



## EVALUATION OF APHRODISIAC POTENTIAL OF ETHANOLIC EXTRACT OF ROSA DAMASCENA MILL PETALS IN RATS

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### ABSTRACT

Erectile dysfunction(ED) is a multidimensional but not unusual male sexual disorder that involves an alteration in any of the components of the erectile response, including natural, relational and psychological. Rosa damascena Mill., belongs to Family Rosaceae, is a rose hybrid, deciduous shrub which is cultivated in many parts of India. The present examination was intended to assess the aphrodisiac activity of Rosa damascena Mill. petals. Animals treated with rose petal extract showed significant difference in Mount frequency, Mount latency, Intromission frequency, Intromission latency as compared to control. It also showed significant difference in sperm count and sperm motility and protected the architecture of testis as compared stress control group. in this investigation can be reasoned that the herb Rosa damascena Mill is a protected medication and can be valuable in improving the male sexual movement and treating different sexual issue like impotency, erectile disappointment, untimely discharge, absence of sexual want and ejaculator inadequacy.

**KEYWORDS:** Aphrodisiac, Erectile dysfunction, Rosa damascena Mill.

### INTRODUCTION

Aphrodite was the goddess of love<sup>[1][2]</sup> and beauty. The present day meaning of love potion can differ, anyway is by and large viewed as a substance that will build sexual want. Male sexual dysfunction, which includes erectile dysfunction (ED) and premature ejaculation, is the most common problem that contributes to infertility, distress, relationship problems, deterioration of self-image and quality of life.<sup>[2]</sup> Erectile dysfunction(ED) is a multidimensional but not unusual male sexual disorder that involves an alteration in any of the components of the erectile response, including natural, relational and psychological. Rosa damascena Mill., belongs to Family Rosaceae, is a rose hybrid, deciduous shrub which is cultivated in many parts of India. Rose is the image of love, purity, faith and beauty since ancient times. Vapour therapy of rose oil is beneficial for headaches and migraine.<sup>[3]</sup> rose petals are acrid, aromatic, appetizer, sour, cardiogenic, expectorant, febrifuge, laxative, refrigerant and aphrodisiac.<sup>[4]</sup> Antitussive activity, Antidiabetic activity, Antimicrobial activity, Anti-inflammatory activity, Anti-depressant, Analgesic, Anti-cancer, Hypnotic effects, Anti cholinesterase activity, Anti -convulsant activity, Anti hyperlipidaemia activity, Anti-viral.<sup>[5]</sup> There was no legitimate logical information referenced with respect to aphrodisiac action of Rosa damascena Mill. petals, the present examination was intended to assess the aphrodisiac activity of Rosa damascena Mill.petals.

### MATERIALS AND METHODS

#### Collection of plant

The petals of herb Rosa Damascena mill were procured from the market of Ongole, Prakasam dist, Andhra Pradesh, India and authenticated.

#### Preparation of alcoholic extract

Shade dried petals of Rosa Damascena mill were powdered and extraction was carried using soxhlet apparatus ethanol as solvent.<sup>[6]</sup>

#### Animals

Albino rats (Mahaveer enterprises, Hyderabad, India) of either sex weighing about 200-250gm were incorporated into the investigations. The animals were maintained under standard lab conditions at an encompassing room temperature of 24±2°C having 50 ± 5% relative dampness with 12h light and dim cycle. The use and care of the animals in the experimental protocol has been endorsed by the Institutional Animal Ethics Committee (Regd. No. 1921/PO/Re/16/CPCSEA) following the guidelines of the Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA).

