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## A studies on Aphrodisiac effect of *Madhuca longifolia* (Flowers) extract in male Poultry

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**Abstract**

Regular use of pesticides in our country decrease the fertility day by day that decrease the aphrodisiac effect in poultry. The present study was conducted to investigate the aphrodisiac effect of ethanolic extract of *Madhuca Longifolia* (Flowers) in male poultry birds. The bird treated with *Madhuca* (flowers) extract showed positively aphrodisiac effect by increase in value of the semen parameter (sperm conc., live & volume). Sperm concentration was increase in both group 1 and group 2, but in group 2 sperm concentration increase more significantly in comparison to group 1. Live spermatozoa was also increase in group 1 and group 2, in group 2 live sperm percentage was significantly higher than group 1. Semen volume was also increase and found greater in group 2 in comparison to group 1 to control group. On the basis of these findings we can say that *Madhuca* has positive aphrodisiac effect.

**Keywords:** poultry, *Madhuca*, spermatozoa, aphrodisiac

**Introduction**

Aphrodisiac is the word derived from Aphrodite, the Greek goddess of sexuality, love & beauty. An aphrodisiac is defined as an agent (food or drug) that arouses sexual desire [1]. But present time regular use of pesticides is increasing day by day particularly in third world countries that decrease the fertility continues [2]. India uses approximately 85,000 tonnes of pesticides annually and an increase of 8% is expected every year. The residue of such environmental pollutant remain in soil, water, air, feed & fodder items for a longer period, to contaminate them [3]. Chicken are especially vulnerable to pesticides toxicity because poultry houses are dusted with pesticides that decrease the all the parameter of semen those related the aphrodisiac potential. The logic that there is general resemblance between man and animal with regard to toxicity profile, suggests that animal population can best be utilized as bio-indicator of pollution.

Exposure of poultry to chemical pesticides causes health consequences to poultry contributing in great economic loss, while also posing a potential threat to public health due to presence of pesticides in poultry meat, ample evidences exist to suggest that the use of pesticides on crop, in store houses, in poultry houses, the no judicious application for spraying or in dipping solution to prevent ectoparasites, leaves behind in residue causing serious health effect [4, 5]. Chronic exposure of chicks to small amount of OPP leads to deleterious effect on metabolism, immune system and reproductive system of birds [6]. In fact, dairy cattle rearing on drinking water contaminated with sewage reduced their reproductive performance [7].

The exposure of males to pesticides can adversely affect pregnancy outcome through a direct genetic or epigenetic effect of their residues on the male germ cells either during spermatogenesis in the testis or sperm maturation in epididymis or by the direct exposure of oocyte during fertilization to the pesticide residues in the seminal plasma [8, 9]. There is growing evidence regarding the adverse impact of certain pesticide residues on reproductive system and such pesticide residue are known as "reproductive toxicants" or "endocrine disrupters". These toxicants modulate and or disrupt reproductive hormone milieu by acting at a variety of sites including hypothalamus, pituitary and reproductive organs [10]. During the course of foetal or early neonatal life, any disruption in the differentiation/ multiplication of sertoli cells in fetal testis by the environmental estrogens in detrimental for the adult to produce sperm is determined by the sertoli cells [11, 12, 13].

**Materials and Methods**

The flowers of *Madhuca longifolia* were collected from the campus of N.D. University during the month of May & June. The plant material were identified and authenticated with the help of scientist of college of Horticulture. After proper identification flowers was shed dried

powdered and passed through 40 meshed and stored in closed vessel for further use. *Madhuca longifolia* flowers were used to prepare ethanolic extract. For this purpose absolute alcohol 95% ethanol was used to prepare the extract. Percent yield (w/w) of *Madhuca longifolia* flowers ethanolic extract was calculated as 42.0%. Percent yield of *Madhuca longifolia* flowers 45 % (w/w) with 95% ethanol [14].

### Experimental design

The experimental design for this study is shown in the table No 1. Twenty four male birds about the age 10-12 month were randomly divided into three groups i.e. A, B, C. Each test group comprised of 8 birds along with control as mentioned in table. Doses were given in drinking water approximately 1/10<sup>th</sup> and 1/5<sup>th</sup> of LD<sub>50</sub> of alcoholic extract of *M. Longifolia* (Flowers).

