Pharmacogn. Res., 2026; 18(4):1379-1385.
https://www.phcogres.com

Original Article

# *Pongamia pinnata* Stem Bark Extract Induced Changes in Testicular Function in Male Wistar Rats

**Kusum Jain[1,*], Suresh Chandra Joshi[2]**

[1]Department of Zoology, Shri Mahaveer College, Jaipur, Rajasthan, INDIA.
[2]Department of Zoology, University of Rajasthan, Jaipur, Rajasthan, INDIA.

## ABSTRACT

**Background:** *Pongamia pinnata* (Linn.) Pierre (Fabaceae), commonly known as Karanja, is widely used in traditional medicine for the treatment of various disorders and is known to possess diverse pharmacological activities. **Objectives:** The present study was designed to evaluate the effects of a 50% ethanolic stem bark extract of *Pongamia pinnata* on testicular function in adult male Wistar rats, with the aim of exploring its potential for the development of a plant-based male antifertility agent. **Materials and Methods:** Twenty-eight adult male Wistar rats were randomly divided into four groups ($n$ = 7). Group I served as the control and received vehicle only, while Groups II, III, and IV were administered the stem bark extract orally at doses of 100, 200, and 300 mg/kg body weight/day, respectively, for 60 days. At the end of the treatment period, testicular weights were recorded, testes were subjected to histopathological and histomorphometric analyses and serum testosterone levels were estimated. **Results:** Administration of *P. pinnata* extract resulted in a significant, dose-dependent reduction in testicular weight and serum testosterone levels compared to controls. Histopathological examination revealed pronounced degenerative alterations in the seminiferous tubules, including disorganization of germinal epithelium, tubular shrinkage, reduced germ cell population, decreased luminal spermatozoa, and atrophy of Leydig cells. **Conclusion:** Chronic oral exposure to the 50% ethanolic stem bark extract of *P. pinnata* adversely affects testicular structure and function in male rats, indicating its potential utility as a male antifertility agent.

**Keywords:** Antifertility, Germ cell population, *Pongamia pinnata*, Testes, Testosterone.

**Correspondence:**
**Dr. Kusum Jain**
Department of Zoology, Shri Mahaveer College, Jaipur, Rajasthan, INDIA.
Email: 22kusumjain@gmail.com

**Received:** 04-02-2026;
**Revised:** 28-04-2026;
**Accepted:** 17-06-2026.

## INTRODUCTION

The unprecedented growth of the global population represents one of the most significant challenges of the modern era. Overpopulation has emerged as a critical global concern, as it directly or indirectly contributes to nearly all major worldwide problems. Issues such as poverty, malnutrition, depletion of natural resources, environmental pollution, global warming, loss of biodiversity, and even armed conflicts are exacerbated by population pressure. Consequently, the regulation of human fertility through effective limitation strategies has become one of the most urgent biosocial and medical challenges confronting humanity today (Hoesl *et al.*, 2005).

Current statistics indicate that the global population has reached approximately 8.2 billion in 2024 and is projected to peak at around 10.3 billion by the mid-2080s. At present, India, with a population of about 1.38 billion, is the second most populous country in the world. Furthermore, projections suggest that India is expected to surpass China and become the world's most populous nation by 2027 (Bhardwaj *et al.*, 2025).

The current scenario necessitates the development of novel contraceptive options that are simple, safe, reversible, and cost-effective, and that are broadly acceptable across diverse cultures, religions, and racial groups worldwide. The availability of such methods would encourage greater adoption by couples, thereby contributing significantly to the reduction of population growth rates (Anderson and Baird, 2002; Page *et al.*, 2008).

There are multiple target sites in the male reproductive physiology for functional disruption in order to develop novel male contraceptives. These include suppression of spermatogenesis in the testes, impairment of sperm maturation within the epididymis, prevention of sperm transport to the female reproductive tract, interference with sperm capacitation, and disruption of sperm functions essential for normal fertilization (Bhardwaj *et al.*, 2025).

Extensive research has been conducted in the field of male contraception. Various hormonal approaches, including androgens, progestogens, estrogens, and GnRH agonists or antagonists, alone or in combination as well as immunological

OPEN ACCESS
Manuscript TECHNOMEDIA

**DOI:** 10.5530/pres.20260004



**Publishing Partner :** Manuscript Technomedia. [www.mstechnomedia.com]

and chemical agents, have been investigated. However, none of these methods has yet proven to be completely effective, safe, and free from adverse side effects (Page *et al*., 2008; Tulsiani and Abou-Haila, 2008).

In recent decades, considerable attention has been directed toward medicinal plants documented in classical Materia Medica, Ayurvedic literature, and folk medicine for their potential antifertility effects in males. Numerous plant species have been systematically screened worldwide, revealing promising leads for the development of plant-based male contraceptives (Unny *et al*., 2003; Obuewu *et al*., 2011).