#### Evaluation for aphrodisiac activity

Male Albino rats weighing about 150-200 g were divided into four groups, each consisting of six rats. **Group-I:** Treated with distilled water (normal control) p.o. **Group-II:** Treated with dose of 0.7 mg/kg p.m. Sildenafil citrate

**Group-III:** Treated with lower dose of *Rosa damascena* Mill petals extract of ethanol (200mg/kg) p.o. **Group-IV:** Treated with higher dose of *Rosa damascena* Mill petals extract of ethanol (400mg/kg) p.o. All the treatments were given continuously for 14 days. On 14th day, after treatment, all animals were allowed for mating and Mount frequency, Mount latency, Intromission frequency, Intromission latency were recorded for evaluating aphrodisiac activity.<sup>[7]</sup>

#### Effect on Stress induced sexual behaviour in male rats

Male Albino rats weighing about 250-300 g were allocated into five groups and each consisting of six rats. Group-I: normal control (treated with distilled water 3ml/kg) (without stress) p.o. Group II: Stress control (treated with distilled water 3ml/kg) (with stress) p.o. Group-III: Treated with dose of 0.05mg per rat i.m Testosterone Group-IV: Treated with lower dose of *Rosa damascena* Mill petals extract of ethanol (200mg/kg) p.o. Group-V: Treated with higher dose of *Rosa damascena* Mill petals extract of ethanol (400mg/kg) p.o All the treatments were given for 28 consecutive days before they were subjected to Immobilization stress.

#### Induction of Immobilization stress

The animals were exposed to IMB stress by Plexiglas chamber (5 cm distance across and 16 cm enormous and 4 openings) for 6 hours daily during light period began from 9 am every day for 28 back to back days. Water and nourishment were evacuated during pressure period.

#### Sexual accessory Organ to body weight ratio

Body weight of each animal was measured on first day before the IMB stress and last 28th day after IMB stress and drug treatment. The percentage change in body weight was calculated. On day 28th light ether Anaesthesia animals were sacrificed. Male reproductive accessory organs like, testis, epididymis, prostate glands, vasdeferens, seminal vesicles and adrenal glands were isolated. Isolated organ are weighed to measure organ to body weight ratio.<sup>[8]</sup>

#### Sperm count

Spermatozoa were isolated by flushing the vas deferens and epididymis in 2.0ml of saline. Pipette the semen by using WBC pipette up to the mark 0.5. Pipette 4% sodium bicarbonate in 1% phenol up to the mark 11 on WBC Pipette and the dilution of 1 in 20. By using high power in the four WBC squares no of sperms were counted.

#### Calculation

Number of sperms in 1 cu.mm of sample =  $N \times 10/4 \times 20$   
 Number of sperms in 1 ml (i.e.1 cu.cm) of sample:  
 =  $N \times 50 \times 1000$ (as 1 cu.cm=1000cu.mm)  
 =  $N \times 50,000$

Where N is the total sperm count observed in outer four square of WBC chamber.

#### Sperm motility

A drop of semen was set on the spread slip and rearranged it on an edge of plasticine on the pit slide. Analysed under high power microscope objective and estimate the immobile to mobile sperms percentage.

#### Histology of testis

Testis of each group was removed by excision and cleaned with 0.9% saline smeared dry of saline and overabundance blood. They are fixed in 12% formalin for 24 hrs. The tissues, after obsession, were washed in water to evacuate abundance fixative. Washed tissues were dried out through a reviewed arrangement of Ethylalcohol, cleared with xylene and inserted in paraffin wax. Segments were cut at 3  $\mu$ m with microtone sharp edge, and mounted on clean glass slide. The segments were recolored with haematoxylin and eosin. The recolored slides were watched (400 X) in research magnifying lens and photographs were caught by utilizing camera.

## RESULTS

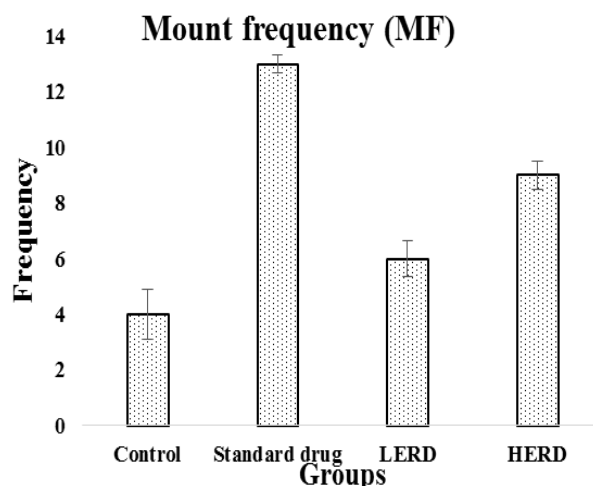
#### Evaluation of Phytochemical constituents in *Rosa damascena* Mill petals extract

From the Phytochemical screening results revealed that ethanolic extract of *Rosa damascena* Mill consists of carbohydrates, proteins, alkaloids, glycosides flavanoids and Volatile oil.

