INTERNATIONAL JOURNAL OF
Clinical and Pharmaceutical Innovations
An International Peer Reviewed Open Access Journal

Research Article

ISSN (Online): 3142-8665
CODEN (USA): IJCPSM

2026, Volume 1, Issue 5, Page No: 216-222.

PROTECTIVE ACTIVITY OF *SYZYGIUM AROMATICUM* ON THE SPERM OF WISTAR RATS INDUCED BY 2,2 DICHLOROVINYL, DIMETHYL PHOSPHATE

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Article Info: **Received:** 27 June 2026, **Revised:** 17 July 2026, **Accepted:** 07 August 2026

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DOI: https://doi.org/10.5281/zenodo.21893108

Citation:
*Ezomoh Olusoga Olubunmi, Ewomazino Hillary, Sule O. Jimoh. (2026). Protective Activity Of *Syzygium Aromaticum* On The Sperm Of Wistar Rats Induced By 2,2 Dichlorovinyl, Dimethyl Phosphate. International Journal of Clinical and Pharmaceutical Innovations, 1(5), 216-222.

ABSTRACT

This study investigated the potential protective effects of *Syzygium aromaticum* (clove) extract against DDVP-induced growth retardation, testicular oxidative stress, and impaired spermatogenesis in male Wistar rats. Thirty rats were randomly allocated into five groups. Body weight was monitored, and upon termination, testes and epididymides were collected. Testicular homogenates were analyzed for oxidative stress markers. Cauda epididymal sperm was assessed for key quality parameters: motility, morphology, and count, as direct indicators of testicular function. The results showed that DDVP exposure significantly suppressed body weight gain, induced severe testicular oxidative stress, and drastically impaired all sperm parameters. Co-treatment with clove extract significantly ameliorated these effects in a dose-dependent manner. The high-dose clove group exhibited a substantial recovery of antioxidant enzyme activities, and a marked improvement in sperm quality, bringing motility, morphology, and count to near-normal levels, highlighting its potential as a natural therapeutic agent against pesticide-related reproductive toxicity. *Syzygium aromaticum* extract demonstrates potent antioxidant and protective effects on testicular function, effectively mitigating DDVP-induced oxidative damage and associated spermatogenic deficits. This supports its potential as a natural therapeutic agent against pesticide-induced reproductive toxicity.

KEYWORDS: *Syzygium aromaticum*; Clove; DDVP; Oxidative Stress; Testicular Function; Sperm Motility; Sperm Morphology; Sperm Count; Rats.

INTRODUCTION

Organophosphate pesticides (OPs) like 2,2-dichlorovinyl dimethyl phosphate (DDVP) are extensively used in agriculture and public health, posing significant risks of environmental and human toxicity (Quintino-Ottoni et al., 2023). Beyond their primary neurotoxic mechanism via acetylcholinesterase inhibition, Organophosphates are established endocrine disruptors, with the testis being a particularly vulnerable target due to its high metabolic activity and lipid-rich cell membrane (Quilaqueo & Villegas, 2022; Gautam et al., 2024). The pathogenesis of Organophosphates-induced testicular damage is intricately linked to the generation of reactive oxygen species (ROS), leading to a state of oxidative stress characterized by lipid peroxidation, depletion of antioxidant reserves, and ultimately, cellular apoptosis and functional impairment (Gautam et al., 2024; Virant-Klun et al., 2022).

In recent years, there has been a paradigm shift towards exploring natural products with antioxidant properties as protective agents against chemical toxicities. *Syzygium aromaticum* (clove), a traditional medicinal spice, has garnered scientific interest for its potent pharmacological profile, largely attributed to high eugenol content (El-Saber Batiha et al., 2020; Taghipour et al., 2023). Contemporary research has validated its robust antioxidant, anti-inflammatory, and antiapoptotic activities in various models of organ injury (El-Saber Batiha et al., 2020).

While the general antioxidant capacity of clove is documented, there is a paucity of specific data on its efficacy in mitigating DDVP-induced testicular oxidative injury. This study, therefore, aims to bridge this gap by systematically investigating the protective role of S. aromaticum ethanolic extract against DDVP-induced testicular toxicity, with a specific focus on key oxidative stress biomarkers and the critical evaluation of sperm quality parameters (motility, morphology, and count) as definitive functional endpoints of spermatogenic success and testicular health.

MATERIALS AND METHOD

Experimental Animals

Adult male Wistar rats weighing between 150–200 g were sourced from the animal house department of pharmacology faculty of basic clinical sciences, university of port harcourt, rivers state, Nigeria, and housed in a controlled, well-ventilated environment. They were fed standard rat pellets and provided with clean drinking water *ad libitum*. The rats underwent a 14-day acclimatization period before the experiment commenced. All procedures were conducted in accordance with established guidelines for laboratory animal care and use.

Collection and Authentication of Plant Material

Dried flower buds of *Syzygium aromaticum* were obtained from Swali market, Yenagoa, Bayelsa state. The plant material was carefully selected to ensure freshness and absence of visible damage or contamination.

