

**PHARMACEUTICO-ANALYTICAL AND EXPERIMENTAL STUDY OF KAPARDA
BHASMA IN SPERMATOGENESIS W.S.R. TO OLIGOSPERMIA****Dr. Ashtakar Gita Narayan^{*1}, Dr. Ravi R. Chavan², Dr. Lalitha³, Dr. Laxmibai Kurle⁴**¹3rd Year Pg Scholar, ²Professor, ³Assistant Professor, ⁴Professor & HOD

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ABSTRACT

Background: In Male infertility, oligospermia characterized by reduced sperm concentration, is one of its major contributing factors. Impaired spermatogenesis may be associated with oxidative stress, altered testicular function and disturbances in the reproductive system. Ayurveda describes Shukra as an important component of the reproductive system and considers nourishment and proper formation of Shukra Dhatu essential for reproductive health. Vrishya, Rasayana and Shukrajanana approaches are therefore important in the management of disorders associated with diminished reproductive capacity. Kaparda, a calcium-containing mineral drug, is described in Rasashastra literature and is processed into Kaparda Bhasma for therapeutic application. **Objective:** The present study was undertaken to evaluate the pharmaceutical, analytical and experimental aspects of Kaparda Bhasma and to assess its potential effect on spermatogenesis in an experimental model of oligospermia. **Materials and Methods:** Kaparda Bhasma was prepared by following classical pharmaceutical procedures, including Shodhana and Marana, and subjected to classical Bhasma Pariksha and contemporary analytical investigations. The experimental study was conducted in Wistar albino rats, in which oligospermia was experimentally induced using cyclophosphamide. The effect of Kaparda Bhasma was assessed using relevant reproductive parameters, including sperm count, sperm motility, sperm morphology and reproductive organ weights. **Results:** The prepared Kaparda Bhasma exhibited characteristics consistent with properly prepared Bhasma. Experimental administration of Kaparda Bhasma showed improvement in selected sperm parameters and reproductive indices in oligospermic rats when compared with the corresponding control group. **Conclusion:** The findings suggest that Kaparda Bhasma have a beneficial influence on spermatogenesis in the experimental model of oligospermia. The study provides a basis for correlating the classical Ayurvedic concept of Vrishya action with experimentally observed Spermatogenic effects.

KEYWORDS: Kaparda, Kaparda Bhasma, Spermatogenesis, Oligospermia, Shukra, Vrishya, Wistar albino rats.**INTRODUCTION**

Ayurveda known as the 'science of life' fosters holistic wellbeing by aligning individuals with nature for physical, mental and spiritual harmony. *Rasashastra and Bhaishajya kalpana* deals with minerals, herbo-mineral/metallic and animal origin formulations with safety processing technologies having wide range of therapeutic efficacy, possessing innate qualities like less dose, better palatability and quick action.

According to Ayurveda classics Shukra is one among the saptha dhatu and Shukrakshaya^[1] is one of the eight shukradoshas leading to male infertility and caused by imbalance in vata and Pitta dosha. Also caused by various factors^[2] like jara, chinta, vyadhi, karma karshana, anashana etc.

VRUSHYA^[3] is one of the Astanga Ayurveda dealing with male potency, aimed at superlative fertility, virility & healthy progeny by tackling common issues like sexual dysfunctions, Infertility, etc. of various etiologies.

Spermatogenesis is a process of cellular differentiation resulting in the formation of functional spermatozoa for successful reproduction. Oligospermia is the condition in which sperm concentration below the limit of 20 million sperm/ml. In the last 50 years a significant decrease in the human fertility has been observed, as per WHO⁴ 60-80 million couples worldwide currently suffering from infertility and stated that 40 - 50% of infertile couples are due to the male factor^[5] like oligospermia, decrease in sperm motility and immature sperm etc.

To address such conditions our classics explained Kaparda Bhasma^[6] is potent, unexplored bhasma indicated in VRUSHYA, netraroga, karnasrava, udarashula, grahaniroga, kshayaroga etc.

Chemically Kaparda is calcium carbonate. Calcium^[7] is a significant element acts as an intracellular second messenger and necessary for many physiological processes in spermatozoa including spermatogenesis, sperm motility, capacitation, acrosome reaction and fertilization.

Animal experimental study is disciplined work to establish the efficacy of formulation in the management of particular disease without causing side effect. Aim of the present research study is to prepare, analyze and scientific evaluation of Kaparda bhasma for its Spermatogenesis w.s.r. to Oligospermia. Hence the study entitled.