### Semen collection

The cocks kept on ambient temperature i.e. 30 °C and relative humidity i.e. 65% during the study period. One month prior to commencement of semen collection, all cocks were kept in individual cages (32×34×53 cm). All cocks were fed with commercial poultry pellets consisting of 18% crude protein and water was provided ad libitum. Semen samples were collected once a week (Monday). Semen collection was done once a week because the time range required for semen to pass from testes to distal region of duct's deferens varies from 1-4 days [15]. First, the cloacae area was cleaned. The back and tail feathers and the abdominal region were stroked gently and repeatedly, which resulted in erection of phallus. Semen was ejaculated after slight pressure was applied to the inverted cloacae. The semen was carefully collected in a test tube and placed in a water bath maintained at 37 °C prior to evaluation.

### Semen evaluation

Sodium citrate and egg yolk were prepared and used as an extender in this study. The volume of the ejaculated semen from each cock was measured using a graduated test tube. The pH was determined using a pH meter. Spermatozoa concentration was determined using the haemocytometer method, and the technique involved in mixing semen with appropriate diluents at a dilution ratio of 1:200 with an eosin solution [16]. The evaluation of sperm motility from the diluted semen was conducted at 400X magnification on a warm stage. A drop of diluted semen was placed on a preheated slide and a cover slip was used to cover the slide; the cover slip helped to prevent overflow, allowed a uniform film to form, and prevented quick drying of semen [17]. The remnant of the semen in each replicate was measured to evaluate the percentage of live and dead spermatozoa as determined from 10µL semen, mixed with 50µL of eosin nigrosin stain to make a thin smear. The smear was air dried for 10 min. At least 200 spermatozoa were examined (400X) under emulsion oil and those with differential morphology were counted. The colour and consistency of the semen were evaluated visually, including varieties that were creamy, grainy, bloody, watery, or contaminate. The following parameters were recorded for the evaluation of aphrodisiac effect of alcoholic extract of *Madhuca longifolia* (Flowers) in male poultry birds.

### Statistical study

In this study statistically, analysis of variance was applied. Completely randomized design was used and significant differences were analysed at 5% level of significance. Comparative study among group 1 and group 2 to control group was done by Das [18].

**Table 1:** Experimental design for evaluation of effect of extract of *Madhuca* (flower) in male poultry birds.

Group	Extract/drug	No. of animal	Dose of Extract (mg/kg b. wt.)	Duration
A	Control (G0)	8	-	
B	1/10 <sup>th</sup> of LD <sub>50</sub> of <i>M. longifolia</i> (flowers)(G1)	8	100 Daily single Dose	28 days
C	1/5 <sup>th</sup> of LD <sub>50</sub> of <i>M. longifolia</i> (flowers)(G2)	8	200 Daily single Dose	28 days

### Result & Discussion

In this study *Madhuca longifolia* flowers extract was given

@ 100 mg/kg b. wt (1/10<sup>th</sup> of LD<sub>50</sub>) and 200 mg/kg b. wt (1/5<sup>th</sup> of LD<sub>50</sub>) daily single dose (OECD, 2008).

**Table 2:** Effect of ethanolic extract of *Madhuca longifolia* (Flowers) at 7 days on below various semen parameter on male poultry birds - (n=8) (mean ± S.E.,)

Parameter	Control	Treated with oral dose/10 LD <sub>50</sub> of <i>Madhuca</i>	Treated with oral dose/5 LD <sub>50</sub> of <i>Madhuca</i>
	G0	G1	G2
Concentration of spermatozoa (10 <sup>9</sup> Sperm/ml)	2.62±.03 <sup>c</sup>	2.88±.03 <sup>d</sup>	3.01±.03 <sup>d</sup>
Live spermatozoa %	97.61±.35 <sup>c</sup>	98.09±.36 <sup>c</sup>	98.58±.36 <sup>c</sup>
Volume (ejaculate/ml)	00.3100±.05	00.33±.05	00.34±.05

Values with different superscripts differ significantly (P<0.05).