The development of novel fertility-regulating drugs derived from medicinal plants represents an attractive proposition, as plant-based contraceptives are generally more acceptable due to their cost-effectiveness, easy availability, and comparatively fewer adverse side effects. Traditional medicine continues to serve as the primary healthcare system for approximately 70-80% of the global population, particularly in developing countries. Medicinal plants constitute a rich source of diverse bioactive phytochemicals that may act either individually or synergistically to exert antifertility effects, offering valuable leads for drug development based on phytochemical investigations (Rates, 2001; Gelani and Rehman, 2005).

*Pongamia pinnata* (Linn.) Pierre is a medium-sized, glabrous tree commonly known as Karanja in Hindi and Indian beech in English. It is an important medicinal plant predominantly found in the tidal forests of India and has long been used in the traditional Indian system of medicine for the treatment of bronchitis, whooping cough, rheumatic arthritis, and diabetes. Numerous pharmacological investigations have demonstrated that *P. pinnata* exhibits a wide spectrum of biological activities, including antioxidant, antimicrobial, antiparasitic, anti-inflammatory, anticonvulsant, antidiabetic, antihyperammonemic, cytotoxic, anthelmintic, insecticidal, and immunomodulatory properties (Akram *et al*., 2021; Kyatham *et al*., 2024).

Phytochemical studies of *P. pinnata* have revealed the presence of a diverse array of bioactive constituents. Flavonoids and their derivatives represent the most commonly isolated compounds, including flavones, flavans, and chalcones. In addition, several other classes of phytochemicals such as sesquiterpenes, diterpenes, triterpenes, steroids, amino acids, disaccharides, fatty acids, and ester compounds have been identified (Muquarrabun *et al*., 2013; Kyatham *et al*., 2024). Notably, seven flavonoids; pongaflavone, karanjin, pongapin, pongachromene, 3,7-dimethoxy-3′,4′-methylenedioxyflavone, millettocalyxin C, and 3,3′,4′,7-tetramethoxyflavone have been isolated from the stem bark of *P. pinnata* (Yin *et al*., 2004; Muquarrabun *et al*., 2013).

Despite its prominent status in Ayurvedic literature and traditional medicine, there is a paucity of scientific data regarding the effects of *P. pinnata* on male reproductive function. Therefore, the present study was undertaken to evaluate the potential contraceptive efficacy of the 50% ethanolic stem bark extract of *P. pinnata* in male rats.

## MATERIALS AND METHODS

### Plant material and Method of extraction

The plant material was collected from the University of Rajasthan Campus and authenticated at the Herbarium, Department of Botany, University of Rajasthan, Jaipur. A voucher specimen of the plant has been deposited in the herbarium (RUBL-20255). The collected stem bark was shade-dried, ground into coarse powder, and extracted with 50% ethanol using a soxhlet apparatus for 36 hr at 60-80°C. The crude extract obtained was filtered and concentrated to dryness under reduced pressure at low temperature (40°C). The resulting viscous brown residue was then suspended in an appropriate volume of distilled water and used for experimental administration.

### Experimental animals

Adult, healthy male albino rats (Wistar strain) of proven fertility were used in the present study. The rats were housed in groups in polypropylene cages (12" × 10" × 8") under standard laboratory conditions, with a 14 hr light: 10 hr dark cycle and a controlled temperature of 22 ± 3°C. Animals were provided with water and a standard laboratory diet (Lipton India Pvt. Ltd.,) *ad libitum*, and occasionally supplemented with soaked grams and leafy vegetables. Animal care and handling were conducted in accordance with the guidelines of the Indian National Science Academy (INSA, 1992, New Delhi, India) for the maintenance and use of laboratory animals. Experimental procedures were approved by the Institutional Animal Ethics Committee (Department of Zoology, University of Rajasthan, Jaipur).

### Experiment design

Rats of nearly equal size; weight (170-210 g) were selected and than randomly divided into four groups each having seven rats and treated as follows:

**Group I:** Control rats treated with vehicle (distilled water, 0.3 mL/rat/day) for 60 days.

**Group II:** Rats treated with *P. pinnata* extract (100mg/kg b.wt./day) orally, suspended in distilled water for 60 days.

**Group III:** Rats treated with *P. pinnata* extract (200 mg/kg b.wt. / day) orally, suspended in distilled water for 60 days.

**Group IV:** Rats treated with *P. pinnata* extract (300 mg/kg b.wt. / day) orally, suspended in distilled water for 60 days.

24 hr after administration of the lost dose, all the overnight fasted rats from various treated groups were sacrificed under ether anesthesia. Blood samples were collected by cardiac puncture.

The blood sample was allowed to clot at 37°C and the serum was separated by centrifugation and stored at-20°C for biochemical analysis.

## Testicular weight

Testicular weights of all rats were recorded at the end of the treatment. Testes and other reproductive organs were dissected out, cleaned of adherent fat and blood clots, and weighed individually on a digital electronic balance. One half of the reproductive tissues was fixed in bouin's fluid for histopathological examination, while the remaining half was stored at -20°C for biochemical estimations in future.

## Serum testosterone

Serum level of testosterone hormone was measured by Chemiluminiscence-CIA by autoanalyzer.

