

Available online on 15.06.2019 at <http://jddtonline.info>

Journal of Drug Delivery and Therapeutics

Open Access to Pharmaceutical and Medical Research

© 2011-18, publisher and licensee JDDT, This is an Open Access article which permits unrestricted non-commercial use, provided the original work is properly cited



Open Access

Research Article

Aphrodisiac activity of hydro-alcoholic extract of *Celosea argentea* dried seeds in male rats

Yadav Nishigandha*, Kolhe Swati, Tembhurne Sachin

Department of pharmacology, AISSMS college of Pharmacy, Savitribai Phule Pune University, Pune, Maharashtra, India

ABSTRACT

Objective: The study was aimed at to investigate the aphrodisiac activity of hydro-alcoholic extract of *Celosea argentea* dried seeds (Amaranthaceae) in male albino rats in different doses. **Material and methods:** The animals were selected in the present investigation on the basis of their performance in the Copulatory arena and runway apparatus. After selection animal (n=6), they were treated with extract of *Celosea argentea* (200 and 400 mg/kg) and sildenafil citrate (5mg/kg) orally. While the control animals were given with normal water. All the treatments were given for 21days. Sexual motivation and mating behavior parameters in male rats were monitored on 11th and 21st day of treatment pairing with receptive females. After termination of drug treatment the parameter such as sexual motivation, mating behavior, serum testosterone level, histological examination of testes, relative organ weight and body weight percent were evaluated. **Result:** The hydro-alcoholic extract of *celosia argentea* seeds showed a significant increase in mating behavior, serum testosterone levels, testes- body weight ratio as compared to vehicle control, while at the dose of 400mg/kg of *Celosea argentea* seeds extract assume closer resemblance of above parameters with the standard used. **Conclusion:** The results of the study strongly suggest that the seed extract of *Celosia argentea* have good aphrodisiac activity.

Keywords: Aphrodisiac, *Celosea argentea*, sexual motivation, mating behavior, Serum testosterone level

Article Info: Received 21 April 2019; Review Completed 29 May 2019; Accepted 01 June 2019; Available online 15 June 2019



Cite this article as:

Yadav N, Kolhe S, Tembhurne S, Aphrodisiac activity of hydro-alcoholic extract of *Celosea argentea* dried seeds in male rats, Journal of Drug Delivery and Therapeutics. 2019; 9(3-s):296-302 <http://dx.doi.org/10.22270/jddt.v9i3-s.3011>

*Address for Correspondence:

Miss. Nishigandha Subhash Yadav, Department of pharmacology, AISSMS college of Pharmacy, Kennedy Road, Near R.T.O, Pune – 411001, Maharashtra, India.

1. INTRODUCTION

An agent (food or drug) that arouses sexual desire is called Aphrodisiac. Aphrodite is derived from Greek goddess of sexual, love and beauty¹. Erectile dysfunction (ED) has been one of the most common complaints among men with sexual health issue². Erectile dysfunction has been identified as the persistent inability to attain and maintain penile erection sufficient for satisfactory sexual performance³. The prevalence rate of ED in Asia ranged widely from 2% to 88%⁴. Today, Drug therapy mainly focuses on phosphodiesterase type 5 inhibitors which increase the levels of cGMP in the cavernosal vasculature leading to facilitation and prolongation of penile erection⁵. WHO estimates that up to 80% of people still rely mainly on traditional remedies⁶.

In Indian system of medicine, several plants are claimed to possess aphrodisiac potential^{7,8}. *Celosia argentea* is medicinal important plant, belong to family amaranthaceae. In India, it is found to be grown as a weed of bajara fields. It is an herbaceous erect and branching

plant⁹. Seeds of *Celosia argentea* are called 'semen celosiae'. According to traditional Chinese medicinal system, seeds are reputed for purging hepatic pathogenic fire to improve eyesight and it is also used for hepatoprotection, anti-diarrhea, anti-diabetics, antioxidant etc¹⁰.

Based on the literature finding and its claim in Indian system of medicine¹¹. Our study was undertaken to evaluate aphrodisiac properties of the *Celosia argentea* seeds extract on the male rat.

2. MATERIAL AND METHOD

2.1 Plant material

The seeds of *Celosia argentea* (CA) were collected from vita, district of sangli, Maharashtra and authenticated by M/s. shamantak enterprises, pune. Certificate of Aunthentication number of *Celosia argentea* is SE/AC/2018/04.

2.2 Preparation of extract

The seeds of CA were collected from the mature plants, shade dried and powdered (80mesh). The powdered seed (253g) was defatted with petroleum ether and later extracted (soxhlet) using 90% ethanol. The solvent free alcoholic extract was suspended in 0.75% sodium carboxy methyl cellulose (CMC) and employed for aphrodisiac activity.