“PHARMACEUTICO-ANALYTICAL AND EXPERIMENTAL STUDY OF KAPARDA BHASMA IN SPERMATOGENESIS W.S.R. TO OLIGOSPERMIA”

is undertaken

METHADODOLOGY

Test drug

Kaparda bhasma preparation^[8]

Kaparda Bhasma was prepared by following Standard guidelines as prescribed in Ayurvedic Classics. Raw kaparda was procured from Bharat trading company, Mumbai. It was subjected to processing i.e *Shodhana* by dolayantra swedana method using nimbu swarasa as media for 1 yama^[3], followed by incineration by mixing kumari swarasa and triturated, pellets were made later placed in *sharava samputa*, *sandhibandana* was done subjected to *Gaja puta*. After 3 *Gaja puta* kaparda Bhasma was obtained. kaparda bhasma(KB) was analysed as per classical parameters of Bhasma pariksha.

Methodology for Analytical Study

Analytical evaluation of the one samples was carried out by adopting both classical Ayurvedic and modern analytical parameters to ensure standardization and quality assessment. Classical analysis included organoleptic evaluation and classical tests such as Rasa, Varna, Sparsa, Gandha, Rekhāpūṇatva.

Physico-chemical parameters including pH, total ash, acid-insoluble ash, water-soluble ash, and loss on drying were assessed to determine purity, stability, and inorganic content. Modern instrumental analyses such as X-ray diffraction (XRD), scanning electron microscopy with energy dispersive spectroscopy (SEM-EDS), and Fourier transform infrared spectroscopy (FTIR) were performed to evaluate crystallographic nature, elemental composition, and chemical bonding. Particle size and distribution were analyzed using Zeta PALS to assess the impact of pharmaceutical processing on drug fineness and potential bioavailability.

EXPERIMENTAL STUDY

The study was conducted at the SDM Centre for Research in Ayurveda and Allied Sciences, Udupi. Animals were acclimatized and maintained under standard laboratory conditions (24 ± 2 °C, 70% RH, 12 h light/dark cycle) in the animal house, Department of Pharmacology. The experiment was carried out in accordance with CPCSEA guidelines after approval from the Institutional Animal Ethics Committee (IAEC Approval No: SDMCRA/IAEC/TGA-B-03).

24 healthy Wistar albino rats were selected and grouped into 4 groups. Selected animal was grouped by randomisation method. 6 animals in each group. The individual rat was weighed and rats with weight around 250-300 only taken for study and marked with picric acid for identification.

Experimental design

Wistar albino rats were randomly divided into 4 groups.

Group 1-normal control

Group 2-disease control

Group 3-test drug group

Group 4- standard drug group

Experimental induction of oligospermia

Cyclophosphamide was administered intraperitoneally once weekly to induce oligospermia. Dose was calculated according to individual rat weight.

DOSE CALCULATION

The dose for Rats was calculated by referring the table of Paget and Barnes.

i.e Rat Dose Human Dose mg/kgbw X Km ratio/kgbw.

(Rat Dose Human dose(mg/kgbw) X Kmratio/kgbw, it gives per kg body weight dose)

The therapeutic Human dose of kaparda bhasma 250 mg per day.

There for Rat dose = Human Dose X 0.018 X 5 = 22.5mg/kgbw. Hence Rat dose of KB =22.5 mg/kg bw.

Dose of Cyclophosphamide : 45mg/kg bw

Dose of tab. Fertisure M : 44 mg / kg bw

DOSAGE FORM

The Dosage of KB (22.5 mg/kgbw) and Tab. Fertisure M was administered in the form of liquid, by adding powdered drug into distilled water.

DRUG DOSING SCHEDULE

In Cyclophosphamide induced oligospermia model, test drug and standard drug were administered daily for 60 days and Cyclophosphamide were administered once weekly.

RANDOMISATION AND GROUPING

Animals are randomly selected and grouped into 4 groups of six animal each.

STATISTICAL ANALYSIS

The data obtained was analyzed by using analysis of variance (ANOVA) followed by Dunnett's 't' test for

determining the level of significance of the observed effects. A 'P' value of less than 0.05 was considered statistically significant.

➤ CYCLOPHOSPHAMIDE INDUCED SPERMATOGENESIS

Male wistar albino rats weighing about 250-300gm will be divided into four groups of six animals each, cyclophosphamide (45mg/kg bw) will be administered to all the groups except normal control group.

Grouping of animals**Table no. 1: Showing grouping of animals.**

Groups	Group name	Treatment	Dose	Duration
i.	Normal control	Tap water with libitum	-	60days
ii.	Positive control	Cyclophosphamide	45mg/kgBW	60days
iii.	Cyclophosphamide induced oligospermia +Standard drug	Cyclophosphamide	45mg/kgBW	60days
		Tab.FertisureM	44mg /kgBW	60days
iv.	Cyclophosphamide induced oligospermia+ Trial drug	Cyclophosphamide	45mg/kgBW	60days
		Kaparda bhasma	22.5mg/kgBW	60days

Group 1 Normal control: Animals received tap water

Group 2 Positive control group received Cyclophosphamide (45mg/kgbw), and distilled water.

Group 3 Standard drug treated group received Tab. FertisureM (44mg/Kgbw) in addition to Cyclophosphamide.