## Histological study

For histological observation, bouin's fixed tissues (testis) were washed in water to remove excess of fixative, dehydrated in graded series of alcohol, cleared in xylene, embedded in paraffin wax and sectioned at 5 µm and counter stained in eosin for the cytoplasmic contrast. Sections were observed for histopathological effects using a Nikon digital camera mounted on a light microscope.

## Germ cell population and Histomorphometric analysis

### *Quantitative evaluation of germ cells*

Quantitative evaluation of germ cells were made using 50 round tubules per group selected randomly at X 400 according to method described by Leblond and Clermont (1952). Quantitative evaluation of spermatogonia, preleptotene, mid-pachytene spermatocytes: round and elongated spermatids were made in stage VII of seminiferous tubule cycle per cross section under X100 magnification. Counting was performed in 10 tubule cross section and mean was calculated.

The diameters of nuclei of various germ cell types were measured by mean of an ocular micrometer. A correction factor was used to obtain the actual numerical density of germ cells (Abercrombie, 1946).

$$\text{True counts} = \text{Crude Counts} \times \frac{\text{Section thickness (µm)}}{\text{Section thickness (µm)} + \text{Nuclear diameter (µm)}}$$

## Seminiferous tubule diameter

Measurements were taken from at least 40 tubular profiles per animal using a light microscope equipped with ocular micrometer caliberated with stage micrometer. Only round tubules were selected randomly in various areas of the section. Two diameters perpendicular to each other were measured at X 100 magnification and averaged and expressed as seminiferous tubule diameter.

## Leydig cell area and nuclear diameter

The diameter of one hundred Leydig cell and their nuclei were measured on five sections from each testis with ocular micrometer at X 1000. The values were averaged and expressed as mean nuclear diameter of the Leydig cells.

## Statistical analysis

All the data were calculated and statistically analyzed with SPSS\V.13.0 computer software package for Windows (SPSS Inc., Chicago, IL., USA). The data were expressed as Mean ± SEM and tested for variance. When variance was homogenous, one way Analysis of Variance (ANOVA) was applied out to determine the significance of observed differences, the test. An alpha probability of less than 5 % ($p$<0.05) was considered as statically significant. All other data were statistically analyzed with one way ANOVA for the comparison between the groups followed by Tukey's multiple comparison procedure as a *post hoc* test.

# RESULTS AND OBSERVATIONS

## Testicular weight

Treatment with *P. pinnata* extract resulted in a significant ($p$ < 0.05), dose-dependent reduction in relative testicular weight in all treated groups (100, 200, and 300 mg/kg body weight/day). The decrease was significant not only when compared with the

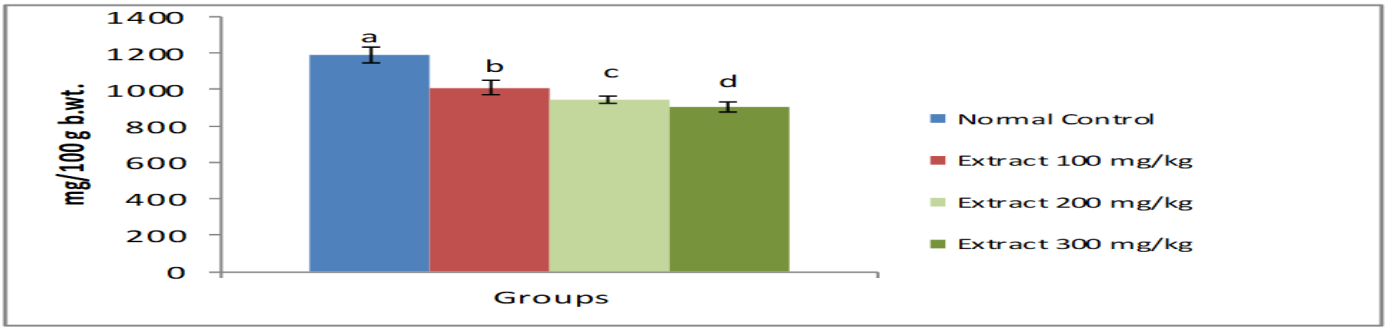


**Figure 1:** Effect of *Pongamia pinnata* on testicular weight. *Similar letters above data points represents no significant different according to Tukey's multiple comparison procedure (at $p$< 0.05).

control group but also among the treated groups themselves (Figure 1).

## Serum testosterone

Serum testosterone levels in rats treated with *P. pinnata* extract exhibited a significant, dose-dependent decline compared with the control group, as well as upon multiple comparisons among the three treated groups (100, 200, and 300 mg/kg body weight/ day). The mean serum testosterone concentration in control rats was 3.81 ng/mL, which decreased to 2.25 ± 0.11 ng/mL in the highest-dose treatment group (Figure 2).

## Histopathology

The histological picture of the testis of control rats displayed normal process of spermatogenesis. The seminiferous epithelium showed characteristic tiered arrangement of all successive germ cell types. The tubular lumen was fully occupied by large number of healthy spermatozoa. The presence of large number of normal Leydig cells was also conspicuous in the interstitium (Figures 3A & 3B, Table 1).

