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## Research Article

### AVRISHYA DRAVYA: PERIL TO FERTILITY; AN EXPERIMENTAL DATA OF SHIGRU BEEJA (*MORINGA OLIEFERA* Lam.) ON SPERMATOGENESIS MODULATION ACTIVITY

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#### ABSTRACT

*Ayurveda* emphasizes on *Bahupraja purusha*, and it compares the one without offspring as fruitless tree. Getting healthy progeny is the main aim of human being; hence *Vajikarana* and *Rasayana* have been given at most importance throughout the literatures of *Ayurveda*. Simultaneously we can trace the references about *Avrishya Dravyas* which interfere in the production of *shukra* and its *pravartana*. *Shigru* (*Moringa oliefera* Lam.) seeds are mentioned as *Avrishya* in *Ashtanga samgraha* and *Nighants*. Fruits form the common eatable vegetables of maximum population. Hence with this background an experimental study was designed to evaluate the effect of *Shigru beeja* on spermatogenesis modulation activity on male albino rats. The activity was evaluated on parameters like sperm count, body weight of rats, motility of sperms, hematological parameters like Hb, RBC, MCV, WBC count, Platelet, histopathological studies like weight of testis, seminiferous tubules etc before and after test design. A careful analysis of data and changes observed after the administration of the test drug SBJ points to the fact that definitely it had modulated spermatogenesis activity in experimental animals.

**KEY WORDS:** *Avrishya*, *Shigru beeja*, *Moringa oliefera*, Spermatogenesis, *Vajikarana*

#### INTRODUCTION

Infertility was attached with a lot of social embarrassment since the period of Brihatrayees<sup>1</sup>. *Vajikarana Tantra* is one among *Ashtanga Ayurveda*, which deals with promotion of sexual health and prevention and cure of sex related disorders. The drugs used for this treatment are known as *Vrishya dravyas*<sup>2</sup>. We can find plenty of drugs mentioned under *Vrishya Karma*. Simultaneously we get reference of certain *Avrishya dravyas* in the texts of *Ayurveda*<sup>3</sup>. Though the word meaning of *Avrishya karma* and its detail explanation is not available, but *Acharyas* have made reference about certain drugs, diet and life style which may result in *Avrishyata*. *Bhavaprakasha* says *Vrishya* as *Shukra vriddhikara* i.e. which increase the quantity and quality of *Shukra* in Human body<sup>2</sup>. As direct reference about *Avrishya Karma* is not available, grammatical meaning of this word denotes which decrease in the quantity and quality of *Shukra* in Human body, thereby results in fertility related disorders<sup>4</sup>.

*Shigru* (*Moringa oliefera* Lam.) is one such popular drug which is extensively used in our system since long time as food as well as medicine and its different parts are mentioned for the treatment of various diseases<sup>5</sup>. *Shigru beeja* is considered as *Avrishya* in *Bhavaprakasha*, *Astanga hridaya* and *Kaiydeva nighantu*<sup>6,7</sup>. Hence an experimental study was planned to conduct spermatogenesis modulation activity<sup>8</sup> of SBJ (*Shigru beeja Moringa oliefera* Lam.)

#### MATERIALS & METHODS

##### Test drug

Matured fruits of *Shigru* (*Moringa oliefera* Lam.) were collected from Udupi district during their fruiting season, authenticated through botanist. Voucher specimen deposited in SDM Centre of Research in Ayurveda and Allied Sciences, Udupi, Karnataka. (Voucher no. 227.13031101). Seeds were separated from fruits, shade dried and fine powder was prepared and used for experimental study.

##### Experimental Animals

The study was carried out in Wistar strain male albino rats maintained under prevailing husbandry conditions in the animal house attached to the pharmacology laboratory of S.D.M. centre for research in Ayurveda and allied sciences. Rats were fed with rat pellet and tap water. They were exposed to natural day and night cycles and maintained at standard laboratory conditions. (Ethical clearance number SDMCAU, IAEC, 2011-12DG 02)

##### Dose

The dose of the test drug was calculated using the following formula:

$$\begin{aligned} \text{Rat dose} &= \text{Human dose} \times \text{Body surface area constant of the rats,} \\ &\text{i.e. } 0.018 \times 5/\text{kg} \\ &= 3000\text{mg} \times 0.018 \times 5/\text{kg body wt.} \\ &= 270 \text{ mg / kg} \end{aligned}$$

### Animal grouping

The selected animals were grouped randomly into three groups, each consisting eight rats. The first group was considered as control and administered with 0.5% gum acacia. The second group was considered as reference standard and administered cyclophosphamide once daily 20mg/kg of body weight for 65 days. The third group was administered with SBJ as a suspension in 0.5% gum acacia once daily in the calculated dose i.e. 27 mg/kg of body weight for 65 days with the help of oral catheter.