Group 4 Test drug group received *kaparda bhasma* (22.5mg/kgbw), in addition to Cyclophosphamide.

At the end of the study, all animals were sacrificed by cervical dislocation. The reproductive organs, including the testes, seminal vesicles, and prostate, were carefully

dissected, cleaned extraneous tissues, blotted to remove blood, and weighed. Portions of the testicular tissue were preserved in 10 % formalin for histopathological analysis to assess spermatogenic changes.

RESULTS

Pharmaceutico-Analytical study results were described under 3 headings.

1. Pharmaceutical results
2. Analytical results
3. Experimental results

1. PHARMACEUTICAL RESULTS**Table no. 2: Showing results of *Kaparda Shodhana*.**

Wt of Ashodhiha <i>Kaparda</i>	Wt of shodhiha <i>Kaparda</i>	Loss of <i>Kaparda</i>	Yield %
495gm	473 gm	22 gm	95.55%

Table no. 3: Showing results of *Kaparda marana*.

Wt of <i>Kaparda</i> before marana	Wt of <i>Kaparda</i> after marana	Loss	Yield %
473 gm	373 gm	100 gm	78.8%

2. ANALYTICAL STUDY RESULTS**1. ORGANOLEPTIC CHARACTERS****Table no. 4: Showing Result Of Organoleptic Character Of *Kaparda Bhasma*.**

Organoleptic Character	<i>Kaparda Bhasma</i>
Colour	White
Odour	Characteristic
Taste	Tasteless
Touch	Fine

Table no. 5: Showing Result Of Physical Test Of *Kaparda Bhasma*.

Contents	<i>Kaparda Bhasma</i>
pH value	9.6
Ash value	98.5%
Acid insoluble ash	40.5%
Water soluble ash	1 %
Loss on drying	1.03%

2. Chemical test

1. X-RAY DIFFRACTION STUDY

Table No. 6: Showing XRD Results Of Kaparda Bhasma.

2-Theta	d	Intensity
22.994	3.8646(19)	74
26.167(11)	3.4028(14)	28
26.438(6)	3.3685(8)	30
28.64(4)	3.115(4)	19
29.3301(12)	3.04263(12)	1795
31.387(9)	2.8477(8)	55
33.071(16)	2.7065(13)	21
34.011(17)	2.6338(13)	66
35.946(6)	2.4963(4)	148
37.860(11)	2.3744(7)	17
38.45(2)	2.3395(15)	8
39.371(4)	2.2867(2)	241
43.141(5)	2.0952(2)	201
45.87(3)	1.9767(14)	20
47.100(7)	1.9279(3)	82
47.486(3)	1.91316(9)	459
48.482(3)	1.87614(13)	315

2. SCANNING ELECTRON MICROSCOPY(EDX) STUDY

Table No. 7: Showing SEM EDS Of Kaparda Bhasma.

Sl.no.	Element	Mass %
1	O	42.47
2	Ca	47.24
3	C	8.10

3. PARTICLE SIZE ANALYSIS

Table no 8: Showing The Result Of Mean Particle Size Of Kaparda Bhasma.

Name of the sample	Mean particle size
Kaparda Bhasma	1181.3nm

4. FTIR ANALYSIS

Table No. 9: Showing FTIR Observations Of Kaparda Bhasma.

Sample peaks Cm^{-1}	Bond	Functional group
7233	O-H	Carboxylic acid
6800	N-H	Amine
3643	O-H	Carboxylic acid
1412	C=O	Acid halide
873	C=O	Unsaturated ester
855	C=O	Ester

5. NPST ANALYSIS

Table No. 10: Showing Result Of NPST Of Kaparda Bhasma.

Sample	1st phase (0-5mins)	2nd phase (5-20mins)	3rd phase (20mins-1day)
kaaprda bhasma	Red circle	Fading of colour started With whitening of inner side of circle	Faded to more than half of intensity after 24 hours

3. EXPERIMENTAL RESULTS

Each rat was observed thoroughly. The observations were recorded.

The results were analyzed for statistical significance on the base of ANOVA one way.

EFFECT OF KAPARDA BHASMA ON SPERM MOTILITY

(a) Table no. 11: Showing Effect Of Kaparda Bhasma On Sluggish Motile Sperm.

Group	sluggish motile sperm	% change
NCG	15.17 ± 3.554	-
PCG	23.83±2.574	57.08↑@
SDG	29.50 ± 6.850	23.79↑#
TDG	27.60± 3.750	15.82↑#

(b) Table no. 12: Showing Effect Of Kaparda Bhasma On Non Motile Sperm.

Group	non motile sperm	% change
NCG	68.17± 13.76	-
PCG	76.17±2.574	11.73↑@
SDG	70.50± 6.850	7.44↓#
TDG	72.40± 3.750	4.94↓#

(c) Table no. 13: Showing Effect Of Kaparda Bhasma On Sperm Count.

