

ANTIFERTILITY EFFECT OF *MADHUCA LATIFOLIA* (ROXB.) MACBRIDE SEED EXTRACT

Bindu Gopalkrishnan* Late. Dr. Shardha N. Shimpi

K .V. Pendharkar College, Dombivili (East), Maharashtra-421203

*Mithibai College of Arts, Chauhan Institute of Science & Amrutben Jivanlal College of
Commerce and Economics, Vile Parle (West), Mumbai-400056
E mail: bindu_phd@rediffmail.com

ABSTRACT: The study involves a standard pharmacological model to test antifertility activity of seeds of *Madhuca latifolia* (Roxb.) Macbride. When the aqueous powdered drug (2gm/body weight) was administered to male albino rats has proved to be an effective antifertility drug. The activity was confirmed by significant decrease in sperm count, biochemical assays so also through histopathological investigations. Hence seeds of *Madhuca latifolia* can be a reliable herbal option after the necessary clinical trials.

Key words: Pharmacology, Antifertility, Male contraceptive, *Madhuca latifolia*.

INTRODUCTION

Exponentially growing population has been adversely affecting the social, economical and technological development of human race. Therefore to reduce/control our number has to be the first on a priority list. A good number of synthetic contraceptives are available in market, each one with either a limited success or side effects. Since herbal drugs are easily available and with no side effects the current study was undertaken.

Madhuca latifolia (Roxb.) Macbride a tall tree commonly known as "Mahuwa" belong to family Sapotaceae [2]. The fruit is edible and medicinally important for curing many ailments. Seeds are used in the treatment of chronic rheumatism, headache, piles, constipation, hemorrhoids, oedema, spermicidal activity etc. [9]. To prove the folklore claims of *Madhuca latifolia* seed as male contraceptive, antifertility activity on male albino rats were carried out.

MATERIAL AND METHODS

Collection and preparation of plant extract

Seeds of *Madhuca latifolia* (Roxb.) Macbride were collected in the month of May from Murbad district, Maharashtra. The seeds were selected in a ripened stage from the mature tree. The specimen was identified from Blatter Herbaria at St. Xavier's College, Mumbai, Maharashtra. A voucher specimen was deposited for future use at K.V. Pendharkar College, Dombivli (E), Maharashtra. (Specimen Number: KVP/BOT/ OO175). Seeds were separated from the ripened fruit and were dried in shade. A fine powder was prepared by grinding the dry seeds in a blender. This powder was stored in airtight container and was used to feed the experimental rats.

Animals: Adult male albino rats, Wistar strain weighing 150-200 gm body weight used for the study were housed under standard laboratory condition. They were fed with standard rodent pellet and water *ad libitum*. The animals were grouped in to two groups of 6 animals each.

- a. Control group - Rats receiving 1% gum acacia for 21 days .
- b. Group II : Rats receiving *Madhuca latifolia* seed powder dissolved in 1% gum acacia (2gm/ kg body weight) for 21 days.

The study was approved by the ethics committee for animal experimentation, the registration no. 525/02/a/CPCSEA.

Acute Toxicity Study: An acute toxicity study was done to select the dose. The dose up to 2gm/ body weight did not produce any sign of toxicity and mortality. The animals were physically active [4 and11] .

Antipyretic test: The crude extract was dissolved in 0.5 ml 1% gum acacia and administered orally to Group II rats for 21 days. On day 22 i.e. 24 hrs after the last dose the animals were sacrificed using ether anesthesia. Reproductive organs were weighed and dissected out [6 and 14].

Bioassay of serum: Blood was collected by cardiac puncture and serum was separated and estimated for protein, cholesterol, alkaline phosphatase and acid phosphatase [7, 8, and 12]

Bioassays of testes homogenate: 0.5 mg of tissue was procured from the testis after sacrificing the animals. This was homogenized with 1ml saline using homogenizer. The homogenate was also analyzed for protein, cholesterol, alkaline phosphatase and acid phosphatase [12].

Hormonal assay: Serum testosterone levels were assayed from frozen samples using radio immuno assay method [1, 3 and 5].

Sperm count: Sperm count were assessed in cauda epididymis by the standard methodology [13] .

Histopathological preparation: The testis were fixed in Bouin's fluid, paraffin sections were made and stained with hematoxylin and eosin. The sections were screened for histopathological effect of the drug [15].

Statistical analysis: The mean and standard error of mean (SEM) were calculated using Student's t- test [10].

