International Journal of Pharmaceutical Research and Applications
Volume 7, Issue 1 Jan-Feb 2022, pp: 1092-1104 www.ijprajournal.com ISSN: 2249-7781

Evaluation of Aphrodisiac Activity of Polyherbal Formulation (Mood ex) In Male Wistar Rat

Ms.Sreelakshmi Rajan, *MrJayachandran T. P, Mr.Soniraj V.S
Mpharm, Department of pharmaceutical sciences CPAS, Cheruvandoor
Managing Director, SPS Biosciences Ernakulam

Submitted: 05-02-2022 Accepted: 20-02-2022

ABSTRACT
Procreation was an important moral and religious issue and aphrodisiacs were sought to ensure both male and female potency. Sexual dysfunction is an inability to achieve a normal sexual intercourse, including premature ejaculation, retrograded, retarded or inhibited ejaculation, erectile dysfunction, arousal difficulties (reduced libido), compulsive sexual behavior, orgasmic disorder, and failure of detumescence. Aphrodisiacs are the agents which are used extensively by the human beings seeking to improve their sexual life and help in erectile dysfunctions (ED) sometimes called 'impotence'.The aim of the present study is to evaluate the aphrodisiac activity of poly-herbal formulation MOOD-EX in male wistar albino rats. The poly herbal formulation MOOD EX which includesGoksura, Mahamedha, Harithaki, Aswagandha, Sathavari and Pipali.Ashwagandha can stimulate production of testosterone and considered the potential aphrodisiac drug in Ayurveda.Tribulusterrestrisadministration has been reported to increase the blood levels of testosterone. The dose was determined by acute toxicity study (200mg/kg for low dose and 400mg/kg for high dose). Aphrodisiac activity was evaluated by mating behaviour test. The low dose and high dose of fraction treated group showed an increase in the mounting frequency, ejaculation frequency, copulatory rate, and intromission frequency when compared to positive control group. There was a decrease in the intromission latency, mounting latency and ejaculatory latency. There was an increase in the sperm count. The present study proposes that the polyherbal formulation [MOOD-EX] showed aphrodisiac activity in male wistar albino rat model.

KEY WORDS : Procreation,MOOD-EX, Sexual dysfunction,Copulatory rate

I. INTRODUCTON

The eternal dream of man and woman have always been the possibility of increasing, preserving and recapturing their sexual capacity, or of stimulating the sexual desire of selected members of the opposite or same sex by various means. One of the most recurrent methods has been the use of aphrodisiacs. These products, causing or increasing sexual desire or arousing sexual response, may simply be exotic food or drinks, rare herbal compounds or pharmaceuticals, amulets or psychic manipulations. Even putatively innocuous foods can be used as aphrodisiacs by suggesting or resembling sex organs (asparagus, oysters, etc.).

Human sexual arousal is a complicated phenomenon, but simple conditioning stimuli of different genres can act as potent aphrodisiacs. (1)The present study of aphrodisiac activity of the polyherbal formulation (MOOD-EX) was conducted on male wistar albino rats.The formulation includes Goksura, MahamedhaHarithaki , Aswagandha, Sathavari and Pipali. Although there are some preliminary reports about the aphrodisiac property of these individual constituents, there has been no systematic study to substantiate this combined activity.

Herbals medicinal plants have a possible to treat the assorted varieties of body ailments. The demand of herbal medicine is increasing day by day in developed yet as developing countries as a result of they are safer and well tolerated as compared to those of allopathic drugs. These plants must be subjected to animal and human studies to figure out their effectiveness in whole organism systems. Many plants have tried helpful within the management of sexual disorders throughout history, even herbs and spices are accustomed increased sexual activities in varied components of the world. There's great would like for substances that are accustomed treat sexual dysfunction in humans. The utilization of aphrodisiacs is

International Journal of Pharmaceutical Research and Applications
Volume 7, Issue 1 Jan-Feb 2022, pp: 1092-1104 www.ijprajournal.com ISSN: 2249-7781

outstanding in several countries of the world as well as Asian country like India, China, Sri Lanka, and Pakistan. (2)

II. MATERIALS AND METHODS

Polyherbalformulation - MOOD EX

INGREDIENTS

- GoksuraTribulusterrestris
- MahamedhaPolygonatumcirrhifolium
- HarithakiTerminaliachebula
- AswagandhaWithaniasomnifera
- SathavariAsparagus racemosus
- PippaliPiper longum

Polyherbal formulation MOOD EX was collected from SPS Biosciences Ernakulam in the month of February2021. The medicine is a polyherbal preparation in which 6 classic herbs have been combined in powder form. All the ingredients have been known in Indian traditional system of medicine for their aphrodisiac potential.

ANIMALS

Adult Wistar Albino rats (170 ± 20 kg) were supplied by kerala veterinary sciences Thissur. The animals were housed in large, spacious poly-acrylic cages at an ambient room temperature with 12-h light/12-h dark cycle. Rats had free access to water and rodent pellets diet. The rats were kept in animal house for ten days before starting the experiments. Ethical clearance was obtained from the Institutional Animal Ethical Committee, CPCSEA, India (Reg No.282/ac/09/CPCSEA).