2.3 Chemicals and Reagents

Estradiol benzoate (EB) AR (Analab Fine Chemicals, Mumbai), Progesterone (P) AR (Himedia Laboratories Pvt. Ltd., Mumbai), Ketamine Hydrochloride Injection (Themis Medicare Ltd. Uttarakhand), Xylazine Hydrochloride Injection (Indian Immunological Ltd., Hyderabad), Sildenafil citrate tablet (Suhagra-25, CIPLA Ltd, solan), Framycetine cream (Safoni India Ltd., Goa), Anesthetic Ether- I.P. (Nandkrishna chemicals PVT LTD, Nardana, Dist- Dhule).

2.4 Animals

Albino rats weighing around 200-250g of either sex housed in standard conditions of temperature ($22\pm 2^{\circ}\text{C}$), relative humidity ($55\pm 5\%$), were used. Animals were acclimatized to a reversed 12h/12h light and dark cycle (lights on from 20:00 to 08:00) for 10 days before the start of experiments. Behavioral studies were carried out during the reversed dark phase (between 11:00 and 16:00 using red light for illumination). They were fed with standard pellet diet and water. The experimental protocol was approved by Institutional Animal Ethical Committee as per the guidelines of CPCSEA, Government of India (Protocol No. CPCSEA/IAEC/PC-09/01-2K18).

2.5 Preparation of sexually receptive females

The ovariectomy of the female rats was done 30 days prior to start of the experiment using standard aseptic surgical techniques under deep anesthesia. Female rats anaesthetized with ketamine (90mg/kg, i.p.) and xylazine (10mg/kg, i.p.). In ovariectomy, incision made through the muscle layer and into the peritoneal cavity and ovary will be located. The fat withdrawn, the ovary separated and the oviduct ligated with surgical cotton thread or catgut¹². Reason for ovariectomy of female rat is avoid unwanted pregnancies during the course of experiment, so that they can be used for desired time period of study without any interruption in their hormonal levels, which occurs during their pregnancy period.

2.6 Induction of behavioral estrous:

Overictomized (Ovx) rats were subcutaneously (sc) injected with $10\mu\text{g}/100\text{g}$ body weight estradiol benzoate (in 0.1ml sesame oil) and $500\mu\text{g}/100\text{g}$ body weight of progesterone (in 0.1ml propylene glycol) before 48hrs and 05 hrs of testing respectively on 11th and 21st day. This treatment assures intense proceptivity and receptivity^{13,14}.

2.7 Equipment used:

Automated Runway Apparatus and Copulatory Arena (VJ Instruments, India)

2.8 Experimental Procedure:

Experiment 1: Motivation performance in Automated Run-Way Apparatus

The Automated run-way apparatus (VJ instruments, India) was a straight-arm runway consisting of a startbox (25X25X20cm), an alley (160X10X20cm), and a cylindrical Plexiglas goalbox (45cm diameter X 40 cm height; Fig 1¹⁵). Removable Plexiglas doors were located between

the startbox and alley and between the alley and goalbox. Infrared photocell emitter-detector pairs were located just outside the startbox and just inside the goalbox. Interruption of the photobeam outside the startbox initiated a timer that stopped when the subject entered the goalbox. This apparatus is comparable to that used successfully by our laboratory for studying other motivating goalbox events including food within the goalbox, a removable Plexiglas partition divided the arena into two semicircular halves. Sixteen 1.2cm diameter holes drilled into the partition and spaced 8cm apart from one another allowed air to pass between the two sides. Thus, the partition prevented even minimal tactile contact between subject and target, although visual, auditory and olfactory were accessible^{14,16}.



Figure 1: Automated runway apparatus to assess sexual motivation in rats.

Performance in the automated run-way apparatus:

On 2 separate days, subjected male rats were allowed individually to explore the entire apparatus for each of the targets (empty, Male, OVX female and OVX+EB+P Female). The animals were given two exposures to the goalbox for duration of 10 minute. Baseline socio-sexual motivation of 18 male rats was measured over the next 6 days. Rats did not receive any drug treatment and each rat tested for their motivation to maintain close proximity to four different goalbox: Empty goalbox; male; OVX Female treated with vehicle (0.1ml sesame oil injected subcutaneously (sc) 48 and 24 h before testing; OVX female treated with both $15\mu\text{g}$ of estradiol benzoate and $500\mu\text{g}$ progesterone. On any given day, all 18 subjected ran for the same target in the goal box; only one trial per day per rat was conducted. Thus, rats run for each goalbox target twice during the baseline phase. The order of goalbox targets was randomly determined.