After 65 days of drug administration, animals of three groups were weighed and anesthetized and blood was collected from retro orbital plexus. An incision was made in the inguinal region and cauda epididymal tissue was identified. Cauda epididymal tissue was excised out carefully and transferred to normal saline (0.5 ml) and teased gently with forceps to liberate the spermatozoa. Cauda epididymis suspension was incubated at 38<sup>o</sup> C for 5 minutes before testing and was examined for sperm count, motility and sperm morphology assessment. Blood samples were collected for hematological and biochemical analysis.

Then the rats were sacrificed by overdose of anesthesia and important organs like testis, seminal vesicles etc were dissected out. These were weighed transferred to fixing solution (10% formalin) for histopathological examinations. Haematological studies were done on following parameters like Hb%, RBC, PCV, MCV, MCHC, WBC, platelet<sup>10</sup>.

### Statistical Analysis

The data generated during the study have been presented as Mean  $\pm$  SEM. Difference between groups were determined by one way ANOVA followed by Dunnett multiple t-test as post hoc test.

## RESULTS

### Effect of SBJ on body weight

The body weight gain was observed in all the three groups when the initial body weight was compared to the body weight recorded at 65<sup>th</sup> day. However, the body weight gain was found to be less in test drug administered group in comparison to reference standard and control groups. In reference standard also the body weight gain was less in comparison to the normal control rats. (Table 1)

### Effect of SBJ churna on sperm count

In the standard and test drug administered groups an apparent decrease in sperm count was observed in comparison to control group. However though the decrease was around 40 to 45 % it was found to be statistically non-significant. (Table 2)

### Effect of SBJ on the effect on sperm motility

An apparent decrease in RLP and SLP was observed in test & standard drug administered groups in comparison with control group. However, the observed decrease was found to be statistically non-significant. An apparent increase in IMM was observed in both test and standard groups in comparison to the control group. However, this increase was also found to be statistically non-significant. (Table 4)

### Effect of SBJ on the weight of testis

Mild decrease in the weight of the testis in test & standard drug administered group was observed when compared with control

group. However, the observed change was found to be statistically non-significant. (Table 3)

### Effect of SBJ on the weight of seminal vesicle

An apparent mild increase in reference standard and an apparent decrease in test group on weight of seminal vesicle was observed in comparison to control group. However, the observed changes were found to be statically non-significant. (Table 3)

### Effect of SBJ on the weight of prostate

An apparent marginal increase observed in prostate weight in reference standard group and moderate decrease observed in test drug administered group was found to be statistically non-significant in comparison to the control group. (Table 3)

### Hematological changes

Haematological parameters like RBC, Hb, PCV, MC, MCH, MCHC and WBC remained unaffected. Platelet count has shown moderate increase in both test and standard drug administered group in comparison to control group. This may indicate stimulation in the formation of platelets and may not be indicative of any adverse effect. Other parameters have not shown significant values related to study. This indicates that the standard and test drug does not have serious toxicological implications. (Table 5)

### Histopathological study on vital organ

Examination of testis from control group exhibit normal cytoarchitecture with features of good spermatogenesis; where as in standard group seminiferous tubules with few and decreased number of sperm. But there was no disorganization of normal cytoarchitecture. Test drug administered group showed features of reduced spermatogenesis, more number of seminiferous tubules with few or comparatively less sperms in ST lumen and increased proportion of Leydig's cells. (Table 6) (Figure 1. a,b,c)

Seminal vesicle from control group show normal cytoarchitecture, whereas from standard group exhibited moderately proliferated epithelial layer. Highly branched epithelial layer with comparatively thicker capsules were the features observed in test drug administered group. (Table 6) (Figure 2. a,b,c)

## DISCUSSION

*Shukra* one among *Sapta Dhatu*, is *sowmya* in nature and *Garbhotpadana* is its *Mukhya Karma*<sup>10</sup>. It is nourished by *Ahara rasa*, according to *dhatu poshana siddhanta*. Brihatrayi texts opines that *Katu*, *Tiktha*, *Kashaya*, *Lavana rasa yuktha ahara sevana* leads to *Avrishya karma*<sup>4</sup>. They use *Pumstvahani*, *Shukrahara* as synonymous to *Avrishya*. Experimental study conducted to evaluate the effect of SBJ (*Moringa oliefera* Lamk.) in spermatogenesis modulation activity on rats has provided certain significant data.

Body weight gain was found to be less in test drug administered group in comparison to reference standard and control groups. SBJ being possessing qualities like *rukshna*, *ushna* may result in decrease in body weight. Intake of *Guru*, *Madhura*, *Brihmana dravyas* results in *dhatu upachaya*. Hence trial drug having opposite qualities resulted in decrease in body weight. In the experimental models SBJ has decreased the sperm count as well as the quality of sperms, which shows it negative effects towards *Shukra dhatu*.