RESULTS

Weight response – The orally tested *Madhuca latifolia* seed extract caused decrease in the weight of testes, epididymis, seminal vesicle + coagulating gland, vasa deferens and ventral prostrate significantly from the control. (Table 1, Graph 1)

Sperm count – There was a decrease in sperm count in tested rats compared to the control rats. (Table 2, Graph 2)

Hormonal assay – Serum testosterone level of *Madhuca latifolia* seed extract treated animals was decreased significantly in comparison to control group (Table 2, Graph 3)

Table 1 : Effect of Madhuca latifolia seed extract on organ weights of male albino rats. [Values are mean ± SE of six determinations]

Groups	Organ weight				
	Testes mg/100g	E.D. mg/100g	V.D. mg/100g	S.V. + C.G. mg/100g	V.P. mg/100g
Control rats	1.4 ± 0.05	0.28 ± 0.02	0.06 ± 0.004	0.15 ± 0.02	0.07 ± 0.02
Madhuca Latifolia seed fed rats	1.25 ± 0.04*	0.27 ± 0.008*	0.05 ± 0.004*	0.07 ± 0.006*	0.04 ± 0.004*

*P<0.1vs Control

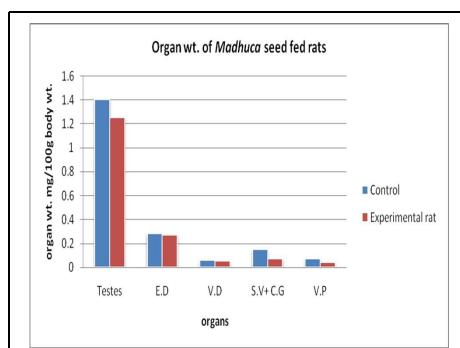
E.D. = Epididymis, V.P. = Ventral prostrate, V.D. = Vasa deferens,

S.V. = Seminal vesicle, C.G. = Coagulating gland

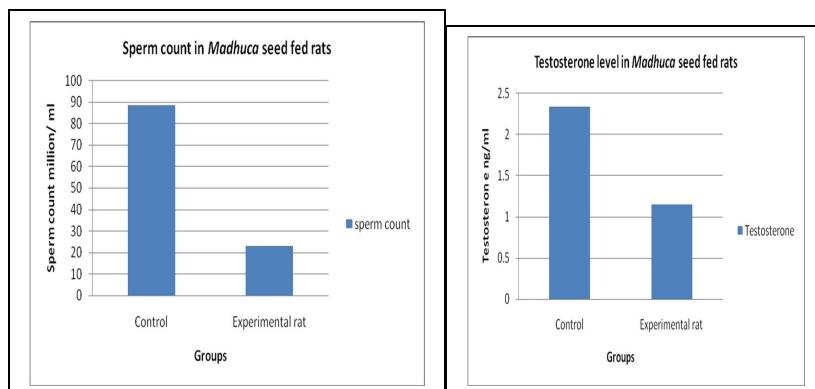
Table 2 : Sperm count and Testosterone level of Madhuca latifolia seed fed male albino rats [Values are mean \pm SE of six determinations]

Groups	Sperm count (million/ml)	Testosterone (ng/ml)
Control rats	88.3 \pm 0.81	2.33 \pm 0.01
Madhuca latifolia seed fed rats	23.0 \pm 1.97	1.15 \pm 0.007

*P<0.02 vs Control



Graph-1



Graph-2

Graph-3

Biochemical findings- A marked reduction in protein, cholesterol, alkaline phosphatase and acid phosphatase in both serum and testes homogenate was observed in treated rats. (Table 3 &and 4, Graph 4 and 5).

Table 3 : Serum biochemistry of Madhuca latifolia seed fed male albino rats [Values are mean \pm SE of six determinations]

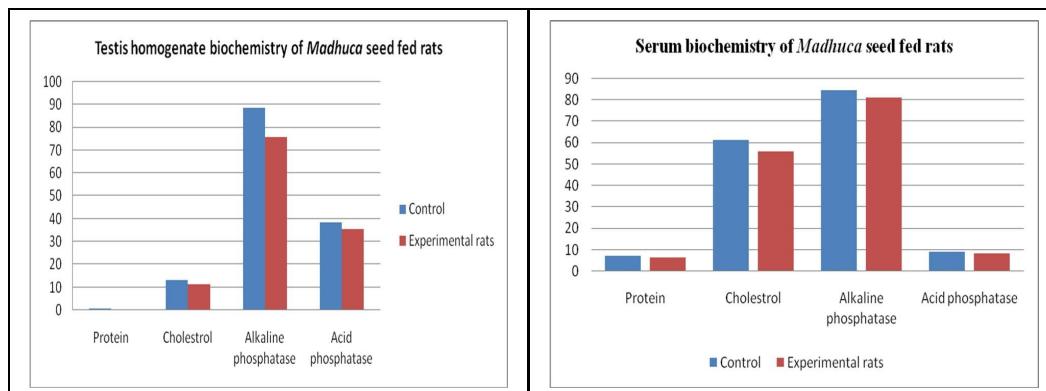
Groups	Protein mg/ml	Cholesterol mg/dl	Alkaline phosphatase u/l	Acid phosphatase u/l
Control	7.3 \pm 0.28	61.3 \pm 4.9	84.8 \pm 29.9	9.31 \pm 2.51
Madhuca latifolia seed fed rats	6.45 \pm 0.44*	56.00 \pm 1.7*	81.1 \pm 1.46**	8.40 \pm 0.17*