ACUTE TOXIITY STUDIES – OECD 423

This method follows a step wise procedure with minimum number of animals per step. The substance is tested using a stepwise procedure, each step using three animals of a single sex. Absence or presence of compound-related mortality of the animals dosed at one step will determine the next step, i.e.

- no further testing is needed,
- dosing of three additional animals, with the same dose
- dosing of three additional animals at the next higher or the next lower dose level

Procedure

The preferred rodent species is the rat, although other rodent species may be used. Normally females are used . This is because literature surveys of conventional LD50 tests show that, although there is little difference in sensitivity between the sexes, in those cases where differences are observed females are generally slightly more sensitive (3). However if knowledge of the toxicological or toxicokinetic properties of structurally related chemicals indicates that males are likely to be more sensitive, then this sex should be used. When the test is conducted in males adequate justification should be provided.

Healthy young adult animals of commonly used laboratory strains should be employed. Females should be nulliparous and non-pregnant. Each animal, at the commencement of its dosing, should be between 8 and 12 weeks old and its weight should fall in an interval within + 20 % of the mean weight of any previously dosed animals.

The temperature in the experimental animal room should be 22ºC (+ 3ºC). Although the relative humidity should be at least 30% and preferably not exceed 70% other than during room cleaning the aim should be 50-60%. Lighting should be artificial, the sequence being 12 hours light, 12 hours dark. For feeding, conventional laboratory diets may be used with an unlimited supply of drinking water. Animals may be group-caged by dose, but the number of animals per cage must not interfere with clear observations of each animal.

The test substance is administered in a single dose by gavage using a stomach tube or a suitable intubation canula. In the unusual circumstance that a single dose is not possible, the dose may be given in smaller fractions over a period not exceeding 24 hours.

Animals should be fasted prior to dosing (e.g. with the rat, food but not water should be withheld over-night, with the mouse, food but not water should be withheld for 3-4 hours). Following the period of fasting, the animals should be weighed and the test substance administered. After the substance has been administered, food may be withheld for a further 3-4 hours in rats or 1-2 hours in mice. Where a dose is administered in fractions over a period it may be necessary to provide the animals with food and water depending on the length of the period.

Three animals are used for each step. The dose level to be used as the starting dose is selected from one of four fixed levels, 5, 50, 300 and 2000 mg/kg body weight. The starting dose level should be that which is most likely to produce mortality in,some of the dosed animals.

International Journal of Pharmaceutical Research and Applications
Volume 7, Issue 1 Jan-Feb 2022, pp: 1092-1104 www.ijprajournal.com ISSN: 2249-7781

When available information suggests that mortality is unlikely at the highest starting dose level

(2000 mg/kg body weight), then a limit test should be conducted. When there is no information on a substance to be tested, for animal welfare reasons it is recommended to use the starting dose of 300 mg/kg body weight.The time interval between treatment groups is determined by the onset, duration, and severity oftoxic signs. Treatment of animals at the next dose, should be delayed until one is confident of survival of the previously dosed animals.

Exceptionally, and only when justified by specific regulatory needs, the use of additional upper dose level of 5000 mg/kg body weight may be considered .For reasons of animal welfare concern, testing of animals in GHS Category 5 ranges (2000-5000mg/kg) is discouraged and should only be considered when there is a strong likelihood that results of such a test have a direct relevance for protecting human or animal health or the environment.

Animals are observed individually after dosing at least once during the first 30 minutes,periodically during the first 24 hours, with special attention given during the first 4 hours, and daily thereafter, for a total of 14 days,except where they need to be removed from the study and humanely killed for animal welfare reasons or are found dead

Additional observations will be necessary if the animals continue to display signs of toxicity.Observations should include changes in skin and fur, eyes and mucous membranes, and also respiratory, circulatory, autonomic and central nervous systems, and somatomotor activity and behaviour pattern. Attention should be directed to observations of tremors, convulsions, salivation, diarrhoea, lethargy, sleep and coma.

Individual weights of animals should be determined shortly before the test substance is administered, and at least weekly thereafter.

MATING BEHAVIOUR TEST

Mating behavior studies were carried out in a separate room under dim red illumination according to the standard procedure. Healthy male albino rats showing brisk sexual activity and female animals showing regular oestrus cycle were selected for the study.

Procedure

After the drug administration at various concentrations to various groups over a fixed period of time mating behaviour study is carried out.. The female rats were brought in oestrous phase by treating them with estradiolvalerate (10 µg/kg S.C. and hydroxy progesterone 1.5mg/kg S.C., for 48 hours and 5 hours prior to experimentation, respectively, to make them sexually acceptable and were selected for the study.(4)

The experiment should be conducted at 20:00h in the same laboratory and under the light of the same intensity. The receptive female animals should be introduced into the cages of male animals in the ratio of 1 female to 1 male.