After completion of above trial phase in automated run way apparatus, male rats were divided into three groups each consisting of six rats. Group-1: Received distilled water (normal control)p.o; Group-2 and 3: received low and high doses of extract of CA 200 and 400 mg/kg p.o. All the treatments were given continuously for 21 days and re-tested in the runway apparatus for their motivation approach to expose towards four goalbox targets (empty, male OVX female, OVX+EB+P female) and their run time (time started from sensor#1 to stopped when the animal triggered sensor #2), proximity time (Sensor #2 and #3 allowed for measurement entry), retreats (Complete return to the startbox after the rat had entered the goalbox) were measured for the period of 03 minutes on 11th and 21st. The entire runway apparatus was cleaned with a 10% ethanol solution at the end of each trial.^{14,16}

Experimental 2: Matting behavior test in Copulatory Arena

The test was carried out by the methods of Agmo (1997)¹² and Dewsbury and Dvis Jr (1970)¹⁷. The Copulatory arena is a rectangular wooden with transparent Plexiglas, illuminated red light. Healthy male rats showing brisk sexual activity were selected for the study. They were divided into 4 groups of 6 animals each and kept singly in separate cages during the experiment Group-1 represented the control group, which received 10ml/kg of distilled water orally. Groups 2 and 3 received extract of CA at doses of 200 and 400 mg/kg p.o, respectively, daily for 21 days. Group 4 served as standard and was given Sildenafil citrate (Viagra tablets) orally at dose of 5mg/kg. The receptiveness of the female rats was confirmed before the test by exposing them to male rats. Female rats with maximum receptivity were selected for the experiment.

In the test, male rat was first placed into Plexiglas box to be habituated to environment for 5 min. Then, a sexually receptive Ovx female rat which had previously treated with subcutaneous injection of 100µg/ml of estradiol benzoate and 500µg/ml of progesterone. The sexual behavior of the male rats was observed for 30 min. The tests for sexual desire was carried out after drug treatments on 11th and 21st day. The sexual behavior observed in the dark phase of the illumination cycle. The following parameters of sexual mating behavior were recorded: number of mounts before ejaculation or mounting frequency (MF), number of intromission before ejaculation or intromission frequency (IF), time from the introduction of female into the cage of male up to the first mount or mounting latency (ML), time from the introduction of the female up to the first intromission by the male or intromission latency (IL), time from the first intromission of series up to the ejaculation or ejaculatory latency (EL), time from the first ejaculation up to the next intromission by the male or post-ejaculatory interval (PEI). The values of the observed parameters for control and experimental groups were recorded.

2.9 Hormonal analysis

After the mating behavior analysis, the blood was collected from each animal from retro orbital venous. Blood samples were centrifuge at 2500 rpm for 10 minutes. The serum

samples were separated to measure the concentration of testosterone level. Total testosterone was measured by a CLIA- Fully Automated chemiluminescence system e411(ROCHE Diagnostics, Germany).¹⁸

2.10 Effect on Relative organ - body weight ratio

After the mating behavior analysis, all the male animals were sacrificed under anesthetic ether and testis, seminal vesicles, epididymis, vas-deference, penis and prostate glands were carefully removed and weighed using digital balance (shimadzu 120). The relative organ body weight ratio of was determined^{19,20}.

$$\text{Relative organ weight} = \frac{\text{absolute organ weight}}{\text{body weight at sacrifice}} \times 100$$

2.11 Histopathological examination of Testis

After decapitation, testis from each rats were collected, rinsed in 0.9% saline and fixed in 10% formalin. After fixation, these tissues were trimmed and processes. Tissue processing was done to dehydrate in ascending grades of alcohol, clearing in xylene and embedded in paraffin wax. Paraffin wax embedded tissue blocks were sections at 3-5 micrometer thickness with rotary Microtome. Slides were stained with Hematoxylin and eosin (H and E) stain. The prepared slides were examined under microscope.

2.12 Statistical analysis

Statistical analysis was done by using ANOVA followed by post-hoc tukey test to compare significant difference by using graph pad prism version 7.

3. RESULTS

3.1 Effect *celosia argentea* seed extract on run time in automated run way apparatus:

As shown in table 1, The 11 day's treatment with CA-400mg/kg showed significant reduction in runtime for OVX female as well as OVX+ EB+P female target. Whereas CA-200mg/kg was effective towards OVX+EB+P female target only. The results of 21 day's pretreatment with CA 200 and 400mg/kg were showed significant reduction in towards OVX female treated with estradiol and progesterone as well as OVX female.

Table 1: Effect of *Celosea argentea* seed extract on Run time of male rats for four targets.