The histoarchitecture of the testis in rats receiving *P. pinnata* extract (100 mg/kg/day) showed degenerative changes. The diameter of seminiferous tubules was reduced ($p<0.05$). There was significant ($p<0.05$) decline in the number of round and elongated spermatids. The characteristic tiered arrangement was also disturbed. The number for spermatogonia and preleptotene spermatocyte remained unaffected. The Leydig cells showed atrophic changes (Figures 3C & 3D, Table 1).

The histoarchitecture of the testis in rats treated with 200 mg/ kg/day dose of *P. pinnata* extract showed marked degenerative changes and disruption of spermatogenesis. The seminiferous tubules were reduced in size and showed disruption of normal epithelial organization. Quantitative determination of germ cells in seminiferous epithelium showed a decline in the numbers of preleptotene and pachytene spermatocyte ($p<0.05$), round and elongated spermatids ($p<0.05$). However, the number of spermatogonia remained significantly unaltered. Additionally, several cells with pycnotic nuclei could be identified. The lumen showed the presence of exfoliated germ cells, sperm debris and few spermatozoa. The Leydig cells were shrunken and showed atrophic changes (Figures 3E & 3F, Table 1).

Atrophic changes in the testis were more pronounced at 300 mg/kg/day dose of the extract. The diameter of seminiferous tubules was further decreased. There was significant decrease in the numbers of spermatogonia ($p<0.05$) preleptotene and pachytene primary spermatocytes, ($p<0.05$), round ($p<0.05$) and elongated spermatids ($p<0.05$). The lumen contained tail reminants of degenerating spermatozoa. Leydig cells also showed marked degeneration, decrease in nuclear diameter and decline in numbers (Figures 3G & 3H, Table 1).

It appears that *P. pinnata* extract induced dose dependent degenerative, atrophic changes and disruption of spermatogenesis in treated male rats.

## Germ cell population and Histomorphometric analysis

The testicular germ cells population i.e. spermatogonia, preleptotene spermatocyte, pachytene spermatocytes, round, elongated spermatids counts/seminiferous tubule cross section, Seminiferous tubule diameter and Leydig cell nuclear diameter are presented in Table 1.

The control value of spermatogonia in per seminiferous tubule was 6.92 ± 0.42. After 60 days of the *P. pinnata* extract treatment a non-significant decline in mean spermatogonia numbers was observed in both 100 mg/kg and 200 mg/kg/ b.wt/ day dose groups. However, in 300 mg/kg b.wt. dose group of *P. pinnata* the decline was significant ($p<0.05$). The mean counts of preleptotene spermatocytes per seminiferous tubule cross section were 20.14±1.02. Treatment of *P. pinnata* extracts at three different doses (100, 200 and 300 mg/kg/ b.wt/day) showed dose dependent significant ($p<0.05$) diminution of preleptotene primary spermatocyte population when compared to control or the three groups compared with each other except in 100mg/kg groups of *P. pinnata*, where the decline was not significant. The population of pachytene spermatocyte in the seminiferous tubules was declined dose dependently ($p<0.05$) in rats treated with *P. pinnata* extract at three different doses. There was significant ($p<0.05$) dose dependent decline in the populations of round and

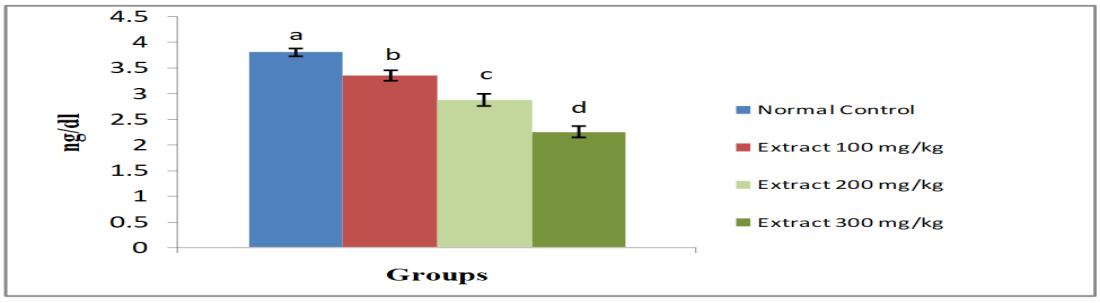


**Figure 2:** Effect of *Pongamia pinnata* on testosterone hormone. *Similar letters above data points represents no significant different according to Tukey's multiple comparison procedure (at $p< 0.05$).