The following mating behavior parameters were recorded:

(a) Mount frequency (MF): The number of mounts without intromission from the time of introduction of the female until ejaculation

(b) Intromission frequency (IF): The number of intromissions from the time of introduction of the female until ejaculation

(c) Mount latency (ML): The time interval between the introduction of the female and the first mount by the male

(d) Intromission latency (IL): The interval from the time of introduction of the female to the first intromission by the male (characterized by pelvic thrusting and springing dismount)

(e) Ejaculation latency (EL): The time interval between the first intromission and ejaculation (characterized by longer, deeper pelvic thrusting, and slow dismount followed by a period of inactivity)

(f) Post-ejaculatory interval (PEI): The time interval between ejaculation and the first intromission of the following series. The experiment was terminated when the male rat begins to mount the female followed by intromission after a brief period of inactivity (which normally results following ejaculation). The values of the observed parameters were measured at first week and third week of drug administration and compared with control.

(g) Copulatory rate: Copulatory rate was calculated by determining the number of mounts plus the number of intromission divided by the time from the first mount until ejaculation.

Copulatory rate = Number of mounts + Number of intromissions/ Time from the first mount till ejaculation

International Journal of Pharmaceutical Research and Applications
Volume 7, Issue 1 Jan-Feb 2022, pp: 1092-1104 www.ijprajournal.com ISSN: 2249-7781

(h) Index of libido : Index of libido is defined as the ratio of number mated to the number paired expressed in percentage

%Index of libido = Number mated / Number paired x 100 (5)

HEMATOLOGICAL ANALYSIS

At the end of experimental period, the next day blood was collected by chod - pap method for the analysis of serum cholesterol and testosterone.

EX VIVO STUDIES

EFFECT ON SPERM COUNT

Epidydymis of rats of each group was homogenized and taken into 5ml of 1% sodium citrate solution and squashed thoroughly with the help of needle an forceps until a milky suspension was obtained. The solution was filtered through 80 mm mesh and the volume was made up to 10ml with the same solution; the made-up volume was inclusive of washings of the filter. The suspension was shaken thoroughly and the spermatozoa were counted in five WBC counting chambers of hemocytometer. The average number of sperms per chamber is reported.

III. RESULTS AND DISCUSSIONS

ACUTE TOXICITY STUDIES

The acute toxicity study as per OECD TG 4239 (Acute toxic class method) was performed to determine the safe dose for the animal. A dose of 2000mg/kg was given and observed for 14 days. The animals exhibited normal behaviour and no signs of toxicity were detected. Their motor activity and secretory status were normal and did not present any signs of depression, tremor, salivation, lethargy, sleeplessness and coma.

The drug was safe at a dose of 2000mg/kg, so we are taking the $1/5^{th}$ and $1/10^{nth}$ of the dose as low dose and high dose.,ie, 200mg/kg as low dose and 400mg/kg as high dose.

MATING BEHAVIOUR TEST

From the mating behaviour study, we determine the different parameters like mounting frequency, mounting latency, intromission frequency, intromission latency, ejaculatory latency, post ejaculatory interval, copulatory rate, index of libido and biochemical parameters like cholesterol and testosterone level.

MOUNTING FREQUENCY

Sl no	Groups	Mounting frequency [MEAN±SEM]
1	Control	3.000±0.447
2	Standard	7.667±0.557
3	High dose	7.000±0.365
4	Low dose	5.167±0.307

Table 1

International Journal of Pharmaceutical Research and Applications
Volume 7, Issue 1 Jan-Feb 2022, pp: 1092-1104 www.ijprajournal.com ISSN: 2249-7781

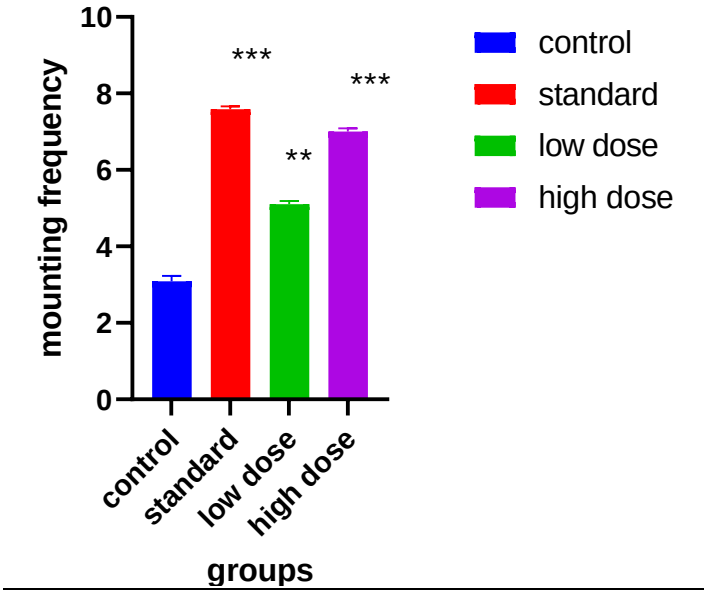

Figure1 Effect of MOOD EX on the mounting frequency of male rats. The values are expressed in Mean ±SEM(n=6). One way ANOVA followed by Dunnett's multiple comparison test.***P<0.001, **P<0.01 as compared to the control.