Group	sperm count	% change
NCG	1.947± 0.4652	-
PCG	3.417±0.5039	75.5↑@
SDG	5.170± 0.6306	51.30↑#
TDG	7.321± 0.7917**	114.25↑#

MORPHOLOGY

(a) Table no. 14: Showing Effect Of Kaparda Bhasma On Normal Sperm.

Group	normal sperm	% change
NCG	68.83± 13.78	-
PCG	83.17±0.6009	20.83↑@
SDG	81.25± 0.4787	2.30↓#
TDG	81.40± 0.6782	2.128↓#

(b)Table no. 15: Showing Effect Of Kaparda Bhasma On Hookless Sperm.

Group	hookless sperm	% change
NCG	12.83± 2.638	-
PCG	15.50±0.6708	20.81↑@
SDG	18.75±0.4787	20.96↑#
TDG	18.40± 0.5099	18.70↑#

(c)Table no. 16: Showing Effect Of *Kaparda Bhasma* On Amorphous Head.

Group	amorphous head	% change
NCG	1.167± 0.3073	-
PCG	1.333±0.2108	14.22↑@
SDG	0.000± 0.000**	100↓#
TDG	0.000± 0.000**	100↓#

(b)Table no. 19: Showing Effect Of *Kaparda Bhasma* On Wt. Of Prostate.

Group	wt. of prostate	% change
NCG	0.9500± 0.005774	-
PCG	0.8300±0.01732	12.63↓@
SDG	0.7400± 0.000	10.84↓#
TDG	0.6250± 0.1126	24.69↓#

(d)Table no. 17: Showing Effect Of *Kaparda Bhasma* On Curled Tail.

Group	curled tail	% change
NCG	0.1667± 0.1667	-
PCG	0.000±0.000	100↓@
SDG	0.000± 0.000	0↑#
TDG	0.2000± 0.2000	20↑#

(c)Table no. 20: Showing Effect Of *Kaparda Bhasma* On Wt. Of Seminal Vesicle.

Group	wt.of seminal vesicle	% change
NCG	0.4050± 0.04907	-
PCG	0.2300±0.01732*	43.20↓@
SDG	0.8000± 0.01155**	247↑#
TDG	0.3800± 0.06351	65.21↑#

Effect on reproductive organs(a)Table no. 18: Showing Effect Of *Kaparda Bhasma* On Wt. Of Testis.

Group	wt.of testis	% change
NCG	2.120± 0.02309	-
PCG	1.605±0.2165*	24.292↓@
SDG	2.895± 0.03753**	80.37↑#
TDG	3.085± 0.02021**	92.21↑#

Table no. 21: Showing The Consolidated Statement Of The Results.

parameters	PC	SDG	TDG
Sluggish motile	NS↑	NS↑	NS↑
Non motile	NS↑	NS↓	NS↓
Sperm count	NS↑	NS↑	NS↑
Normal sperm	NS↑	NS↓	NS↓
Hookless	NS↑	NS↑	NS↑
Amorphous head	NS↑	VS↓	VS↓
Curled tail	NS↓	NS↑	NS↑
Testis weight	S↓	VS↑	VS↑
Prostate weight	NS↓	NS↓	NS↓
Seminal vesicle weight	S↓	VS↑	NS↑

HISTOPATHOLOGICAL STUDIES

Table no. 22: Showing Histopathology Of Testis.

Groups	Depletion and degeneration of elongated spermatids	Exfoliation of germ cells	Spermatid retention	Atrophic tubules
NC1	-	-	-	-
NC2	-	-	-	-
PC1	-	++	-	-
PC2	++	+	-	-
S1	-	-	-	-
S2	-	-	-	-
T1	-	-	-	-
T2	-	-	-	-

Nil, + mild, ++ moderate, +++ severe

Table no. 23: Showing Histopathology Of Prostate.

Group	Decrease of Secretion	Inflammatory infiltrate	Apoptosis
NC1	-	+	-
NC2	-	+	-
PC1	-	+	-
PC2	-	+	-
S1	-	-	-
S2	-	-	-
T1	+	-	-
T2	++	-	-

Table no. 24: Showing Histopathology Of Seminal Vesicle.