#### Aphrodisiac activity

##### Mount frequency

The results shown that a significant increase in mount frequency was observed in animals treated with ethanolic extract of *Rosa damascena* Mill petals at lower and higher concentrations when compared to control group rats.



**LERD:** Low dose of ethanolic extract of *Rosa damascena mill* (200mg/kg)

**HERD:** High dose of ethanolic extract of *Rosa damascena mill* (400mg/kg)

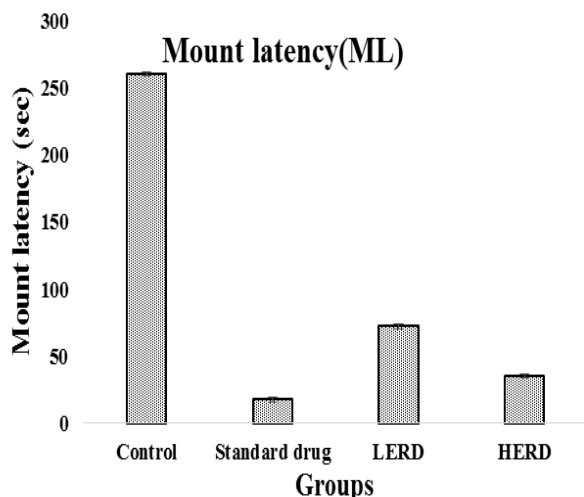
Values are Mean  $\pm$  SEM (n=6) one way ANOVA followed by Dunnett's 't' test.

Where \* represents very significant at  $p < 0.05$  when compared to control group

**Fig 1: Effect of *Rosa damascena* Mill petals extract on Mount frequency in male rats.**

**Mount latency**

Mount latency of control rats showed  $260 \pm 1.22$ . Mount latency significantly decreased in standard Sildenafil treated rats. Rats treated with *Rosa damascena* petals extract 200 and 400mg/kg. b.w showed significant decrease in mount latency as compared to control group rats.



**LERD:** Low dose of ethanolic extract of *Rosa damascena* mill (200mg/kg)

**HERD:** High dose of ethanolic extract of *Rosa damascena* mill (400mg/kg)

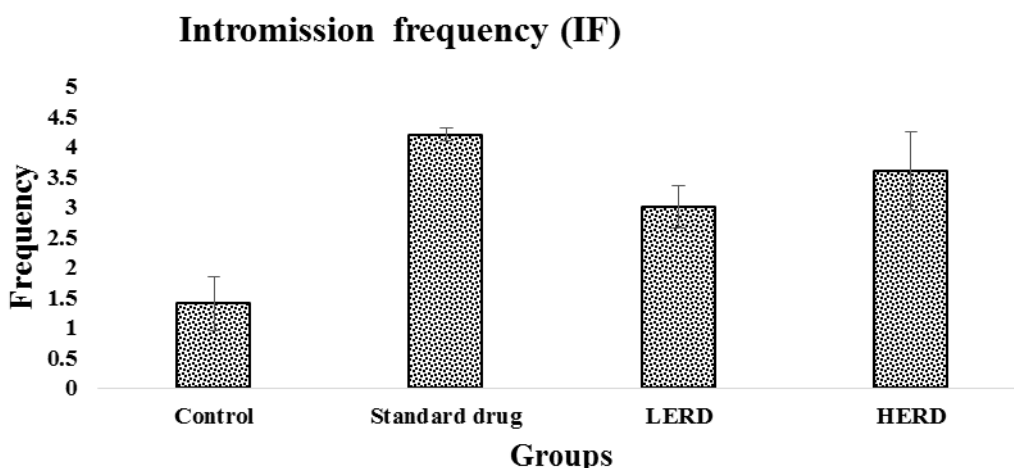
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Where \* represents very significant at  $p < 0.05$  when compared to control group