Treatment (mg/kg)		Run time (Seconds)			
		Empty target	Male target	OVX Fmale	OVX + EB+P Female
After 11 days treatment	Control (vehicle 10ml/kg	32±0.98	28.66±0.21	26.66±0.21	22.5±0.56
	CA-200mg/kg	31.33±0.49	28.16±0.30	25.66±0.49	19.66±0.42**
	CA- 400mg/kg	31±0.57	27.83±0.30	25.16±0.30*	18.66±0.55***
After 21 days treatment	Control (10ml/kg vehicle)	31.83±0.87	28.33±0.21	26.5±0.22	22.16±0.40
	CA- 200mg/kg	30.5±0.34	27.66±0.21	24.83±0.30*	19±0.44***
	CA-400mg/kg	29.5±0.54	27±0.36**	24.33±0.62**	15.16±0.31***

Values are expressed a mean±SEM (n=6). One way ANOVA followed by the Tukey-Kramers Test *P<0.05, **P<0.01, ***P<0.001 compared with vehicle-treated animals.

3.2 Effect *celosia argentea* seed extract on Proximity time in automated run way apparatus:

The results of the study demonstrated that, after 11 days of treatment, *celosia argentea* at doses of 200 and 400mg/kg significantly improved the proximity time (P<0.01, P<0.001)

for two target i.e. OVX female and OVX+EB+P treated female while the results of 21 days of treatment showed significant (P<0.05, P<0.01, P<0.001) improvement of proximity time for almost all four targets compared to vehicle control group (Table 2).

Table 2: Effect of *Celosea argentea* seed extract on Proximity time in automated run way apparatus.

Treatment		Proximity time (seconds)			
		Empty target	Male rat	OVX Female	OVX+EB+P
After 11 days treatment	Control (vehicle 10ml/kg)	77.33±1.28	82.66±2.36	94.66±0.33	101.5±0.76
	CA- 200mg/kg	85.66±2.26	83.16±2.42	98±0.36**	106.33±0.49**
	CA-400mg/kg	78.5±2.20	83.66±2.53	97.5±0.84**	109.16±1.138***
After 21 days tretment	Control (10ml/kg vehicle)	80.16±1.49	83.16±2.18	96.66±0.61	102.2±0.86
	CA- 200mg/kg	92.5±1.85**	85.16±2.21	100±0.47**	109±0.70***
	CA- 400mg/kg	88.16±2.52*	90.66±0.88*	101.83±0.54***	118.6±0.74***

Values are expressed a mean±SEM (Standard Error Mean) (n=6). One way ANOVA followed by the Tukey-Kramers Test *P<0.05, **P<0.01, ***P<0.001 compared with vehicle-treated animals.

3.3 Effect *celosia argentea* extract on Retreat in automated run way apparatus:

The results of the study indicates that, 11 days treatment with CA-200mg/kg showed significant decreased the retreats in two target (ovx female as well as OVX+EB+P

female) while at 400mg/kg there was decreased in the retreat only in one target (OVX_EB+P female). The results of 21 days treatment, with *Celosia argentea* at all doses significant decreased the retreats (P<0.05, P<0.01, P<0.001) in two goalbox targets (Ovx female and Ovx+EB+P females) as compared to vehicle control (Table 3).

Table 3: Effect of *Celosea argentea* seed extract on Retreats in automated run way apparatus

Treatment (mg/kg)		Retreat (number)			
		Empty target	Male target	OVX female	OVX+EB+P female
After 11 days treatment	Control (vehicle 10ml/kg)	10.66±0.33	8.66±0.33	6.66±0.21	6.33±0.21
	CA-200mg/kg	9.83±0.16	7.66±0.21	5.66±0.33*	4.66±0.33**
	CA-400mg/kg	10±0.25	7.83±0.75	6.33±0.21	4.83±0.30**
After 21 days treatment	Control (vehicle 10ml/kg)	10.5±0.22	8.66±0.21	6.83±0.16	6.33±0.21
	CA-200mg/kg	10±0.25	7.66±0.22	5.83±0.30*	4.66±0.22***
	CA-400mg/kg	9.83±0.16	8.5±0.22	5.5±0.22**	4.33±0.28***

Values are expressed as mean±SEM (n=6), One way ANOVA followed by Tukey-Kramer Test *P<0.05, **P<0.01, ***P<0.001 compared with vehicle control animals.

3.4 Effect of *Celosia argentea* extract on mating behavior in Copulatory Arena

A mating behavior study revealed that continuous administration for 21 days, all the doses of CA extract were able to significantly decrease the mount and intromission latencies, when compared to vehicle control and standard drug-treated rats. It also significantly decreased the ejaculatory latency and post ejaculatory interval. Ultimately

it resulted in an increased of mounting frequency and intromission frequency in comparison to control and standard drug. All the doses of CA extract were followed by dose-dependent progression on 11th and 21st day interval consecutively on mating behavior of male rats as shown in table 4. There was an overall increase in the sexual behavior parameters in CA-extract treated groups of rats as increased in MF and IF, and reduction in ML, IL, and PEI. These results were also statistically significant.