Groups	Decrease of Secretion Compared with NC	Sloughing of cells	Apoptosis
NC1	-	-	-
NC2	-	-	-
PC1	+++	++	++
PC2	-	++	+
S1	-	-	-
S2	-	-	-
T1	-	-	-
T2	-	-	-

FIGURES

Rajiv Gandhi Education Society's
Ayurvedic Medical College, Hospital, PG Research Centre
 Dist: Gadag State: Karnataka
RON – 582 209,
 (AYUSH approved ASU drug testing laboratory Lic. No. TL-13/2023)
DRUG TESTING LAB

Outward number: RGE/SDTL/LR/25/0211
 Ref No: DTLRG/25/178 Registration date: 10/11/2025 Report date: 25/11/25
 Submitted by: Dr. Geeta Requestion no: Sample Qty: 25GM
 Sample name: Kaparda yoga Batch no: NA Date of expiry: NA
 Reference: Compound drug Mfg Date: NA Part/Form: Powder

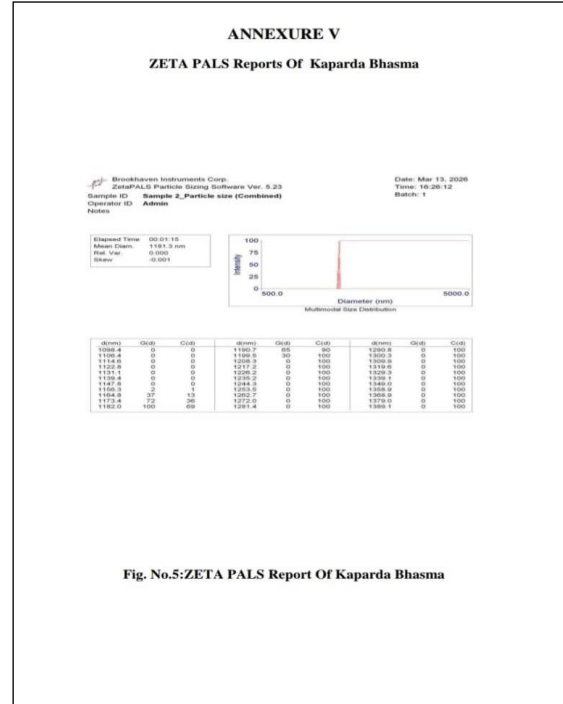
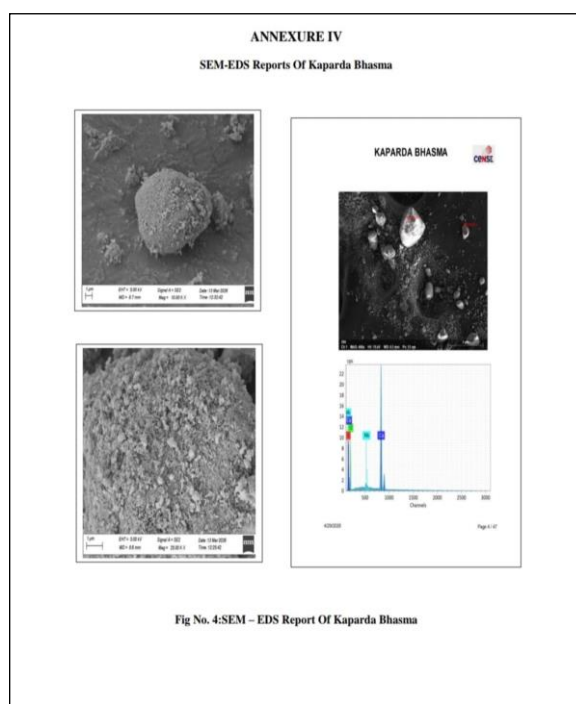
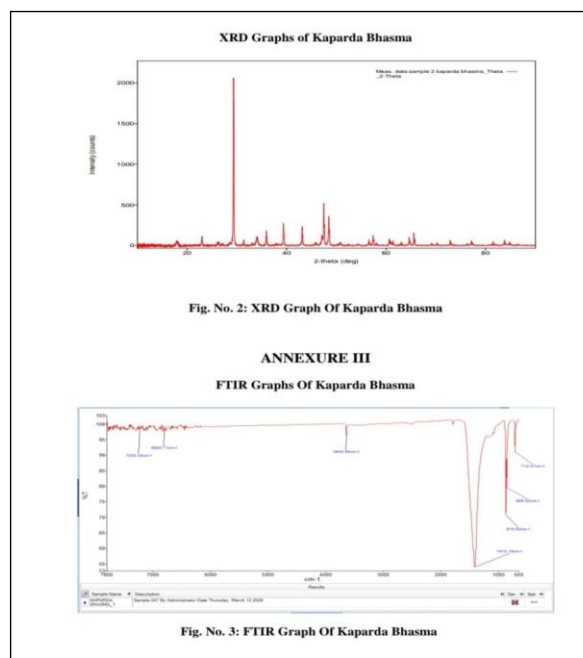
TEST REPORT
 Form – 50 [see rule: 160-D (f)]
 (As per Drug and cosmetic act 1940 and the rules there under)

Sl. No	TEST	RESULTS
A. Organoleptic study		
1.	Description	Form: Fine amorphous powder Colour: White Taste: Tasteless Odour: Characteristic
B. Physico-Chemical study		
2.	Ash value	98.5%
3.	Acid insoluble ash	40.5%
4.	Water soluble ash	1%
5.	pH	9.6
6.	Loss on drying	1.03%