**Fig 2: Effect of *Rosa damascena* Mill petals extract on Mount latency in male rats.**

**Intromission frequency**

The results expressed that ethanolic extract of *Rosa damascena* Mill at lower and higher concentration significantly increased Intromission frequency according to the dose concentration as compared to control group rats, similar activity as that of standard drug. However, ethanolic extract of *Rosa damascena* Mill at lower dose has less intromission frequency as compared to high dose.



**LERD:** Low dose of ethanolic extract of *Rosa damascena* mill (200mg/kg)

**HERD:** High dose of ethanolic extract of *Rosa damascena* mill (400mg/kg)

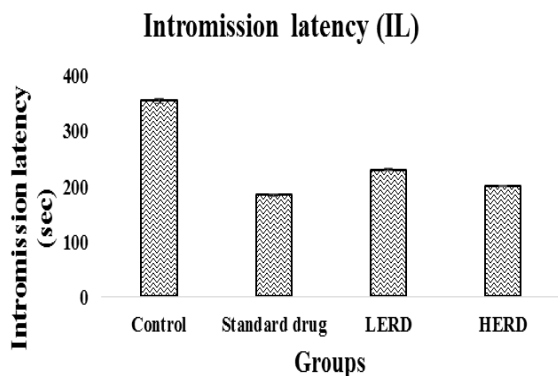
Values are Mean  $\pm$  SEM (n=6) one way ANOVA followed by Dunnett's 't' test.

Where \* represents very significant at  $p < 0.05$  when compared to control group

**Fig 3: Effect of *Rosa damascena* Mill petals extract on Intromission frequency in male rats.**

**Intromission latency**

The rats treated with standard drug and *Rosa damascena* Mill extract significantly reduced the intromission latency as compared to the control group of rats.



Values are Mean ± SEM (n=6) one way ANOVA followed by Dunnett's 't' test.

Where \* represents very significant at p< 0.05 when compared to control group

**Fig. 4: Effect of *Rosa damascena* Mill petals extract on Intromission latency in male rats.**

**Sexual accessory Organ to body weight ratio**

**LERD:** Low dose of ethanolic extract of *Rosa damascena* mill (200mg/kg)

**HERD:** High dose of ethanolic extract of *Rosa damascena* mill (400mg/kg)

Values are Mean ± SEM (n=6) one way ANOVA followed by Dunnett's 't' test.