**Table 3:** Effect of ethanolic extract of *Madhuca longifolia* (flowers) at 14 days on below various semen parameter on male poultry birds - (n=8) (mean ± S.E.,)

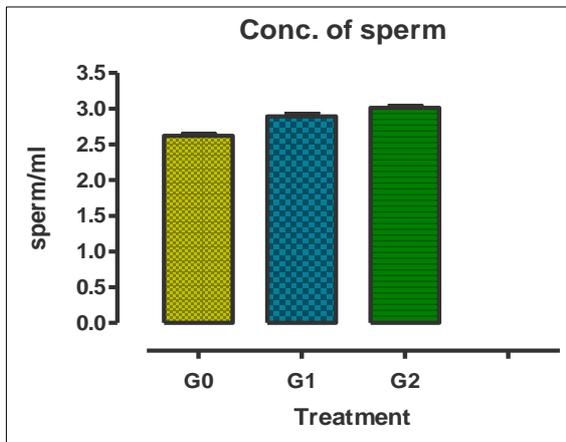
Parameter	Control	Treated with oral dose/10 LD <sub>50</sub> of <i>Madhuca</i>	Treated with oral dose/5 LD <sub>50</sub> of <i>Madhuca</i>
	G0	G1	G2
Concentration of spermatozoa (10 <sup>9</sup> Sperm/ml)	2.62±.03 <sup>c</sup>	2.93±.03 <sup>d</sup>	3.09±.04 <sup>e</sup>
Live Spermatozoa (%)	97.61±.35 <sup>c</sup>	98.35±.34 <sup>c</sup>	98.74±.35 <sup>c</sup>
Volume (ejaculate/ml)	00.31±.05	00.34±.05	00.35±.05

Values with different superscripts differ significantly (P<0.05).

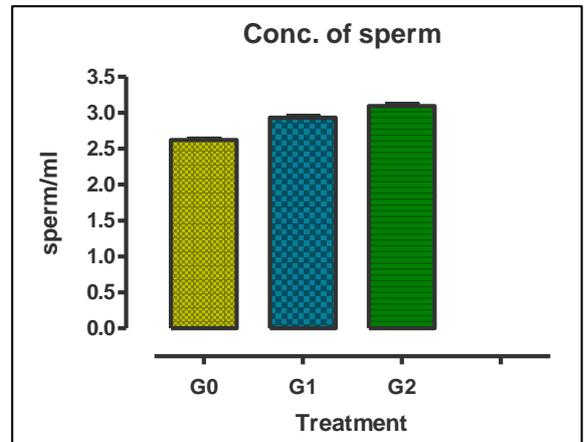
**Table 4:** Effect of ethanolic extract of *Madhuca longifolia* (Flowers) at 28 days on below various semen parameter on male poultry birds - (n=8) (mean ± S.E.,).

Parameter	Control G0	Treated with oral dose/10 LD <sub>50</sub> of <i>Madhuca</i> G1	Treated with oral dose/5 LD <sub>50</sub> of <i>Madhuca</i> G2
Concentration of spermatozoa (10 <sup>9</sup> sperm/ml)	2.62±.029 <sup>c</sup>	3.01±.03 <sup>d</sup>	3.14±.04 <sup>e</sup>
Live spermatozoa (%)	97.61±.35 <sup>c</sup>	98.58±.36 <sup>cd</sup>	99.01±.34 <sup>d</sup>
Volume(ejaculate/ml)	00.31±.05 <sup>ab</sup>	00.34±.05 <sup>b</sup>	00.37±.15 <sup>b</sup>

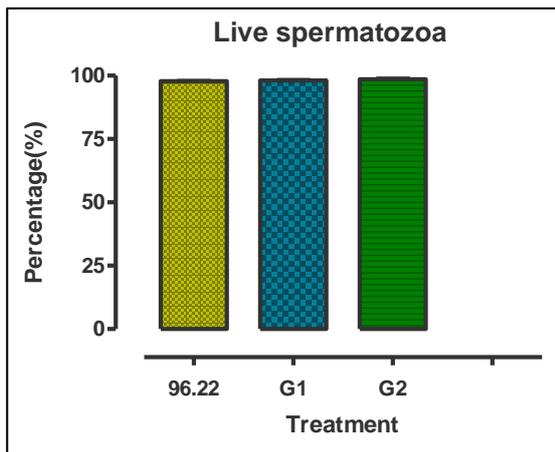
Values with different superscripts in a differ significantly (P<0.05).