**Table 1 Effect of SBJ on body weight**

Groups	Initial Body Weight	Body weight on 65 <sup>th</sup> day	Weight gain in %
Control	285.714±7.190	335±9.449	17.54
Standard	275.714±8.621	308.571±11.686	12↓
Test	250.625±12.728	274.375±16.324	9.6↓

Data in Mean ± SEM

**Table 2 Effect of SBJ on sperm count**

Group	Sperm count/cumm MEAN±SEM	% Change
Control	73150±44360	
Standard	43878.57±1000	40.0156↓
Test	40012.5±5980.3	45.3007↓

Data in Mean ± SEM

**Table 3 Effect of SBJ on the weight of Testis, seminal vesicles and prostrate**

Group	Testis weight(g) mean	Seminal vesicle	Prostate weight(g)
Control	2.99 ± 0.18	1.02 ± 0.05	0.88 ± 0.09
Standard	2.95 ± 0.14	1.12 ± 0.11	0.92 ± 0.06
Test	2.83 ± 0.11	0.81 ± 0.08	0.77 ± 0.05

Data in Mean ± SEM

**Table 4 Effect of SBJ on sperm motility**

Group	RLP%	%	SLP%	%	IMM%	%
	Mean ±SEM	Change	Mean± SEM	Change	Mean ±SEM	Change
Control	0.28± 0.18		27.42±6.740		72.28±6.711	
Standard	0.25±0.25	12.49↓	21.14±3.181	22.91↓	78.57±3.206	8.69↑
Test	0.25±0.25	12.49↓	19.87±3.297	27.53↓	79.87±3.308	10.49↑

Data in Mean ± SEM, RLP=Rapid Linear Progressive, SLP = Slow linear progressive, IMM= Immotile

**Table 5 Effect of SBJ on Heamatological parameters**

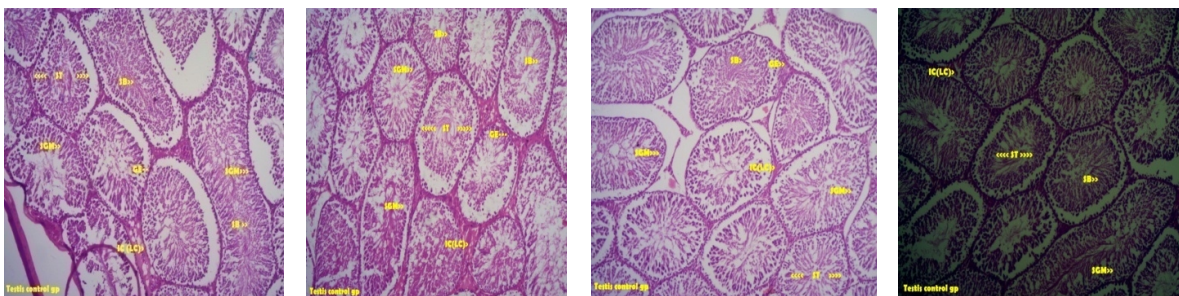
Group	WBC (10 <sup>3</sup> //µl)	RBC (10 <sup>6</sup> /µl)	Hb g/dl	MCH (pg)	PCV (%)	MCHC (g/dl)	Platelet (10 <sup>3</sup> /µl)
Control	10385.712±12	8.304±0.207	15.9±0.392	19.1±0.241	45.985±1.344	34.557±0.19	6.668±0.321
Standard	4233.33±61**	7.708±0.242	15.1±0.342	20.116±0.357*	44.9±1.053	34.51±0.234	7.168±0.418
Test	8328.571±36	8.264±0.104	15.81±0.311	19.071±0.226	46.528±1.085	33.928±0.305	7.091±0.427

Data expressed in Mean ± SEM, \* P<0.05, \*\*p<0.01 in comparison to normal control group.

**Table 6 Histopathological changes**

Sl.no	Organs	Control	Standard	Test
1	Testis	NC	Few seminiferous tubules with decreased number of sperms were observed	Reduced spermatogenesis, more number of seminiferous tubules with few or comparatively less sperms.
2	Seminal vesical	NC	Moderately proliferated epithelial layer.	Highly branched epithelial layer with comparatively thicker capsule.