Where \* represents very significant at p< 0.05 when compared to control group

**LERD:** Low dose of ethanolic extract of *Rosa damascena* mill (200mg/kg)

**HERD:** High dose of ethanolic extract of *Rosa damascena* mill (400mg/kg)

**Table 1: Sexual accessory Organ to body weight ratio.**

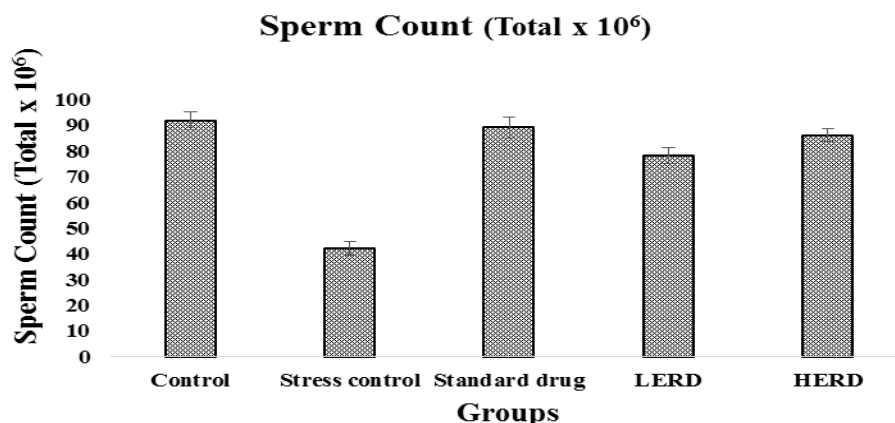
Groups	Dose	change in body weight (% in g)	Testes (mg/g)	Vasdeferens (mg/g)	Seminal vesicles (mg/g)	Adrenal Glands (mg/g)	Prostate Gland (mg/g)	Epididymis (mg/g)
Control	3ml DL	39±3.94	9.2±2.34	0.19±0.09	2.5±0.19	0.11±0.02	1.89±0.12	1.78±0.32
Stress control	3ml DL	15±2.66	4.2±1.29	0.11±0.06	1.2±0.13	0.05±0.01	0.99±0.29	0.72±0.12
Standard drug	0.7 mg/kg	20±3.12*	8.8±3.23*	0.18±0.02*	2.2±0.14*	0.09±0.02*	1.55±0.32*	1.3±0.25*
LERD	200mg/kg	21±4.82*	6.9±2.25*	0.15±0.04*	1.9±0.17*	0.07±0.01*	1.2±0.51*	0.98±0.26*
HERD	400mg/kg	19±3.33*	8.1±1.69*	0.17±0.03*	2.1±0.18*	0.08±0.02*	1.45±0.23*	1.25±0.11*

Sexual accessory Organ to body weight ratio was depicted in Table.1 Stress control group rats testis, epididymis vasdeferens, seminal vesicles, prostate glands and adrenal glands weight ratio was significantly decreased when compared Normal control. Reduction of all organ to body weight ratio was prevented significantly in testosterone, *Rosa damascena* mill treated groups.

**Effect on Stress induced sexual behaviour in male rats**

**Sperm count**

The Control group rats sperm count was found to be 91.66±3.49. The sperm count of stress control rats shown 42.22±2.66. Significant decrease in sperm count was observed in stress control rats as compared to the control group rats. A significant increase in Sperm count was observed in animals treated with standard drug and *Rosa damascena* Mill petals extract.



**LERD:** Low dose of ethanolic extract of *Rosa damascena* mill (200mg/kg)

**HERD:** High dose of ethanolic extract of *Rosa damascena* mill (400mg/kg)

Values are Mean ± SEM (n=6) one way ANOVA followed by Dunnett's 't' test.

Where \* represents very significant at p< 0.05 when compared to stress control group

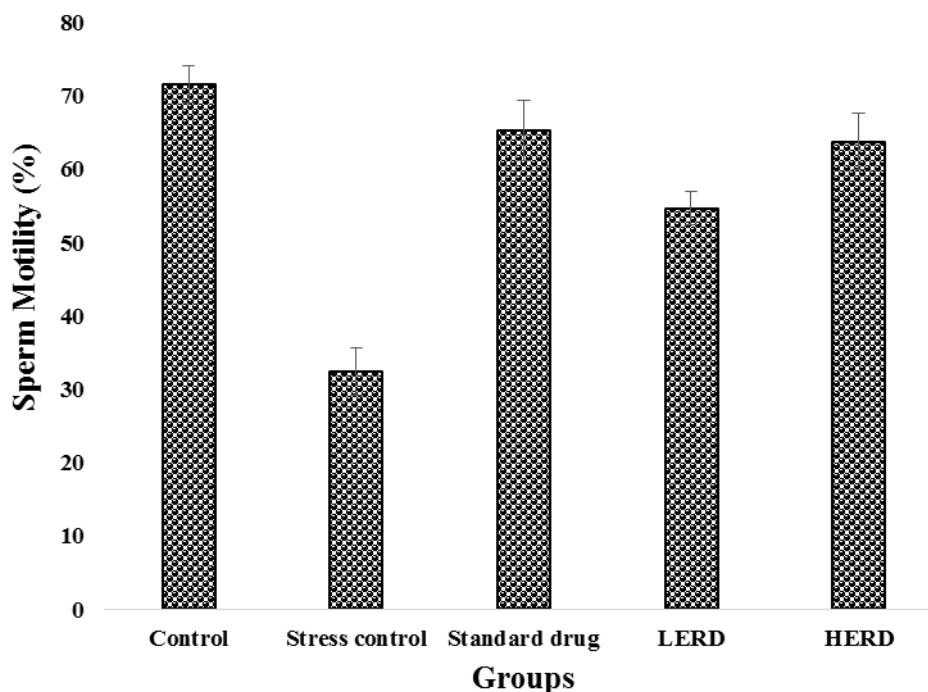
**Fig. 5: Effect of *Rosa damascena* Mill petals extract on Sperm Count (Total x 10<sup>6</sup>) in male rats.**