MOUNTING LATENCY

Sl no	Groups	Mounting latency(sec) [Mean±SEM]
1	Control	134.2±9.867
2	Standard	21.67±2.108
3	High dose	35.3±6.009
4	Low dose	41.67±4.216

Table 2

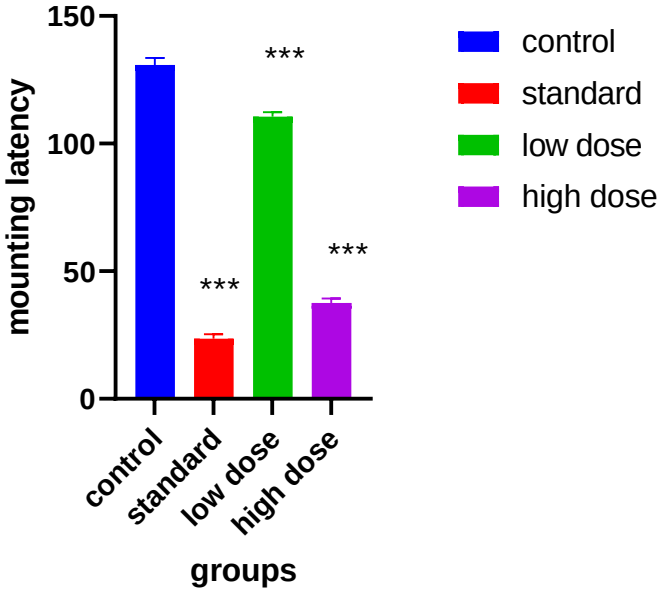

Figure 2 Effect of polyherbal formulation on mounting latency of male rats. The values were expressed in Mean±SEM(n=6). One way ANOVA followed by Dunnet's multiple comparison test.***p<0.001 as compared to control.

International Journal of Pharmaceutical Research and Applications
Volume 7, Issue 1 Jan-Feb 2022, pp: 1092-1104 www.ijprajournal.com ISSN: 2249-7781

INTROMISSION FREQUENCY

Sl no	Groups	Intromission frequency [Mean±SEM]
1	Control	3.667 ±0.333
2	Standard	10.67±0.494
3	High dose	7.667±0.421
4	Lowdose	5.667±0.666

Table 3

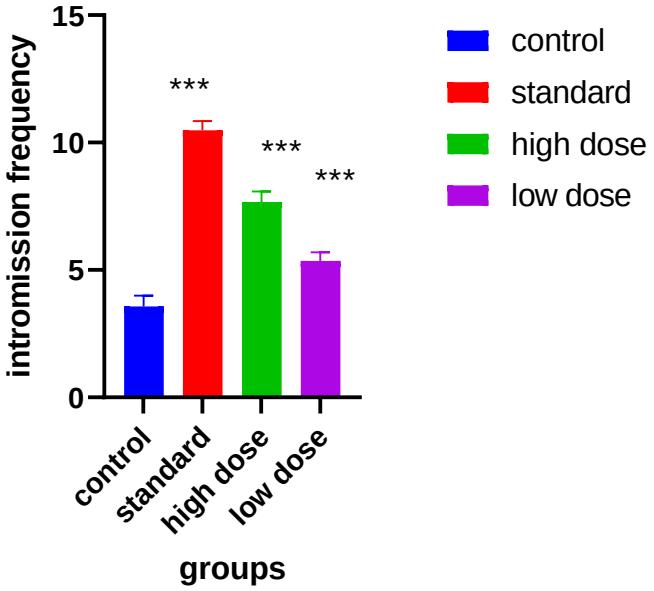

Figure 3 Effect of polyherbalfdormulatuion on intromission frequency of male rats. The values were expressed in Mean±SEM(n=6). One way ANOVA followed byDunnett's multiple comparison test. ***p<0.001 as compared to control.

INTROMISSION LATENCY

Sl no	Groups	Intromission latency(sec) [Mean±SEM]
1	Control	189.0±5.854
2	Standard	49.67±2.940
3	High dose	92.50±5.188
4	Low dose	151.2±10.12

Table 4

International Journal of Pharmaceutical Research and Applications
Volume 7, Issue 1 Jan-Feb 2022, pp: 1092-1104 www.ijprajournal.com ISSN: 2249-7781

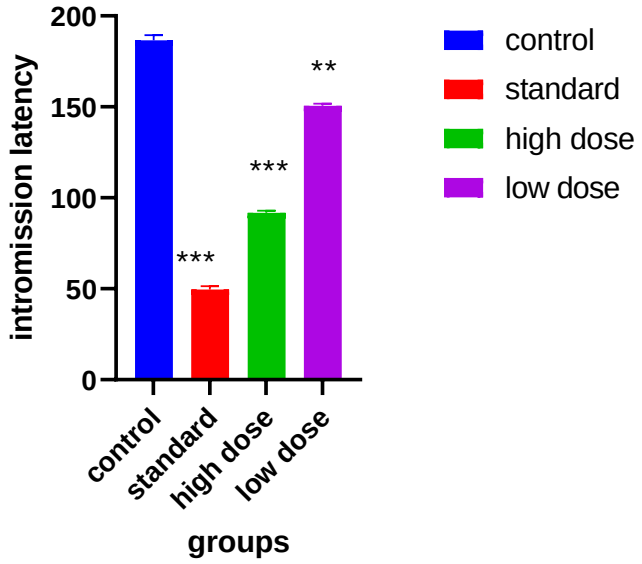

Figure 4 Effect of polyherbal formulation on intromsiion latency on male rats.The values were expressed in Mean±SEM(n=6). One way ANOVA followed byDunnett's multiple comparison test. ***p<0.001,**p<0.01 as compared to control.

