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**Title: Studying the effect of Long pepper plant extract on  
spermatogenesis in busulfan-induced azoospermia rat model based  
on histopathological criteria and sperm analysis**

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## **Abstract**

### **Background:**

Male-factor infertility accounts for approximately 60% of infertility cases, with azoospermia being one of the major causes. Despite advances in medical treatments, therapeutic options remain limited. Medicinal plants with antioxidant and regenerative properties are increasingly explored as potential alternatives. This study evaluated the spermatogenic effect of long pepper plant extract in busulfan-induced azoospermia rats' model with using histopathological criteria.

### **Methods:**

Twenty-one adult male rats were assigned to three groups: healthy control (n=7), azoospermic control (n=7), and treatment (n=7; gavages with Long pepper plant extract at a 5mg/cc dose for 48 days). Azoospermia was induced using busulfan (30 mg/kg). At the end of treatment, testes and epididymis's were harvested for histopathological and morphometric evaluations of spermatogenesis and seminiferous tubule characteristics.

### **Results:**

In control group, seminiferous tubules exhibited degenerative changes and absence of mature spermatozoa. In contrast, the Long pepper-treated group showed significant recovery in seminiferous tubule architecture, presence of mature spermatozoa within the lumen, and partial restoration of sperm parameters compared with azoospermic groups ( $p<0.05$ ). Body weight gain and testicular size improved, approaching values observed in healthy controls.

### **Conclusion:**

Long pepper plant extract demonstrated promising spermatogenic and histoprotective effects in azoospermic rats, suggesting its potential as an adjunctive therapeutic agent for male infertility related to impaired



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spermatogenesis. Further studies are warranted to elucidate its molecular mechanisms and clinical applicability.

**Keywords:** Azoospermia, Long pepper, Spermatogenesis, Histopathology.

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