**Sperm motility**

The sperm motility of normal control rats was found to be  $71.6 \pm 2.49$ . A significant decrease in sperm motility

was observed in stress control rats. The rats treated with standard drug and *Rosa damascena Mill* petals showed significant increase in sperm motility.

**Sperm Motility (%)**

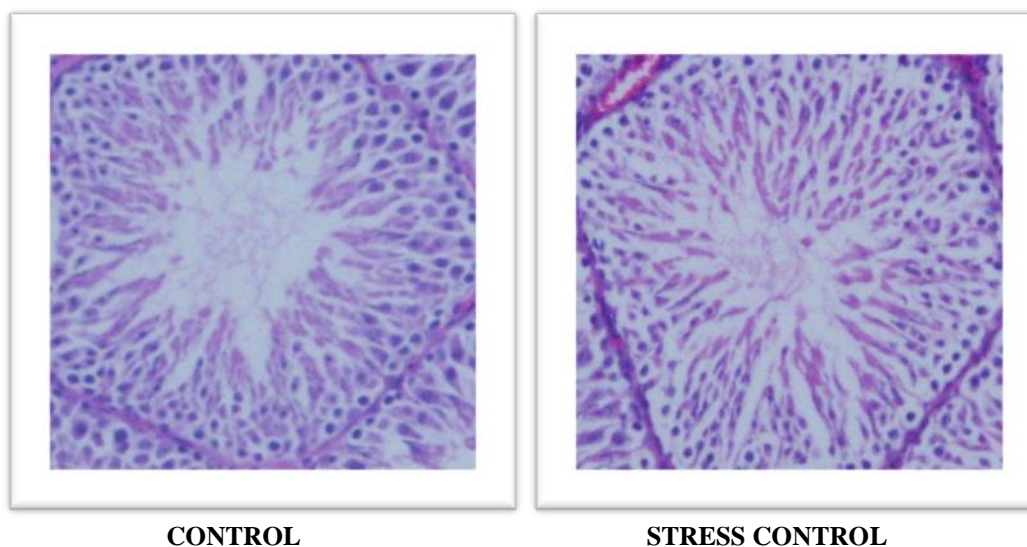
**LERD:** Low dose of ethanolic extract of *Rosa damascene mill* (200mg/kg)