EJACULATION LATENCY

Sl no	Groups	Ejaculatory Latency(sec) [Mean±SEM]
1	Control	18.00±2.503
2	Standard	147.5±7.388
3	Low dose	95.67±4.088
4	High dose	132.5±5.117

Table 5

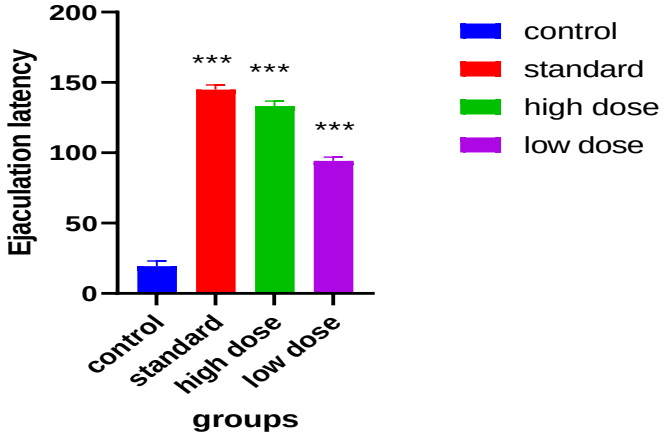

Figure 5 Effect of polyherbal formulation on ejaculation latency on male rats.The values were expressed in Mean±SEM(n=6). One way ANOVA followed byDunnett's multiple comparison test. ***p<0.001 as compared to control

International Journal of Pharmaceutical Research and Applications
Volume 7, Issue 1 Jan-Feb 2022, pp: 1092-1104 www.ijprajournal.com ISSN: 2249-7781

POST EJACULATORY INTERVAL

Sl no	Groups	Post ejaculatory interval Mean±SEM
1	Control	509.2±27.26
2	Standard	235.0±10.02
3	Low dose	417.8±15.43
4	High dose	315.2±12.14

Table 6

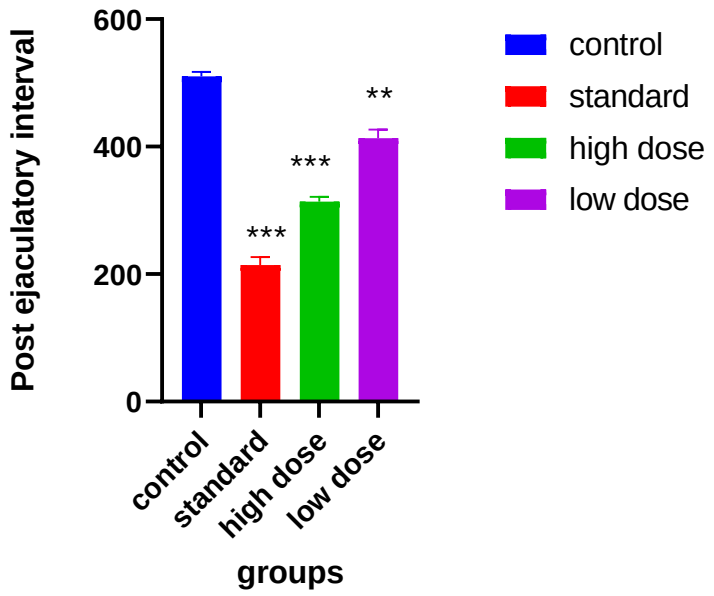

Figure 6 Effect of polyherbal formulation on post ejaculatory interval on male rats.The values were expressed in Mean±SEM(n=6). One way ANOVA followed byDunnett's multiple comparison test. ***p<0.001,**p<0.01 as compared to control.

COPULATORY RATE

The copulatory rate was calculated by determining the number of mounts plus the number of intromissions divided by the time from the first mount until ejaculation.

Copulatory rate = Number of mounts + Number of intromissions/Time from the first mount until ejaculation

Copulatory rate of control group = 3 + 3.66/341.16 = 0.019

Copulatory rate of standard groups = 7.66 + 10.66 /218.82 = 0.083

Copulatory rate of low dose groups = 5.16 +4.5 /300.32 = 0.032

Copulatory rate of high dose groups = 7+7.33/246.66 =0.058

INDEX OF LIBIDO

Index of libido is defined as the ratio of number mated to the number paired expressed in percentage.

%index of libido = Number mated/Number paired X 100

The index o libido of all groups (control, standard, low dose, high dose) is 100% because all animals in each group were paired in a ratio of 1:1.