Table 4: Effect of *Celosia aregentea* seed extract on mating behavior in Copulatory Arena

Mating behavior	Control		CA-200mg/kg		CA-400mg/kg		Sildenafil Citrate 5mg/kg	
	On 11 th day	On 21 st day	On 11 th day	On 21 st day	On 11 th day	On 21 st day	On 11 th day	On 21 st day
ML	3.44±0.24	3.37±0.13	3.09±0.13 ^{ns}	2.89±0.14 ^{ns}	2.75±0.20 ^{ns}	2.63±0.26*	2.23±0.18**	1.34±0.09**
IL	3.87±0.21	3.67±0.15	3.12±0.12*	2.91±0.14*	2.79±0.20**	2.64±0.25**	2.31±0.12***	1.44±0.15***
EL	9.10±0.04	8.99±0.09	8.18±0.07*	8.01±0.11**	8.17±0.3*	8.03±0.27**	5.79±0.21***	4.76±0.19***
PEI	5.43±0.19	5.61±0.18	4.88±0.2 ^{ns}	4.66±0.19*	4.47±0.33*	4.33±0.31**	2.93±0.19***	2.62±0.17***
MF	21.5±1.17	22±0.93	25.66±0.66 ^{ns}	29.66±0.65***	24±0.57 ^{ns}	27.66±1.05**	28.66±1.02***	38.5±1.6***
IF	19.66±1.22	20.33±0.76	24.16±0.79*	27.83±0.6***	23.5±0.56*	26.16±0.79**	27.16±1.01***	37.33±1.76***

All the values expressed as Mean±SEM (N=6); significant difference from control, One way ANOVA followed by Tukey-Kramers Test (*P <0.05, **P< 0.01, ***P<0.001) ns= Not significant, ML= Mount Latency, IL= Intromission Latency, EL= Ejaculation Latency, PEI= Post Ejaculation Latency, MF= Mount Frequency, IF= Intromission Frequency.

3.5 Effect of *Celosia argentea* seed extract on serum testosterone level

The *Celosia argentea* extract shows significant ($P < 0.01$, $P < 0.001$) effect on serum testosterone level as compared to

vehicle control animals (Fig 2). There was dose dependant increased in the level of serum testosterone drug treated animals. *Celosia argentea* at 400 mg/kg produce more increased in serum testosterone which was nearly equal to the value of testosterone of standard Sildenafil citrate.

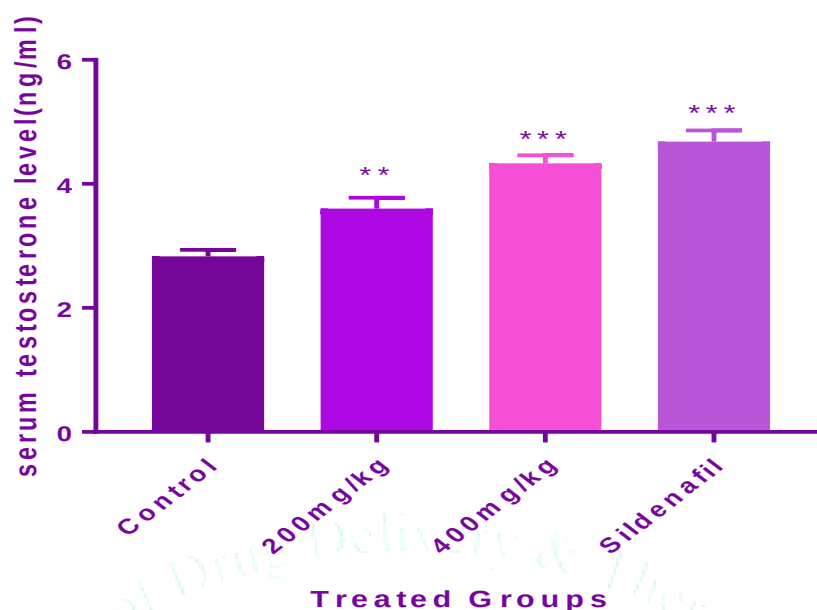


Figure 2: Effects of *C. argentea* extract on serum testosterone level in male rats. All values were expressed as mean $\pm$ SEM ($n=6$); One way ANOVA followed by the Tukey-Kramer's, ** $P < 0.01$, *** $P < 0.001$ considered significant as compared to vehicle control

3.6 Effect of *Celosia argentea* seed extract on reproductive organ weight

The result of the study indicates to significant increased the body weight of all treated animals as compared to vehicle

control. The relative weight of reproductive organ like testis and epididymis and seminal vesicle ($P < 0.05$, $P < 0.01$, $P < 0.001$) increased significantly (Table 5). The significant increase in the weight of reproductive organs was also observed in standard Sildenafil citrate treated group.