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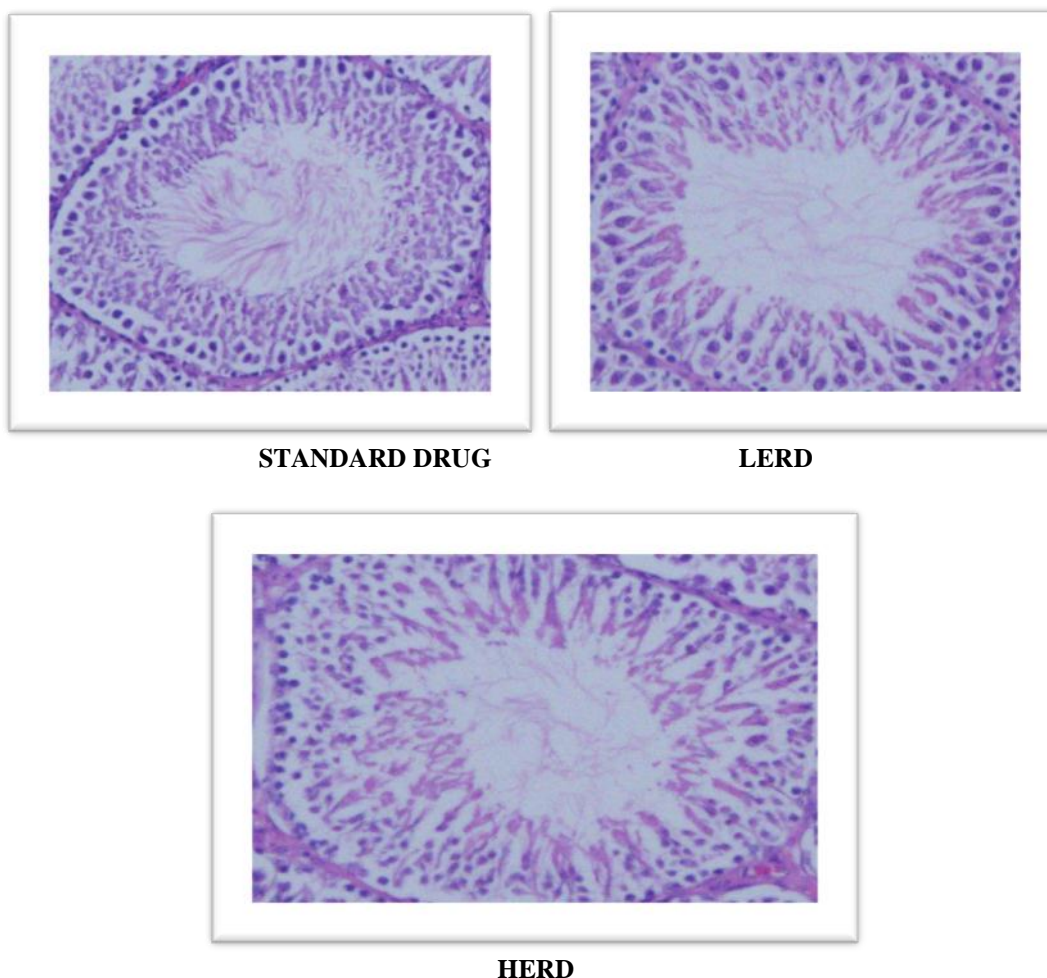
Where \* represents very significant at  $p < 0.05$  when compared to stress control group

**Fig. 6: Effect of *Rosa damascena Mill* petals extract on Sperm Motility (%) in male rats.**

**HISTOPATHOLOGY OF TESTIS**

**CONTROL**

**STRESS CONTROL**



**Fig. 7: Effect of *Rosa damascena Mill* petals extract on testis histopathology.**

### HISTOPATHOLOGY OF TESTIS

In this study, histopathology of rat testis was carried to find out the changes produced due to IMB stress for a period of 28 days.

Ordinary histology of testis was seen in typical control rats like, firmly packed seminiferous tubules and basement membrane. Spermatogenic cells and spermatozoa were found numerously. Lumen was filled with tailed sperms.

Absence of spermatozoa, basement membrane thickening, a layer of spermatogenic cells were observed in stress control group. Normal structure of basement membrane, sperm cells, maturation and dominant spermatocytes were observed in testosterone treated group. Treatment with *Rosa damascena Mill* shown thick layer of basement membrane, more number of sertoli cells and dominant spermatocytes. Maturation was arrested in cells of seminiferous tubules.

Treatment with *Rosa damascena Mill* extract to the rats protected from stress induced thin spermatogenesis layer, spermatozoa cell absence and constricted blood vessel. The treatment with this extract increased number of sertoli cells and spermatocytes.

### CONCLUSION

From results acquired in this investigation can be reasoned that the herb *Rosa damascena Mill* is a protected medication and can be valuable in improving the male sexual movement and treating different sexual issue like impotency, erectile disappointment, untimely discharge, absence of sexual want and ejaculator inadequacy. Be that as it may, further definite investigations are expected to affirm the helpfulness this plant extricate in treating sexual issue.

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