**Table 1: Effects of *Pongamia pinnata* on Germ cell population and Histomorphometric analysis *Values for a given group in a row followed by same letter as superscript are not significantly different according to Tukey's multiple comparison procedure (at $p < 0.05$).**

| Treatment | Control | *Pongamia pinnata* | | |
|---|---|---|---|---|
| Parameter | | 100 mg/kg b.wt. | 200 mg/kg b.wt. | 300 mg/kg b.wt. |
| Spermatogonia | 6.92±0.42$^{a*}$ | 6.79±0.24 $^{a}$ | 5.94±0.66 $^{a}$ | 5.37±0.92 $^{b}$ |
| Preleptolene spermatocyte | 20.14±1.02 $^{a}$ | 19.42±0.97 $^{a}$ | 17.85±0.67 $^{b}$ | 16.57±0.75 $^{c}$ |
| Mid-pachytene spermatocyte | 23.92±0.94 $^{a}$ | 20.28±0.93 $^{b}$ | 19.27±0.65 $^{c}$ | 17.07±0.82 $^{d}$ |
| Round spermatid | 42.0±3.02 $^{a}$ | 33.28±1.88 $^{b}$ | 23.85±2.48 $^{c}$ | 15.71±1.04 $^{d}$ |
| Elongated spermatid | 34.66±1.88 $^{a}$ | 22.64±1.66 $^{b}$ | 16.0±1.04 $^{c}$ | 8.42±0.77 $^{d}$ |
| Seminiferous tubular diameter (μm) | 277.85± 9.93 $^{a*}$ | 244.85± 10.24 $^{b}$ | 237.00±9.00 $^{c}$ | 219.71±9.30 $^{d}$ |
| Leydig cell nuclear diameter (μm) | 6.94± 0.09 $^{a}$ | 6.22±0.11 $^{b}$ | 6.11±0.15 $^{c}$ | 5.82±0.15 $^{d}$ |

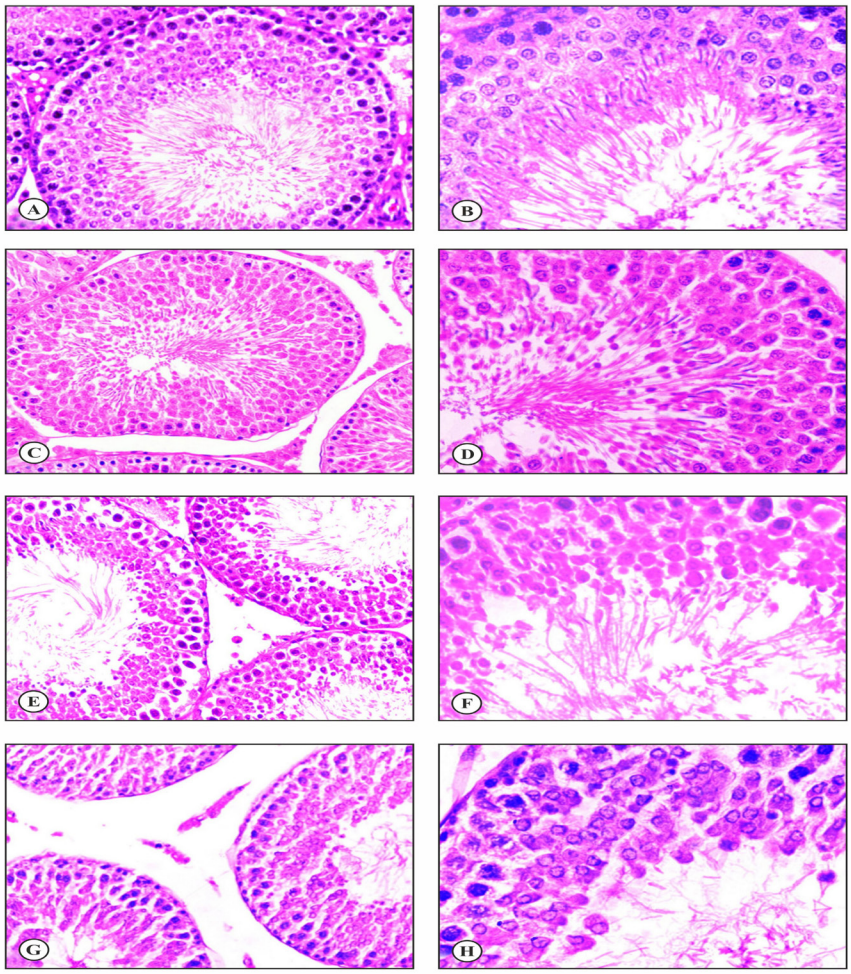


**Figure 3:** Photomicrographs of rat testis showing dose-dependent histological changes following *P. pinnata* extract treatment. The control group exhibits normal testicular histoarchitecture and orderly spermatogenesis with a characteristic tiered arrangement of germ cells (A: ×200; B: ×400). Rats treated with 100 mg/kg/day show degenerative changes, including shrinkage of seminiferous tubules, impaired spermatogenesis, and presence of sperm debris in the tubular lumen (C: ×200; D: ×400). At 200 mg/kg/day, degeneration is more pronounced with arrest of spermatogenesis at the secondary spermatocyte stage, disorganization of the seminiferous epithelium, loss of post-meiotic germ cells, and reduced spermatozoa (E: ×200; F: ×400). Treatment with 300 mg/kg/day results in marked shrinkage of seminiferous tubules, increased intertubular space, severe depletion of germ cells, and profound disruption of spermatogenesis with abundant luminal sperm debris (G: ×200; H: ×400) (H.E.).

elongated spermatids per seminiferous tubules cross section in all the groups of rats treated with *P. pinnata* extract for 60 days.