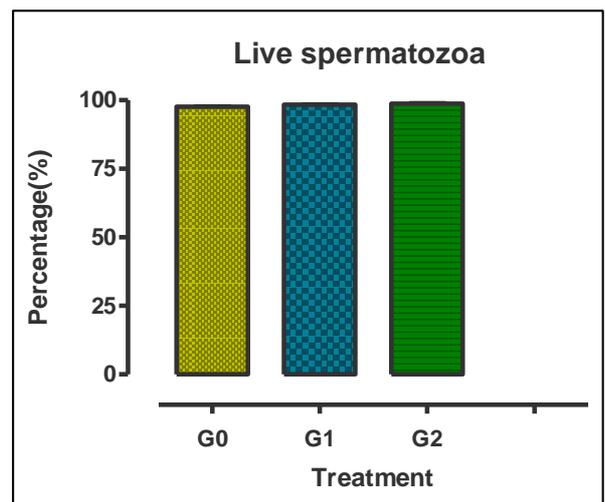
**Fig 1:** Effect *Madhuca* flower (1/10<sup>th</sup> & 1/5<sup>th</sup> of LD<sub>50</sub>) on conc. of sperm after 7<sup>th</sup> days of semen evaluation in poultry (n=8)



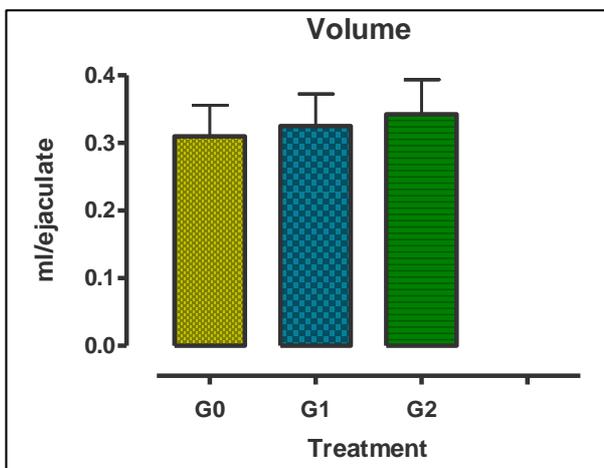
**Fig 4:** Effect *Madhuca* flower (1/10<sup>th</sup> & 1/5<sup>th</sup> of LD<sub>50</sub>) on conc. of sperm after 14<sup>th</sup> days of semen evaluation in poultry (n=8)



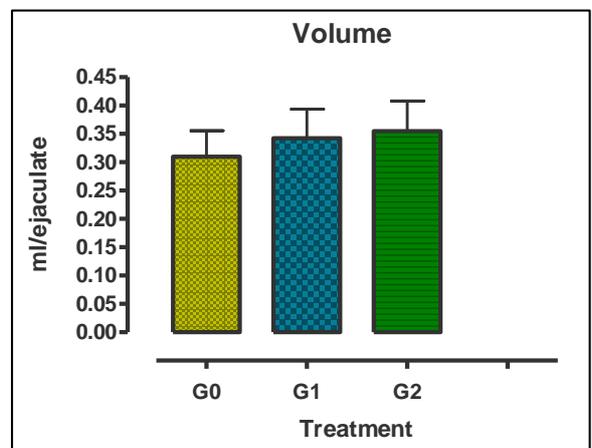
**Fig 2:** Effect *Madhuca* flower (1/10<sup>th</sup> & 1/5<sup>th</sup> of LD<sub>50</sub>) on live spermatozoa after 7<sup>th</sup> days of semen evaluation in poultry (n=8)



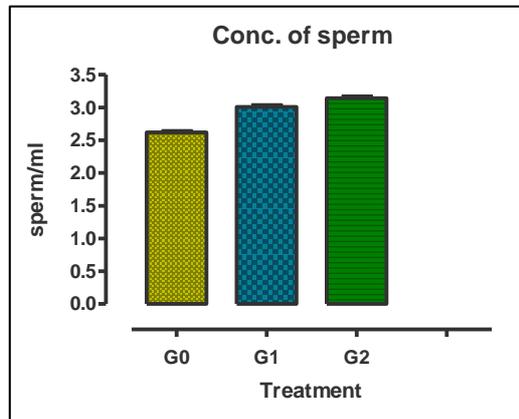
**Fig 5:** Effect *Madhuca* flower (1/10<sup>th</sup> & 1/5<sup>th</sup> of LD<sub>50</sub>) on live spermatozoa after 14<sup>th</sup> days of semen evaluation in poultry (n=8)