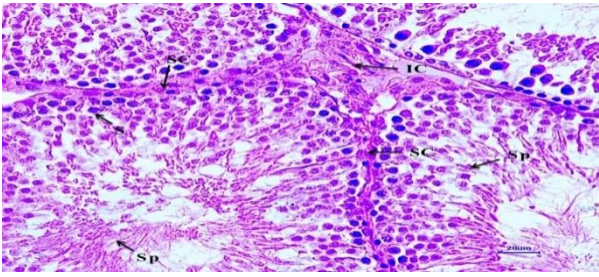
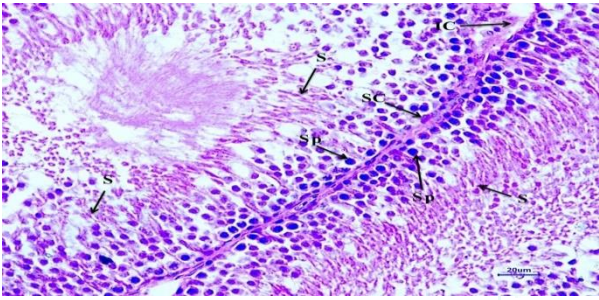
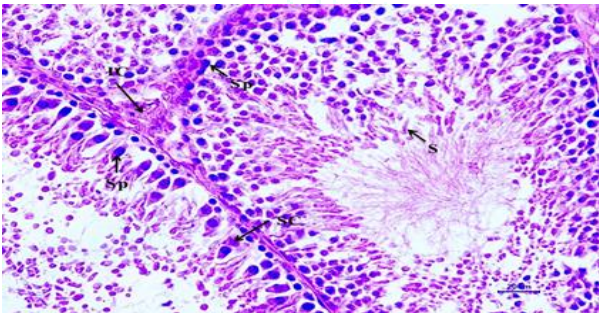
Table 5: Effect of *Celosia argentea* seed extract on relative body weight and reproductive organ weight ratio.

Treatment groups	Doses (mg/kg body wt.)	Relative organ body weight (%)					
		Testis	Epididymis	Seminal vesicle	Prostate gland	Vas-deferens	Penis
Control	Vehicle 10ml/kg	1.22 $\pm$ 0.158	0.50 $\pm$ 0.05	0.33 $\pm$ 0.01	0.10 $\pm$ 0.01	0.17 $\pm$ 0.008	0.13 $\pm$ 0.007
Group-1	200 mg/kg	1.74 $\pm$ 0.05*	0.68 $\pm$ 0.008*	0.33 $\pm$ 0.04 ^{ns}	0.13 $\pm$ 0.02 ^{ns}	0.22 $\pm$ 0.018**	0.16 $\pm$ 0.003**
Group-2	400 mg/kg	2.06 $\pm$ 0.2**	0.7 $\pm$ 0.03*	0.44 $\pm$ 0.01 ^{ns}	0.17 $\pm$ 0.01*	0.18 $\pm$ 0.01 ^{ns}	0.15 $\pm$ 0.002*
Group-3 Sildenafil citrate	5 mg/kg	1.88 $\pm$ 0.04**	0.83 $\pm$ 0.04***	0.46 $\pm$ 0.013*	0.17 $\pm$ 0.02*	0.19 $\pm$ 0.01 ^{ns}	0.15 $\pm$ 0.003*

Values are considered a Mean $\pm$ SEM (Standard Error Mean) ($n=6$), One way ANOVA followed by Tukey-Kramer's Test * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$, when compared with control, ns- non significant.

3.7 Histology Analysis:

Histopathology examination of testes from treated and vehicle control animal did not reveal any lesion or pathology significance at sertoli and spermatogonial cells of testes.