There was significant ($p<0.05$) dose dependent decrease in the mean diameter of seminiferous tubules in the rats treated with three different doses (100,200 and 300 mg/kg/ b.wt/day) of *P. pinnata* extracts as compared to control and also on multiple comparison test.

Treatment of *P. pinnata* extract at 100,200 and 300 mg/kg/ b.wt/ day,dose levels for 60 days resulted in a significant ($p<0.05$) reduction of Leydig cell nuclear diameter when compared to control and also between three groups themselves.

## DISCUSSION

Traditional medicine has long played a vital role in healthcare systems worldwide, particularly in India, where plant-based remedies form the foundation of indigenous systems such as Ayurveda. The widespread use of medicinal plants is largely attributed to their perceived low toxicity and the long history of human exposure. In this context, the present investigation was undertaken to evaluate the antispermatogenic and antifertility potential of the medicinally important plant *Pongamia pinnata* in male Wistar albino rats.

In the present study, a dose-dependent decline in the relative weight of the testes was observed following administration of *P. pinnata* stem bark extract. Similar reductions in testicular weight have been reported earlier in animals treated with various plant extracts possessing antispermatogenic and antiandrogenic activities (Purohit and Bhagat, 2004; Jain and Meerwal, 2016). The decrease in testicular weight may be attributed to the loss of spermatids and spermatozoa, reduction in seminiferous tubular diameter, and inhibition of steroid biosynthesis by Leydig cells (Nandi *et al*., 1999; Jana *et al*., 2006; Jain and Meerwal, 2016).

A significant dose-dependent decrease in serum testosterone levels was also recorded in rats treated with *P. pinnata* extract. This reduction in androgen levels may result from a direct inhibitory action of the extract on Leydig cells or indirectly through interference with the hypothalamic-pituitary-gonadal axis, leading to reduced gonadotropin secretion and impaired androgen biosynthesis. The observed decline in testosterone levels may be due to the adverse effects of phytoconstituents present in the extract on steroidogenic activity. Phytochemical studies of *P. pinnata* have demonstrated the presence of flavonoids, flavones, and terpenes, compounds known to exhibit antiandrogenic as well as estrogenic properties (Bandivdekar and Moodbidri, 2002; Jain and Meerwal, 2016). It is well established that estrogens inhibit androgen biosynthesis in Leydig cells, thereby affecting normal spermatogenesis (Abney, 1999; Dias *et al*., 2014).

Histopathological examination of the testis is considered the most sensitive endpoint in the evaluation of male reproductive toxicity (Creasy, 2001). In the present study, histological analysis revealed marked dose-dependent degenerative and atrophic changes in the testes of rats treated with *P. pinnata* extract. The seminiferous epithelium showed exfoliation and disorganization of germ cells, along with a significant reduction in the number of preleptotene spermatocytes, pachytene spermatocytes, round and elongated spermatids, and spermatozoa within the lumen. In the higher dose groups, depletion of spermatogonia was also evident. Additionally, the seminiferous tubules exhibited reduced diameter, and Leydig cells showed signs of atrophy.

These histological alterations are likely a consequence of decreased serum testosterone levels, which are essential for maintaining normal spermatogenesis and the structural integrity of seminiferous tubules (Sharpe *et al*., 1988). Seminiferous epithelial sloughing may result from damage to Sertoli cells and disruption of intercellular bridges. Moreover, it is well documented that a decline in intratesticular testosterone induces germ cell apoptosis, further contributing to impaired spermatogenesis (Kim *et al*., 2001; O'Donnell *et al*., 2006; Yang *et al*., 2006). Furthermore, the present observations are consistent with several studies reporting testicular lesions, reduced germ cell populations at various stages of development, and Leydig cell atrophy following treatment with plant extracts exhibiting antiandrogenic activity (Jain and Ali, 2007; Nusier *et al*., 2007; Yakubu *et al*., 2007; Jain and Meerwal, 2016).

Phytochemical investigations have shown that *P. pinnata* stem bark extract is rich in phenylpropanoids, flavonoids, and pongamones (Kitagawa *et al*., 1992; Yin *et al*., 2004; Li *et al*., 2006). Flavonoids and isoflavonoids such as rutin and quercetin are well-known phytoestrogens and have been reported to exert antifertility and antiandrogenic effects in male rats (Bhargava, 1989; Das *et al*., 2004). Farnsworth *et al*. (1975) also reported the antifertility potential of rutin and quercetin. Several estrogenic compounds that bind to estrogen receptors in male reproductive organs have been shown to induce similar degenerative changes and suppression of spermatogenesis through a decline in serum testosterone levels (Boockfor and Blake, 1997; Jana *et al*., 2006; Yang *et al*., 2007).

Overall, the results of the present investigation strongly suggest that the phytoconstituents present in *P. pinnata* stem bark extract exert deleterious effects on male reproductive function, primarily through antiandrogenic and estrogenic mechanisms, leading to impaired spermatogenesis and reduced fertility.