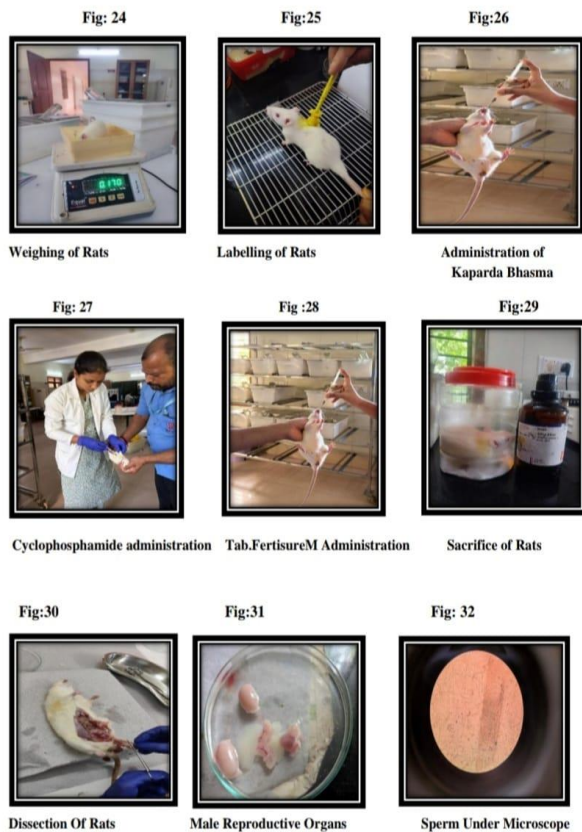
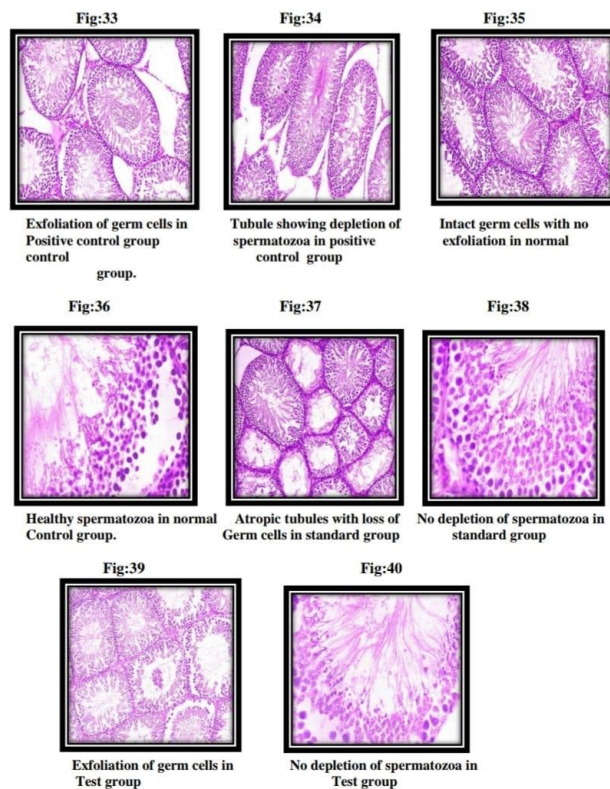
End of Report
 (Standards referred above are as per API and PSAT)
 In my opinion the sample is standard quality.

Analyst
ANALYST

Lab in-charge (Authorized signature)
IN-CHARGE
 DTL R.G.E.S. A.M.C. & H.RON

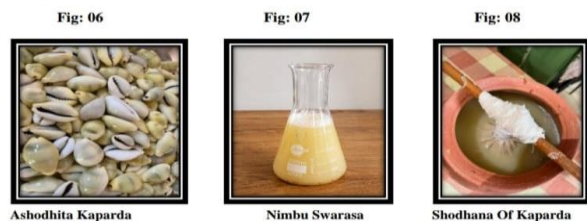


EXPERIMENTAL STUDY

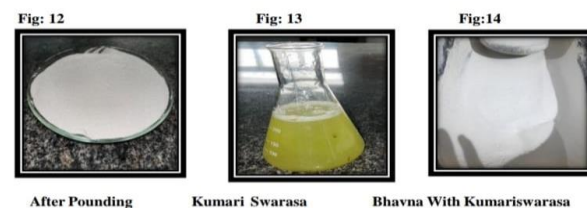
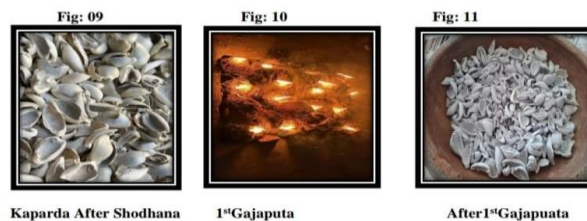
HISTOPATHOLOGY
(A) Testes