%index of libido = Number mated/Number paired x 100 = 1/1 x 100 = 100%

International Journal of Pharmaceutical Research and Applications
Volume 7, Issue 1 Jan-Feb 2022, pp: 1092-1104 www.ijprajournal.com ISSN: 2249-7781

BIOCHEMICAL PARAMETERS
SERUM CHOLESTEROL

Table 7

Sl no	Groups	Serum cholesterol level (mg/dl) Mean±SEM
1	Control	57.17 ± 4.909
2	Standard	179.0±4.524
3	Low dose	87.00±4.227
4	High dose	135.7±5.655

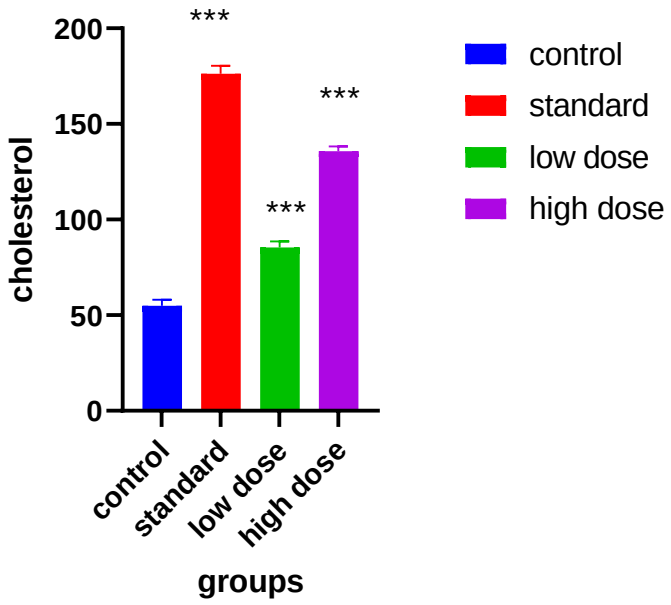

Figure 7 Effect of MOOD EX on serum cholesterol level of male rats. The values were expressed in Mean ±SEM (n=6). One way ANOVA followed by Dunnett's multiple comparison test. ***p<0.001, as compared to control.

SERUM TESTOSTERONE LEVEL

Sl no	Groups	Serum testosterone level(mg/ml)
1	Control	1.360±0.057
2	Standard	2.615±0.035
3	Low dose	1.817±0.050
4	High dose	2.255±0.0343

Table 8

International Journal of Pharmaceutical Research and Applications
Volume 7, Issue 1 Jan-Feb 2022, pp: 1092-1104 www.ijprajournal.com ISSN: 2249-7781

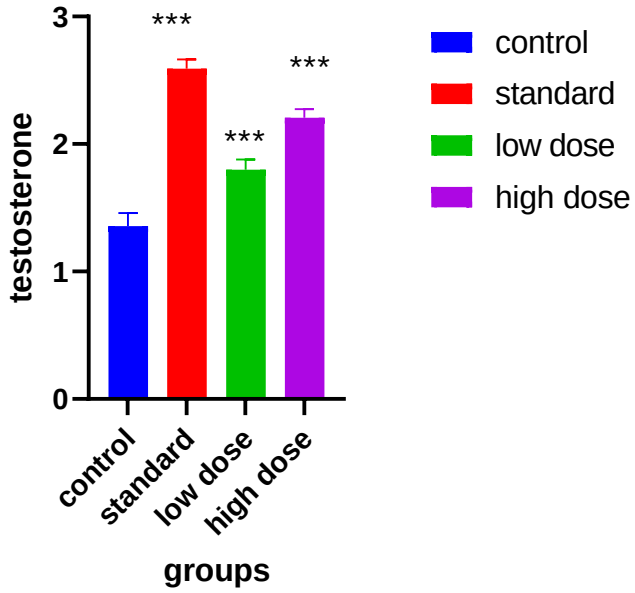

Figure 8 Effect of MOOD EX on serum testosterone level of male rats. The values were expressed in Mean ±SEM (n=6). One way ANOVA followed by Dunnett's multiple comparison test. ***p<0.001, as compared to control.

EFFECT OF POLYHERBAL FORMULATION ON SPERM COUNT

Sl no	Groups	Sperm count(x 10^6) Mean±SEM
1	Control	105.9±1.827
2	Standard	156.5±3.651
3	Low dose	120.9±1.984
4	High dose	138.2±1.951

Table 9

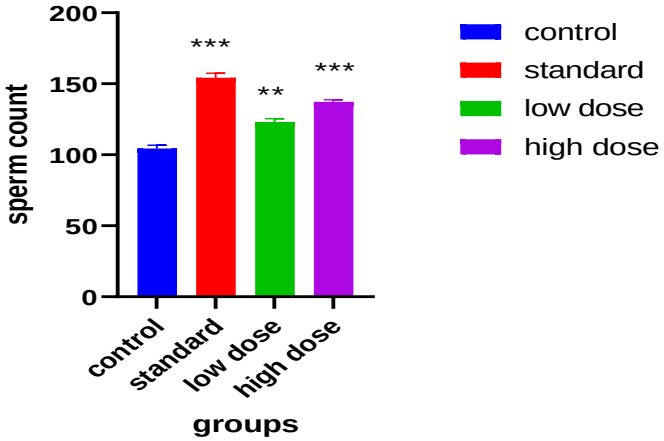

Figure 9 Effect of MOOD EX on sperm count of male rats. The values were expressed in Mean ±SEM (n=6). One way ANOVA followed by Dunnett's multiple comparison test. ***p<0.001, **p,<0.01 as compared to control.