## CONCLUSION

Based on the findings of the present investigation, it can be concluded that the phytoconstituents present in the 50% ethanolic stem bark extract of *Pongamia pinnata*, acting either individually or synergistically, produced marked adverse effects on the testes of male Wistar rats. Administration of the extract led to pronounced degenerative changes in testicular histoarchitecture, along with significant impairment of spermatogenesis. Taken together, these

alterations reflect a compromised functional integrity of the testes, which ultimately resulted in a decline in male fertility.

## ACKNOWLEDGEMENT

The authors gratefully acknowledge the Head of the Department for providing the necessary facilities and extend their sincere thanks to Prof. G. C. Jain for his support and valuable guidance.

## ABBREVIATIONS

**ANOVA:** Analysis of Variance; **b.wt.:** Body weight; **g:** Gram(s); **GnRH:** Gonadotropin-Releasing Hormone; **H.E.:** Hematoxylin and Eosin; **h:** Hr(s); **kg:** Kilogram(s); **mg:** Milligram(s); **P. pinnata:** *Pongamia pinnata*; **SEM:** Standard Error of Mean.

## CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

## SUMMARY

Oral administration of a 50% ethanolic stem bark extract of *Pongamia pinnata* to male Wistar rats for 60 days produced dose-dependent decreases in testicular weight and serum testosterone, accompanied by marked histopathological alterations in seminiferous tubules and Leydig cells, suggesting its potential use as a plant-based male antifertility agent.

## REFERENCES

Abercrombie, M. (1946). Estimation of nuclear population from microtome sections. *Anatomical Record, 94*, 239-247.

Abney, T. O. (1999). Roles of estrogens in Leydig cell development and function. *Steroids, 64*, 610-617.

Akram, M., Nimesh, S., Chishti, M. A., *et al.* (2021). *Pongamia pinnata*: An updated review on phytochemistry and pharmacological uses. *Pharmaceutical and Pharmacology International Journal, 9*(5), 194-199. https://doi.org/10.15406/ppij.2021.09.00344

Anderson, R. A., & Baird, D. T. (2002). Male contraception. *Endocrine Reviews, 23*, 735-762.

Bandivdekar, A. H., & Moodbidri, S. B. (2002). Spermicidal activity of seed oil of *Pongamia glabra*. *Archives of Andrology, 48*(1), 9-13. https://doi.org/10.1080/01485102753385152

Bhardwaj, G. S., Jain, A., Jangid, T., & Jangir, R. N. (2025). Exploring the male antifertility potential of medicinal plants: A comprehensive review. *Pharmacognosy Research, 17*(1), 265-275.

Bhargava, S. K. (1989). Antiandrogenic effects of a flavonoid-rich fraction of *Vitex negundo* seeds. *Journal of Ethnopharmacology, 27*, 327-333.

Boockfor, F. R., & Blake, C. A. (1997). Chronic exposure to 4-tert-octylphenol disrupts spermatogenesis in adult male rats. *Biology of Reproduction, 57*, 267-277.

Creasy, D. M. (2001). Pathogenesis of male reproductive toxicity. *Toxicologic Pathology, 29*, 64-76.

Das, S., Parveen, S., Kundra, C. P., & Pereira, B. M. J. (2004). Male rat reproduction is vulnerable to flavonoid-rich seed extracts of *Vitex negundo*. *Phytotherapy Research, 18*, 8-13.

Dias, T. R., Alves, M. G., Oliveira, P. F., & Silva, B. M. (2014). Natural products as modulators of spermatogenesis: The search for a male contraceptive. *Current Molecular Pharmacology, 7*(2), 154-166. https://doi.org/10.2174/1874467208666150126155912

Farnsworth, N. R., Bingel, A. S., Cordell, G. A., Crane, F. A., & Fong, H. H. S. (1975). Plants as sources of new antifertility agents. *Journal of Pharmaceutical Sciences, 64*, 535-598.

Gilani, A. H., & Rahman, A. U. (2005). Trends in ethnopharmacology. *Journal of Ethnopharmacology, 100*, 43-49.

Hoesl, C. E., Saad, F., Poppel, M., & Altwein, J. E. (2005). Reversible non-barrier male contraception: Status and prospects. *European Urology, 48*, 712-723.

Jain, G. C., & Ali, S. M. (2007). Effect of ethanolic extract of *Cassia alata* (L.) flowers on reproductive function of male albino rats. *Journal of Experimental Zoology India, 10*, 129-132.

Jana, K., Jana, S., & Samanta, P. K. (2006). Chronic exposure to sodium arsenate alters hypothalamic-pituitary-testicular activity in adult rats. *Reproductive Biology and Endocrinology, 4*, 9. https://doi.org/10.1186/1477-7829-4-9

Kim, J. M., Ghosh, S. R., Wells, A. C. P., & Zirkin, B. R. (2001). Caspase-3 and caspase-activated DNase are associated with germ cell apoptosis. *Endocrinology, 142*, 3809-3816.