Treatment Groups	Decription
Control 	Control: Testes: Showing normal histology of seminiferous tubule, Spermatogonial cell (S), Spermatid (SP), Interstitial cells (IC), Sertoli Cell (SC) {H &E, 400X}
Group 1 (200mg/kg) 	Test Drug 200 mg/kg: Testes: Showing normal histology of Seminiferous tubule, Spermatogonial cell (S), Spermatid (SP), Interstitial cells (IC), Sertoli Cell (SC) {H &E, 400X}
Group 2 (400mg/kg) 	Test Drug 400 mg/kg: Testes: Showing normal histology of seminiferous tubule, Spermatogonial cell (S), Spermatid (SP), Interstitial cells (IC), Sertoli Cell (SC) {H &E, 400X}

4 DISCUSSIONS

The objective of present investigation was to assess the effect of *Celosea argentea* seed extract on various sexuality aspects such as: Sexual motivation and mating behavior of male rats. The first experiment successfully demonstrated the predictive validity of the runway methodology in assessing experimentally-induced changes in male sexual motivation. Treatment with *Celosea argentea* significantly increased male interest in female targets. This enhancement of sexual motivation was manifested in shorter latencies (run time) to approach female (OVX+EB+P), as well as greater amounts of time spent in close physical proximity to Female (OVX) and Female(OVX+EB+P) target located on the other side of a Plexiglas barrier (Proximity time). These two variables are behaviorally independent and are affected by the treatment with seed extract of *Celosea argentea*, from this results it indicates that the animals reflect underlying incentive-motivational processes after drug treatments. Notably, there was less motivation after treatment with *Celosea argentea* to approach an empty goalbox and male target goalbox.

The results of mating behavior study revealed that *Celosea argentea* seed extract produce dose dependant increased in MF and IF as compared to vehicle treated animals. All the doses of *Celosea argentea* seed extract also caused significant reductions in ML and IL and EL, compared with vehicle

control animals. There was highly significant decreased in ML and IL observed in animals treated with standard Sildenafil citrate. All doses of *Celosea argentea* seed extract caused significant reduction in EL and PEI as compared to vehicle control animals, while the results indicated to more significant decreased in the standard Sildenafil citrate.

The continued administration of various doses of *Celosea argentea* seed extract for 21 days demonstrated to increases in the serum testosterone levels. An increased in serum testosterone level has been associated with increase of sexual desire, penile tumescence, and rigidity as well as the accessory muscle which help to provide additional sexual activity.⁽²¹⁻²³⁾ Increased serum testosterone levels after administration of *Celosea argentea* seed extract could thus be considered as one of the contributing factors responsible for the overall increased in sexual performance. There are some possible bioactive agents responsible for increasing endogenous testosterone levels and enhancing male sexual behavior. The mechanism of these agents includes rising of androgen production,^(24,25) flavonoids by enhancing testosterone synthesis or by preventing its degradation;^(26,27) saponins by activating gonadal tissues and CNS via nitric oxide dependent mechanism.⁽²⁸⁾ Thus, the improvements in sexual function demonstrated in the current study might be due to the presence of such compounds in *Celosea argentea* seed extract.

In the present study, hydro-alcoholic extract of *Celosia argentea* seed at the dose level of 200mg/kg and 400mg/kg resulted to increase in the relative reproductive organ- body weight ratio of animals. The reported literature indicates that there is increased in the weight of reproductive organ in testosterone therapy.⁽²⁹⁾ Thus it reveals that, increased in the relative organ-body weight ratio in the present study might be due to increase in genesis of testosterone and it is an indicator for the effectiveness of the *Celosia argentea* seed extract in improving sexual motivation and mating behavior in the present investigation.

5 CONCLUSIONS

The study concluded that the cumulative dose of *Celosea argentea* extract could enhance overall sexual function and performance in male rats by increasing the level of testosterone. Thus, this study may prove to be an effective and safe alternative remedy in sexual disorders. However, further detailed studies are needed to confirm the usefulness of this plant extract in treating sexual disorders. This includes separation, purification and characterization of different chemical constituents of this extracts and testing the aphrodisiac activity of purified compounds.

ACKNOWLEDGEMENT

Authors would like to thank, The Principal, The HOD and other faculty members of Pharmacology department, AISSMS collage of pharmacy, Pune, University of Savitribai Phule pune, India for providing lab facilities and necessary atmosphere for carrying out this research work.

