression has become commonest form of mental disability, so it may demonstrate an economic and pharmacological impact in modulating behavioral of humans.

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Conflict of Interest

None.

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