Kirtikar, K. R., & Basu, B. D. (1933). *Indian medicinal plants* (Vol. 1, p. 2226). Allahabad, India: Lalit Mohan Basu.

Kitagawa, I., Zhang, R., Hori, K., Tsuchiya, N., & Shibuya, H. (1992). Pongapinones A and B from *Pongamia pinnata* bark. *Chemical and Pharmaceutical Bulletin, 40*, 2041-2043.

Leblond, C. P., & Clermont, Y. (1952). Spermatogenesis of rat, mouse, hamster and guinea pig as revealed by the periodic acid-fuchsin sulfurous acid technique. *American Journal of Anatomy, 90*, 167-215.

Li, L., Li, X., Shi, C., Deng, Z., Fu, H., Proksch, P., & Lin, W. (2006). Pongamone A-E, five flavonoids from *Pongamia pinnata*. *Phytochemistry, 67*, 1347-1352.

Meerwal, P., & Jain, G. C. (2016). Antifertility effect of *Caesalpinia bonducella* (L.) Fleming in male Wistar rats. *International Journal of Pharmacognosy, 3*(6), 265-275.

Muqarrabun, L. M. R., Ahmat, N., Ruzaina, S. A. S., Ismail, N. H., & Sahidin, I. (2013). Medicinal uses, phytochemistry and pharmacology of *Pongamia pinnata* (L.) Pierre: A review. *Journal of Ethnopharmacology, 150*(2), 395-420. https://doi.org/10.1016/j.jep.2013.08.041

Nandi, S., Banerjee, P. P., & Zirkin, B. R. (1999). Germ cell apoptosis in the testis of Sprague-Dawley rats following testosterone withdrawal. *Biology of Reproduction, 61*, 70-75.

Nusier, M. K., Bataineh, H. N., & Daradka, H. M. (2007). Adverse effects of rosemary on reproductive function in adult male rats. *Experimental Biology and Medicine, 232*, 809-813.

O'Donnell, L., *et al.* (2006). Endocrine regulation of spermatogenesis. In J. D. Neill (Ed.),*Knobil and Neill's physiology of reproduction* (pp. 1017-1069). Elsevier Academic Press.

Ogbuewu, I. P., Unamba-Oparah, I. C., Odoemenam, V. U., Etuk, I. F., & Okoli, I. C. (2011). Potentiality of medicinal plants as sources of new contraceptive principles in males. *North American Journal of Medical Sciences, 3*(6), 255-263. https://doi.org/10.4297/najms.2011.3250

Page, S. T., Amory, J. K., & Bremner, W. J. (2008). Advances in male contraception. *Endocrine Reviews, 29*, 465-493. https://doi.org/10.1210/er.2007-0041

Purohit, A., & Bhagat, M. (2005). Contraceptive effect of *Curcuma longa* (L.) in male albino rats. *Asian Journal of Andrology, 6*, 71-74.

Rates, S. M. K. (2001). Plants as source of drugs. *Toxicon, 39*, 603-613.

Sharpe, R. M., Donachie, K., & Cooper, I. (1988). Re-evaluation of the intratesticular level of testosterone required for maintenance of spermatogenesis in the rat. *Journal of Endocrinology, 117*, 19-26.

Tulsiani, D. R., & Abou-Haila, A. (2008). Male contraception: An overview of potential target events. *Endocrine, Metabolic & Immune Disorders - Drug Targets, 8*, 122-131.

Unny, R., Chauhan, A. K., *et al.* (2003). Potentiality of medicinal plants as a source of new contraceptive principles. *Phytomedicine, 10*, 233-260.

Vidhale, D. K., Mora, V. R., Pasam, S., Kotha, A., Konda, U., & Boggula, N. (2024). Critical review on *Pongamia pinnata*: A versatile medicinal plant. *Asian Journal of Pharmaceutical Research and Development, 12*(4), 138-146. https://doi.org/10.22270/ajprd.v12i4.1455

Yakubu, M. T., Oladiji, A. T., & Akanji, M. A. (2007). Biochemical indices and testicular histology following chronic administration of *Fadogia agrestis* extract. *African Journal of Biochemistry Research, 1*, 156-163.

Yang, J. Y., Wang, G. X., Liu, J. L., Fan, J. J., & Cui, S. (2007). Toxic effects of zearalenone and α-zearalenol on the male reproductive system in mice. *Reproductive Toxicology, 24*, 381-387.

Yang, Z. W., Kong, L. S., Guo, Y., Yin, J. Q., & Mills, N. (2006). Histological changes of the testis and epididymis following Leydig cell destruction. *Asian Journal of Andrology, 8*, 289-299.

Yin, H., Zhang, S., & Wu, J. (2004). Study on flavonoids from stem bark of *Pongamia pinnata*. *Zhong Yao Cai, 27*, 493-495.

**Cite this article:** Jain K, Joshi SC. *Pongamia pinnata* Stem Bark Extract Induced Changes in Testicular Function in Male Wistar Rats. Pharmacog Res. 2026;18(4):1379-85.