PHARMACEUTICAL STUDY

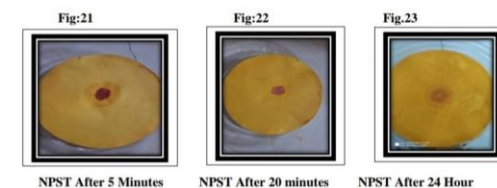
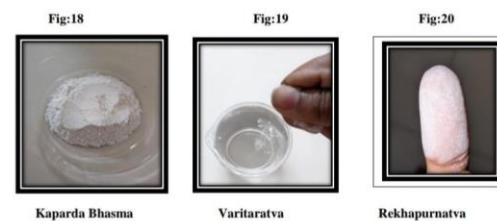
KAPARDA SHODHANA

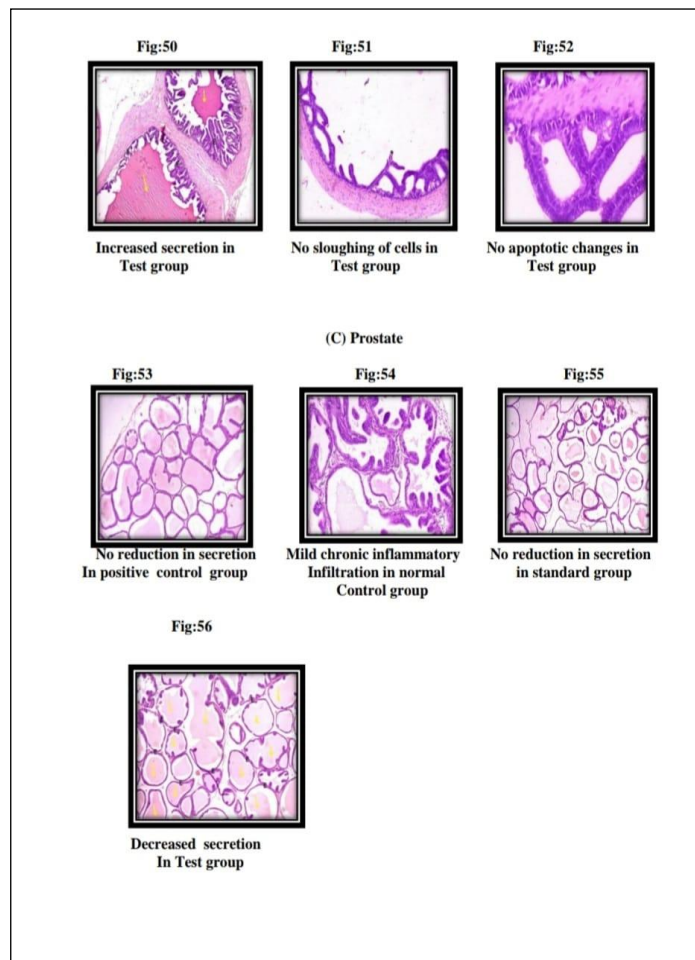
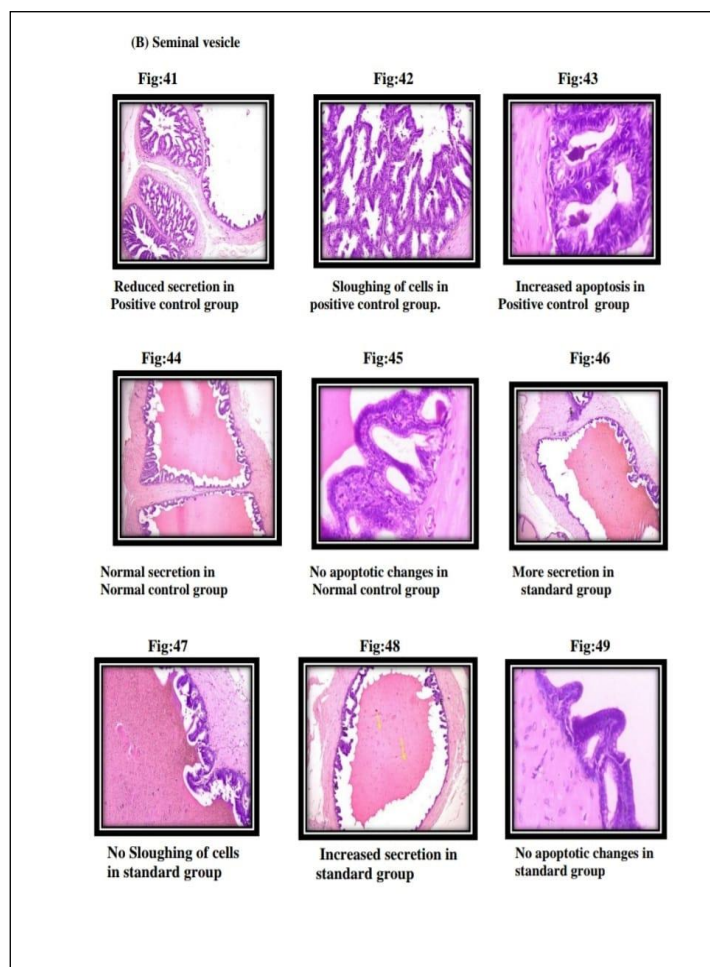