* P<0.1; **P<0.01vs Control

Table 4 : Testis homogenate biochemistry of *Madhuca latifolia* seed fed male albino rats [Values are mean \pm SE of six determinations]

Groups	Protein mg/ml	Cholesterol mg/dl	Alkaline phosphatase u/l	Acid phosphatase u/l
Control	0.7 \pm 0.02	13.1 \pm 0.2	88.3 \pm 8.2	38.2 \pm 2.5
Madhuca latifolia seed fed rats	0.42 \pm 0.01**	11.3 \pm 0.85***	75.7 \pm 12.4*	35.5 \pm 1.4*

*P<0.1; **P<0.001; ***P<0.02 vs Control



Graph-4

Graph-5

Histopathology of testis – Administration of crude extract caused an effective inhibition of spermatogenesis in male albino rats. Shrinkage in few seminiferous tubules was seen. Thinning of basement membrane of seminiferous tubule was prominent. There was an increase in the diameter of lumen, compared to control. Distance between the interstitial spaces showed widening within the two seminiferous tubules. Disruption of sertoli cell was significant. (Figs 1 and 2).

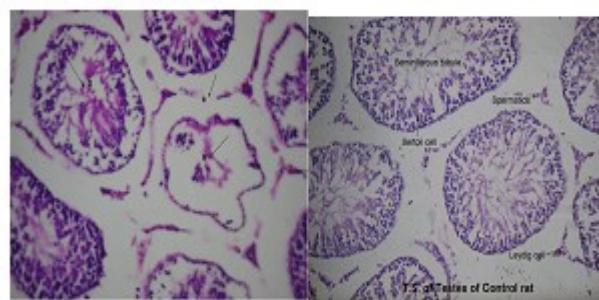


Fig. 1 T.S. of testes of control male albino rat

- a. Shrunken seminiferous tubule , absence of sperms
- b. Inhibition of spermatogenesis
- c. Thinning of basement membrane of seminiferous tubule.

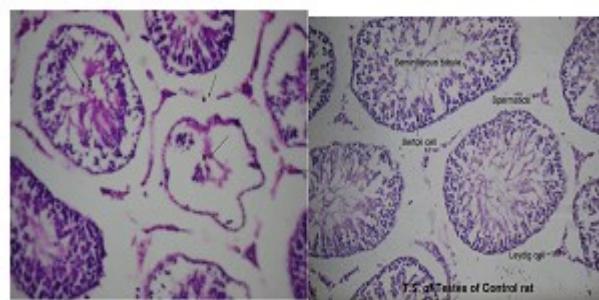


Fig. 2 T.S. of testes of experimental male albino rat

DISCUSSION

In the present study, administration of *Madhuca latifolia* seed extract to rats caused decrease in weight of testes, epididymis, seminal vesicle + coagulating gland, vasa deferens and ventral prostrate which may be due to low plasma level of testosterone. The decrease in weight of accessory sex organs indicates the atrophy of glandular tissue and also reduction in secretory cells thus reflecting the decrease level of testosterone. The decrease in sperm count might be due to partial arrest of spermatogenesis as it was also confirmed by Histopathological findings. A decrease in sperm reserve may be a reasonable cause for reduction in the weight of epididymis. The effect of the said drug on the sex organ has also affected the biochemical assay of serum and testes homogenate.

Thus, the organ weight, biochemical and hormonal assay along with sperm count go concurrent with histopathological evidences. These results confirm that, the seed of *Madhuca latifolia* have antifertility potentials in male albino rats. Further clinical trials will confirm the safety and efficacy of the said drug of plant origin. The crude seed extracts are given to the rats due to the presence of mixture of bioactive compounds, hence detailed phytochemical analysis will reveal the exact compound responsible for antifertility activity.

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