International Journal of Pharmaceutical Research and Applications
Volume 7, Issue 1 Jan-Feb 2022, pp: 1092-1104 www.ijprajournal.com ISSN: 2249-7781

IV. CONCLUSION

Male impotence is a significant problem that may contribute to infertility function decreases spontaneously with advanced aging. It occurs commonly in middle aged and older men. Aphrodisiacs are the substances which are used to increase sexual activity and help in fertility. The present study was aimed to evaluate the aphrodisiac activity of polyherbal formulation [MOOD EX] in male wistar rats.

The invivo mating behaviour was performed to evaluate the aphrodisiac activity of MOOD EX in male wistar rats. The animals are given the test compound for 21 days and on the 21st day , the experiment is conducted at 8pm and male rats are observed with the help of a camera. From the observation, it is observed that significant increase in mounting frequencies [MF] and intromission frequencies [IF] with corresponding decrease in mount latency [ML] and intromission latency [IL] are indications that the male rats were aroused. It also reflects enhanced performance , motivation, and vigour. Also the prolonged ejaculatory latency is a strong indication that the sexual function of male rats were enhanced suggesting aphrodisiac activity.

It has been reported earlier that androgens are important modulators of ale sexual behaviour including erection and libido. These androgens may act both at the central and peripheral nervous system levels. Testosterone is one of the main androgens in the male gonads produced by the interstitial Leydig cells of the testis. An increase in testosterone had been linked with a moderate but corresponding increase in sexual desire and libido. Cholesterol is a requirement for the normal activity of testicles. Cholesterol is also a known precursor in the synthesis of the steroids including bile acids, steroid hormones, and vitamin D. An increase in testicular and/serum cholesterol concentrations led to a corresponding increase in the aphrodisiac activity of the polyherbal formulation MOOD EX. This increase in the cholesterol concentrations corroborationg the increased testosterone concentration observed.

The increase in sperm count shows that the rats were aroused. While comparing the low dose and high dose of the polyherbal formulation, the high dose possess more activity than the low dose.Overall, this study showed that the polyherbalformulation has aphrodisiac potential. The highest dose of the extract has the best aphrodisiac effect. The extract has a functional capacity to increased concentrations of testosterone and cholesterol which are possible mechanism of action for its aphrodisiac activity.

V. ACKNOWLEDGEMENT

I take this opportunity with pride and enormous gratification to express the feeling of thanks and gratefulness to all those people who have helped, encouraged and inspired me for the successful completion of this work. First in foremost I want to thank almighty which I felt by my soul that always blessed and guided me from the beginning until I finished writing this work.

With great pleasure and deep satisfaction, I express my humble gratitude and respect to my honored research guide Jayachandran T.P, Associate professor, Department of Pharmaceutical Sciences, Cheruvandoor, for his invaluable guidance, never diminishing encouragement, valuable suggestions, moral support, patience and timely help in my studies. I consider myself very much lucky to have him as my guide. This period has been an enriching experience of working under his guidance.

I extend my heartfelt gratitude to Prof. Dr.JyotiHarindran, Dr. Abdul Vahab A, Dr.Litty Joseph, Dr SibyP.Ittiyavirah and other Lecturers, Department of Pharmaceutical Sciences, Cheruvandoor, for their continuous support, valuable and generous help in carrying out my dissertation work in this institution.

I owe my sincere thanks non-teaching staffs of the department who has helped me immensely in this project. Also, I express my love and thanks to my classmates for their intense help. I wish to express my deep sense of gratitude to Mr.Soni Raj V S Managing Director, S.P.S Biosciences Ernakulam and all the staffs in Path Centre Thrissur for providing me the necessary requirements for the work.

I am forever indebted to my beloved parents and my Sister, who always stood beside us in trouble and happiness and without whose prayers, support, understanding and encouragement I wouldn't have survived till here.

Finally, I take this privilege to express my heartfelt thanks to all my near and dear ones who have been involved directly or indirectly with the successful completion of this project work.

REFERENCES

[1]. Sciences FDES. Natural aphrodisiacs . Studies of commercially-available herbal recipes , and phytochemical investigation of Erythroxylumvacciniifolium Mart . (

International Journal of Pharmaceutical Research and Applications
Volume 7, Issue 1 Jan-Feb 2022, pp: 1092-1104 www.ijprajournal.com ISSN: 2249-7781