REFERENCES

- Singh R, Ashraf A, Jeyabalan G, Semwal A, Jaikishan. A overview of the current methodologies used for evaluation of aphrodisiac agent. *Journal of Acute Diseases*, 2013; 85-91.
- Ahn TY, et al. Prevalence and risk factors for erectile dysfunction in Korean men: results of an epidemiological study. *J Sex Med*, 2007; 4(5):1269-1276.
- Wagner G, Saenzde TI. Clinical review update on male erectile dysfunction. *Br Med J*, 1998; 316:678-682
- Park k, Hwang EC, Kim SO. Prevalence and medical management of erectile dysfunction in Asia. *Asian J Androl*, 2011; 13(4):543-549.
- Meinhardt W, Kropman RF, Vermeji P. Comparative tolerability and efficacy of treatments of impotence. *drug saf*, 1999; 20:133-146
- Arunkumar S, Muthuse vamuth. Analysis of Phytochemical constitutes and antimicrobial activities of Aloe vera against clinical pathogens. *World J. Agricultural Sci*, 2009; 5(5):572-576
- Indurwade NH, Kawtikar PS, Kosalge SB, Janbandhu NV. *Indian Drugs* 2005:67
- Patel DK, Kumar R, Prasad SK, Hemalatha S. Pharmacologically screened aphrodisiac plant- A review of current scientific literature. *Asian Pacific Journal of Tropical Biomedicine*, 2011; S131-S38.
- Hara H. *The Flora of Eastern Himalaya*. Tokyo University Press, Japan, 1966. P.407
- Ying Tang, Hai-liang Xin, Mei-li Guo. Review on research of the phytochemistry and pharmacological activities of *celosea argentea*. *Revista Brasileira de Farmacognosia*, 2016; 26:787-796
- Kirtikar KR, Basu BD. *Indian Medicinal Plants*. 2nd ed. Allahabad India: LM Basu Publishers; 1975. P.2508-2509.
- Agmo Anders. Male rat sexual behavior. *Brain Research Protocols*, 1997; 1:203-209
- Singh S, Nair V, Gupta YK. Evaluation of the aphrodisiac activity of *Tribulus terrestris* Linn. In sexually sluggish male albino rats. *Journal of Pharmacology and Pharmacotherapeutics*, 2012; 3(1):43-47.
- Lopez HH, Wurzel G, Ragen B. The effect of acute bupropion on sexual motivation and behavior in the female rat. *Pharmacology, Biochemistry and Behavior*, 2007; 87:369-379.
- Kaspate D, et al. To study an Aphrodisiac activity of hydroalcoholic extract of *withanea somnifera* dried roots in female wistar rats. *IJPSR*, 2015; 6(7):2820-2836.
- Lopez HH, Olster HD, Ettenberg A. Sexual motivation in the male rat: The role of primary Incentives and Copulatory experience. *Hormones and Behavior*, 1999; 36:176-185.
- Dewsbury DA, Davis HN, JR. Effects of reserpine on the Copulatory behavior of male rats. *Physiology and Behavior*, 1970; 5: 1331-1333.
- Sahoo HB, Nandy S, Senapati AK, Sarangi SP, Sah00 SK. Aphrodisiac activity of polyherbal formulation in experimental models on male rats. *Pharmacognosy Research*, 2014; 6(2):120-126.
- Zade V, Dabhadkar D. Evaluation of the potential aphrodisiac activity *Psoralea corylifolia* in male Albino Rats. *Asian Journal of Biomedical and Pharmaceutical Sciences*, 2013; 3(22):18-27.
- Sekar suresh, Elumalai Prithiviraj, Seppan Prakash. Dose- and time-depended effects of ethanolic extract of *Mucuna pruriens* Linn. Seed on sexual behavior of normal male rats. *Journal of Ethanopharmacology*, 2009; 122:497-501.
- Cao JF, Zhang PY, Xu CW, Huang TT, Bal YG, Chen KS, et al. Effect of aqueous extract of *arctium lappa* L. (burdock) roots on the sexual behavior of male rats. *BMC complement altem Med*, 2012; 12:1-8
- Aversa A, Fabbri A. New oral agents for erectile dysfunction: What is changing in our practice? *Asian J Androl*, 2001; 3:175-179.
- Gauthaman K, Adalkan PG, Prasad RN. Aphrodisiac proprieties of *tribulus terrestris* extract (potodiascin) in normal and castrated rats. *Life sci*, 2002; 71:1385-1396.
- Drewes SE, Gerge J, Khan F. Recent finding on natural products with erectile dysfunction activity. *Phytochemistry*, 2003; 62: 1019-1025
- Gauthaman K, Adalkan PG. The hormonal effects of *Tribulus terrestris* and its role in the management of male erectile dysfunction-An evaluation using primates, rabbit and rat. *Phytomedicine*, 2008; 15:44-54.
- Rathasooriya WD, Femando TS. Effect of black tea brew of *camella sinesis* on sexual competence of male rats. *J Ethanopharmacol*, 2008; 118:373-377.
- Yang NJ, Kaphle K, Wang P, Jang D, Wa L, Lin J, et al. Effect of aqueous extract of 'Betel Quild' and its constituents on testosterone production by dispersed mouse Intestinal cell. *Am J Chin Med* 2004; 32: 705-715.
- Murphy LL, Lec T. Ginseng, Sex behavior and nitric oxide. *Ann NY Acad Sci*, 2002; 962:372-377.
- Thakur M, Dixit VK. Effect of *chlorophytum borivilianum* on androgenic sexual behavior of male rats. *Indian drugs*, 2006; 43:300-306.