NC- No changes



**Figure 1.a Photomicrographs of section Testis (Control group)**

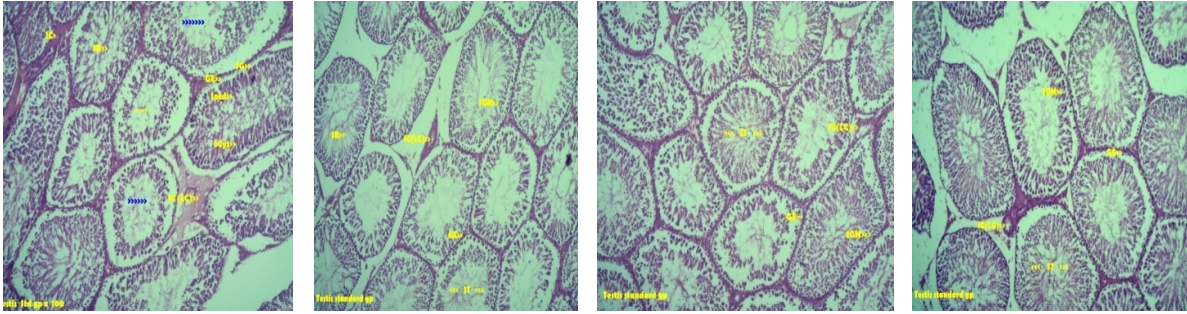


Figure 1.b Photomicrographs of section Testis (Standard group)

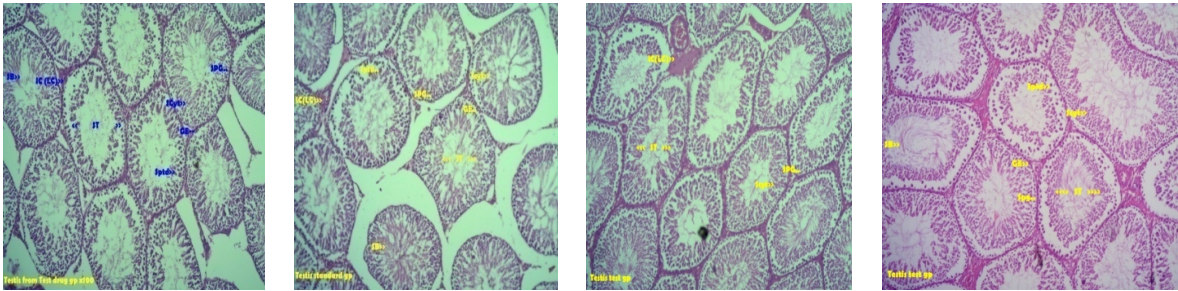


Figure 1.c Photomicrographs of section Testis (Test group)



Figure 2.a Photomicrographs of section Seminal vesicals (Control group)

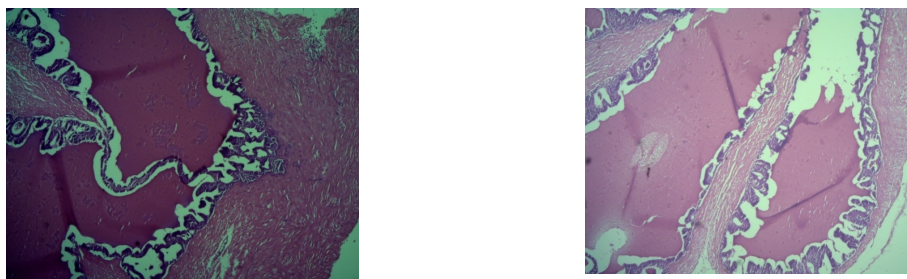


Figure 2.b Photomicrographs of section Seminal vesicals (Standard group)



Figure 2.c Photomicrographs of section Seminal vesicals (Test group)

Mild decrease in the weight of the testis in test & standard drug administered group was observed when compared with control group. *Vrishana*(Testis) is said to be *Shukravaha srotomoola*. Decrease in the weight of same may hamper production of sperm. Mode of action of *Avrishya Dravyas* can be well understood with this result. In histopathological studies reduced spermatogenesis in the testis was observed in comparison to the normal control. A careful analysis of the various data such as sperm count, quality of sperm, weight of testis, weight of seminal vesicle and histopathological changes observed after the administration of the test drug points to the fact that it has mild to moderate spermatogenesis modulation activity when consumed over a long period in continuous manner. Heamatological reports have not shown much change. This indicates that standard and test drug does not have serious toxicological effect.

## CONCLUSION

Getting healthy progeny is the main aim of human being; hence *Vajikarana* and *Rasayana* have been given at most importance throughout the literatures of *Ayurveda*. *Avrishya* drugs are the one which produce harm to *Shukra*, thereby affecting male fertility. *Shigru* (*Moringa oliefera* Lamk) seeds are mentioned as *Avrishya* in *Ashtanga samgraha* and *Nighantu* texts. Data obtained in this experimental study proved the test drug definitely has spermatogenesis modulation activity. *Ruksha, Tikshna guna* and *Katu rasa* of *Shigru* which are opposite to the qualities of *Shukra dhatu* might have been resulted in its normal physiological production, leading to pathological condition *Avrishyata*.

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