Erythroxylaceae) from Brazil Thèse de doctorat. 2003;
[2]. Nimesh S, DhwajAshwlayan V, Barman P. Medicinal Plants as Aphrodisiac Agents: A Current Status. ActaSci Pharm Sci. 2019;3(8):137–44.
[3]. Brunetti P, Fabrizio A, Faro L, Tini A, Paolo F, Carlier J. Pharmacology of Herbal Sexual Enhancers : A Review of Psychiatric and Neurological Adverse E ffects. 2020;1–51.
[4]. Gupta RB, Ahuja A, Kabra MP, Soni S. Study of Fenugreek Effect on Aphrodisiac Activity in Diabetes Induced Rat. 2015;3(3):232–7.
[5]. Abhyankar N, Kathrins M, Niederberger C: Use of testicular versus ejaculated sperm for intracytoplasmic sperm injection among men with cryptozoospermia: a meta-analysis. FertilSteril. 2016; 105(6): 1469–1475.e1
[6]. Semwal A, Sahib P, Kumar R, Singh R, Shaheed A, Ajit B, et al. Nature ' s Aphrodisiacs -A Review of Current Scientific Literature I nternational J ournal of R ecent A dvances in P harmaceutical R esearch Nature ' s Aphrodisiacs - A Review of Current Scientific Literature. 2013;(October 2014).
[7]. Toxic potentials of ten herbs commonly used for aphrodisiac effect in Turkey. 2015;496–506.
[8]. Sharma M, Arya D, Bhagour K, Gupta RS. Natural aphrodisiac and fertility enhancement measures in males : A review Current Medicine Research and Practice Natural aphrodisiac and fertility enhancement measures in males : A review. Indian J Rheumatol [Internet]. 2017;7(2):51–8. Available from: http://dx.doi.org/10.1016/j.cmrp.2017.02.007
[9]. Mathur M. Herbal Aphrodisiac their Need , Biology and Status : Global and Regional Journal of Natural Products Herbal Aphrodisiac their Need , Biology and Status : Global and Regional Scenario. 2018;(January 2012).
[10]. Histology of testis.
[11]. Brugo-olmedo S, Chillik C, Kopelman S. Review Definition and causes of infertility. Reprod Biomed Online [Internet]. 2002;2(1):173–85. Available from: http://dx.doi.org/10.1016/S1472-6483(10)62193-1
[12]. Karavolos S, Panagiotopoulou N, Alahwany H, Authority E. An update on the management of male infertility. 2020;267–74.
[13]. Viagra D. Tablets. 5:1–30.
[14]. Singh B, Gupta V, Bansal P, Singh R, Kumar D. Pharmacological potential of plant used as aphrodisiacs. Int J Pharm Sci Rev Res. 2010;5(1):104–13.
[15]. Malaysiana S, Potensi P, Ekstrak A, Sawah B, Jantan T. Evaluation of the Aphrodisiac Potential of Rice Field Eel , Monopterusalbus Extracts in Male Mice. 2017;46(6):845–9.
[16]. Narayanswamy VB, Setty MM, Malini S, Shirwaikar A. Preliminary aphrodisiac activity of. 2007;161:152–61.
[17]. Jungwirth a, diemer t, dohle gr, giwercman a, kopa z, krausz c, et al. Guidelines for the investigation. 2012;(february):176–88.
[18]. Jedrzejowska RW, Wolski JK, Hilczer JS. The role of oxidative stress and antioxidants in male fertility. 2012;60–7.
[19]. Wilkinson JM, Halley S, Towers PA. Comparison of male reproductive parameters in three rat strains : Dark Agouti , Sprague-Dawley and Wistar . 2000;70–5.
[20]. Subramoniam A, Gangaprasad A, Sureshkumar PK, Radhika J, Arun BK. ORIGINAL ARTICLE A novel aphrodisiac compound from an orchid that activates nitric oxide synthases. 2013;(July 2012):212–6.
[21]. Barki A. Mating Behaviour. 2017.
[22]. Chhatre S, Nesari T, Somani G, Kanchan D, Sathaye S. Phytopharmacological overview of Tribulusterrestris. 2014;8(15).
[23]. Akram M, Asif HM, Akhtar N, Shah PA, Uzair M, Shaheen G. Tribulusterrestris Linn.: A review article. 2011;(February 2014).
[24]. Develop JP, Poonam B, Pratti P, Prasad NB. Polygonatumverticillatum(linn .) All . And polygonatumcirrhifolium(wall .) Royle : two threatened vital healers from asthaverga nurtured by garhwalhimalaya ,india. 2011;18:159–67.
[25]. Rathinamoorthy R, Thilagavathi G. TerminaliaChebula - Review on Pharmacological and Biochemical Studies. 2014;6(1):97–116.
[26]. Gupta GL, Rana AC. Withaniasomnifera(Ashwagandha): A Review PHCOG MAG .:

Plant Review Withaniasomnifera (Ashwagandha): A Review. 2017;(July).
[27]. Selvaraj K, Sivakumar G, Veeraraghavan VP, Dandannavar VS, Veeraraghavan GR. Review article Asparagus Racemosus - A Review. 2019;10(1):87–9.
[28]. Hasan N, Ahmad N, Zohrameena S, Khalid M. Asparagus racemosus : for medicinal uses & pharmacological Introduction : -. 2016;(March).
[29]. Pabuccu EG, Caglar GS, Tangal S, et al.: Testicular versus ejaculated spermatozoa in ICSI cycles of normozoospermic men with high sperm DNA fragmentation and previous ART failures. Andrologia.2017; 49(2).