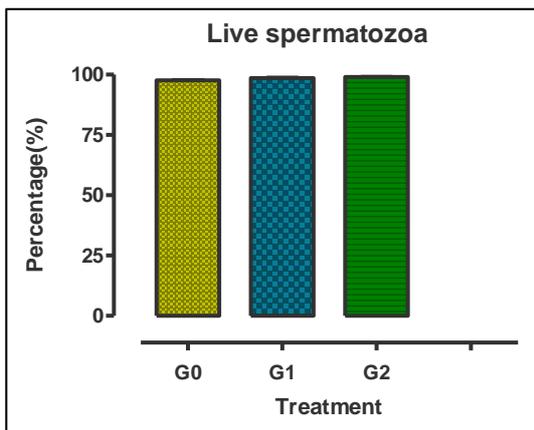
**Fig 3:** Effect *Madhuca* flower (1/10<sup>th</sup> & 1/5<sup>th</sup> of LD<sub>50</sub>) on volume of semen after 7<sup>th</sup> days of semen evaluation in poultry (n=8)



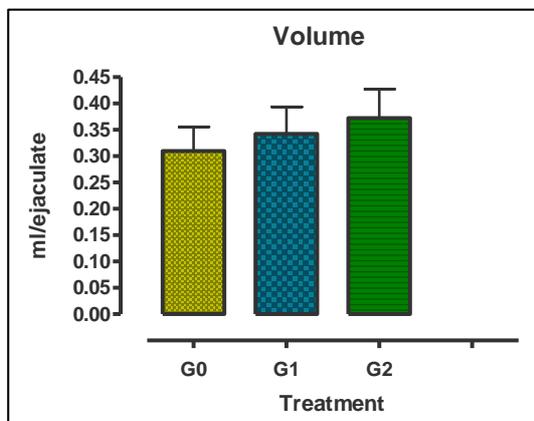
**Fig 6:** Effect *Madhuca* flower (1/10<sup>th</sup> & 1/5<sup>th</sup> of LD<sub>50</sub>) on volume of semen after 14<sup>th</sup> days of semen evaluation in poultry (n=8)



**Fig 7:** Effect *Madhuca* flower (1/10<sup>th</sup> & 1/5<sup>th</sup> of LD<sub>50</sub>) on conc. of sperm after 28<sup>th</sup> days of semen evaluation in poultry (n=8)



**Fig 8:** Effect *Madhuca* flower (1/10<sup>th</sup> & 1/5<sup>th</sup> of LD<sub>50</sub>) on live spermatozoa after 28<sup>th</sup> days of semen evaluation in poultry (n=8)



**Fig 9:** Effect *Madhuca* flower (1/10<sup>th</sup> & 1/5<sup>th</sup> of LD<sub>50</sub>) on volume of semen after 28<sup>th</sup> days of semen evaluation in poultry (n=8)

Effect of these extracts on various semen parameters (sperm conc., live and volume) at 7 days are presented in table. No. 2 and showed by graph no. 1, 2, and 3.

The bird treated with *Madhuca* flowers extract showed positively aphrodisiac effect by increase in value of all the semen parameter. Sperm concentration was increase in both group 1 and group 2 but in group 2 sperm concentration increase more significantly in group 1. Live spermatozoa was also increase in group 1 and group 2, in group 2 live sperm percentage was significantly higher than group. Semen volume was also increase and found greater in group 2 in comparison to control group.

After 14 days effect of *Madhuca* represented in table no 3 and showed by graph no. 4, 5 and 6. At 7 days both dose (1/5 and

1/10 LD<sub>50</sub>) of *Madhuca longifolia* flowers extract had positively significant result in all the parameters. In group 2 where 1/5 dose of *Madhuca* was given result was highly positively significant in comparison to group 1.

After 28 days effect of *Madhuca* represented in table no. 4 and showed by graph no. 7, 8, and 9. At the 28 days both doses of *Madhuca* were continue to increase the value of all the parameter (sperm conc., live and volume). The animal of group 2 exhibited more positively significant in comparison of group 1.

Comparing 1/10<sup>th</sup> & 1/5<sup>th</sup> of LD<sub>50</sub> of *Madhuca*, 1/5<sup>th</sup> showed better result i.e. better aphrodisiac effect. These finding suggests that *Madhuca* has positive aphrodisiac effect [20]. Similar findings were observed at 14 and 28 days, but there was a continuous improvement recorded in all the parameters.

## Conclusions

On the basis of these findings we can say that *Madhuca* has positive aphrodisiac effect. There is a scope of further investigation regarding *Madhuca* flower extract.

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## Competing Interests

The authors declare that they have no competing interests.

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