KAPARDA MARANA



BHASMA PAREEKSHA





DISCUSSION

Pharmaceutical Study

Kaparda was subjected to classical Shodhana using Nimbu Swarasa in Dolayantra Swedana, resulting in a reduction of initial weight from 495 g to 473 g, indicating removal of adherent and soluble impurities during processing. The acidic medium of Nimbu Swarasa facilitates surface purification and may promote physicochemical modification of the raw mineral through prolonged contact and heating. The change in pH of the processing medium from 2 to 4 after Shodhana suggests interaction between the mineral matrix and acidic constituents of the medium, with partial neutralization during processing. Subsequent Marana facilitates repeated trituration and thermal transformation, resulting in conversion of the processed Kaparda into a fine and therapeutically suitable Bhasma. The pharmaceutical process therefore involves sequential physical and chemical transformations, including purification, particle-size reduction and modification of the mineral matrix.

Analytical Study

The analytical characterization demonstrated that Kaparda Bhasma possesses a predominantly mineral composition, with calcium and oxygen constituting the major elemental components. The acid-insoluble ash value of 40.5% and water-soluble ash value of 1%

further provide physicochemical indices for characterization and quality assessment of the prepared Bhasma. SEM-EDX analysis showed calcium (47.24%) and oxygen (42.47%) as predominant elements, with carbon (8.10%) representing a comparatively smaller fraction, supporting the calcium-rich nature of the prepared Bhasma. XRD analysis revealed multiple sharp diffraction peaks, indicating the presence of crystalline phases; the prominent diffraction peak observed at $20-29.3301^\circ$ with an intensity of 1795 counts represents a characteristic crystalline reflection of the sample. FTIR analysis demonstrated absorption bands corresponding to O-H, N-H and C-O functional groups, indicating the presence of surface chemical functionalities associated with the processed material. Particle-size analysis showed a measured particle size of 1376.8 nm, demonstrating substantial particle-size reduction following the pharmaceutical processing of Kaparda. Collectively, the classical Bhasma Pariksha and instrumental findings establish a reproducible physicochemical profile of Kaparda Bhasma and provide an analytical basis for correlating its pharmaceutical processing with its observed biological activity.

Experimental Study

Cyclophosphamide-treated PCG showed increased sluggish and non-motile sperm, indicating oxidative stress and impaired spermatogenesis. Both SDG and

TDG improved sperm quality by increasing sperm count and reducing non-motile sperm. TDG showed better improvement, with higher sperm count (7.321 vs. 5.170 million/ml) and lower non-motile sperm (70.50% vs. 72.40%), suggesting stronger spermatoprotective activity.

PCG showed increased hookless (15.50%) and amorphous head (1.33%) sperm, indicating oxidative damage. Both SDG and TDG completely prevented amorphous head defects. Hookless sperm and normal morphology were comparable between SDG and TDG, indicating that Kaparda Bhasma has spermatoprotective and antioxidant effects similar to the standard drug.

Cyclophosphamide reduced testicular and seminal vesicle weights, indicating reproductive tissue damage and impaired androgenic function. TDG showed greater improvement in testicular weight (92.21%) than SDG (80.37%), while SDG showed better recovery of seminal vesicle weight. Prostate changes were non-significant, suggesting minimal prostatic effects.

PCG showed degeneration and depletion of germ cells, spermatid exfoliation, reduced seminal secretion, cellular sloughing, and apoptosis. Both SDG and TDG restored normal tissue architecture without significant degeneration or germ-cell loss. TDG showed increased prostatic secretion, suggesting slightly better functional recovery and overall reproductive tissue protection.

CONCLUSION

The present study demonstrated the successful pharmaceutical preparation of Kaparda Bhasma through classical Shodhana and Marana procedures, with attainment of Bhasma Siddhi Lakshanas. Physicochemical and instrumental analyses confirmed its quality, purity, crystalline nature, calcium-rich composition, and fine particle size. The experimental study in male Wistar albino rats showed significant improvement in sperm count, motility, and morphology following administration of Kaparda Bhasma. Histopathological findings further demonstrated active spermatogenesis with preserved seminiferous tubules and normal reproductive tissue architecture. Overall, the pharmaceutical and analytical findings established the quality and authenticity of the prepared Kaparda Bhasma. The experimental findings provide scientific support for its classical Vrushya activity and traditional use in promoting male reproductive health. Thus, Kaparda Bhasma may have potential therapeutic utility in the management of Shukra Kshaya (Oligospermia).

REFERENCE

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