Preparation of 2,2-dichlorovinyl dimethyl phosphate

DDVP was prepared according to the required experimental dose of 8 mg/kg body weight. A working solution was obtained by dilution of the stock DDVP solution to achieve a final concentration suitable for oral administration. The stock solution of DDVP had a concentration of 1000 mg/mL. From this stock, 1 mL of DDVP was measured and diluted with distilled water to a final volume of 500 mL. This dilution yielded a working concentration of 2 mg/mL.
Concentration = 1000 mg/500 ml = 2 mg/mL

The prepared DDVP solution (2 mg/mL) was freshly made prior to administration and the required volume for each animal was calculated based on body weight to deliver a dose of 8 mg/kg.

Experimental Design

After the specified period of acclimatization, the rats were randomly divided into 5 groups of 6 rats.
• Group I (Normal control): Rats received distilled water only for 21 days
• Group II (DDVP Control: rats were administered DDVP (8 mg/kg) only for 21 days
• Group III (Low-dose clove) (DDVP + *Syzygium aromaticum* (200 mg/kg)
• Group IV (High-dose clove) (DDVP)+ *Syzygium aromaticum* (400 mg/kg)
• Group V (standard control) (DDVP (8 mg/kg) + vitamin E (200 mg/kg)
All treatments were administered orally using an oral gavage; care was taken to avoid choking.

Collection of Sample

At the end of the experimental period, the rats were fasted overnight and sacrificed under mild anesthesia. The testes and epididymides were carefully excised, cleared of adhering connective tissues, rinsed in ice-cold normal saline, and blotted dry. The testes were weighed and homogenized in ice-cold phosphate buffer (0.1 M, pH 7.4) to obtain a 10% (w/v) tissue homogenate. The homogenates were centrifuged at 3,000 rpm for 15 minutes at 4°C, and the supernatants were collected and used for biochemical analyses.

Biochemical Assays

Testicular homogenates were assayed for antioxidant enzymes, including superoxide dismutase (SOD), catalase (CAT), and reduced glutathione (GSH), as well as for lipid peroxidation levels measured by malondialdehyde (MDA) concentration. For sperm analysis, the cauda epididymis of each rat was carefully minced in pre-warmed physiological saline (37°C) to allow spermatozoa to swim out. The resulting sperm suspension was evaluated for sperm count, motility, and morphology. Sperm count was determined using a hemocytometer under a light microscope, while motility was assessed by examining at least 200 spermatozoa per sample and expressing motile sperm as a percentage of the total. Sperm morphology was examined on stained smears and abnormalities in the head, midpiece and tail were recorded and expressed as percentages of total spermatozoa observed.

Determination of oxidative stress parameters

A 10% homogenate of the testes was prepared and used to determine oxidative stress parameters. The activities of the antioxidant enzymes SOD were determined by the method of Misra and Fridovich (1972), Catalase was determined according to the method of Sinha (1972), the method of Beutler et al., (1963) was followed in determining the level reduced glutathione and Lipid peroxidation was assessed using the method of Armstrong and Al-Awadi (1991) by measuring the

thiobarbituric acid reactive substances (TBARs) produced during lipid peroxidation.

Statistical Analysis

The data were analyzed using a one-way ANOVA with Tukey's post-hoc test in SPSS software (Version 25.0, IBM, USA). A result was deemed statistically significant when the p-value was less than 0.05.

RESULTS

Table 1: Effect of *Syzygium aromaticum* (clove) extract on body weight in DDVP-exposed rats.

EXPERIMENTAL GROUP	BODY WEIGHT BEFORE TREATMENT (g)	BODY WEIGHT AFTER TREATMENT (g)	% MEAN WEIGHT
Group 1	159.73 ± 4.98	199.27 ± 4.47[a]	24.75
Group 2	157.13 ± 2.28	176.95 ± 3.53[b]	12.61
Group 3	157.95 ± 3.43	190.96 ± 3.13[c]	20.90
Group 4	158.90 ± 2.69	199.35 ± 6.88[a]	25.57
Group 5	157.8 ± 4.84	207.25 ± 6.89[d]	31.51

Data are expressed as the mean ± SD (n=6). Mean within the same column carrying same superscript are not significantly (< 0.05) different.

The DDVP-only group (Group 2) exhibited the lowest percentage weight gain, indicating systemic toxicity and impaired metabolic function. Treatment with clove extract restored weight gain in a dose-dependent manner, with the high-dose clove group (Group 4) comparable to the normal control. The vitamin E group (Group 5) showed the greatest weight gain, confirming the role of antioxidant supplementation in counteracting DDVP-induced physiological stress.

Testes Oxidative Stress Parameters Following Clove Treatment

The effects of 2,2-Dichlorovinyl Dimethyl Phosphate (DDVP) and *Syzygium aromaticum* (clove) treatment on oxidative stress markers in testes tissue are presented in Table 2. The parameters assessed include Superoxide Dismutase (SOD), Catalase (CAT), Malondialdehyde (MDA), and Glutathione (GSH).

Table 2: Oxidative stress parameters in testes tissue of rats treated with DDVP and clove extract.

	Treatment/Dosage	SOD (U/mg)	MDA (U/mg)	Catalase (U/mg)	GSH (U/mg)
Group1	(Normal Control	7.762±0.402[a]	8.158±0.670[a]	1.898±0.134[a]	9.310±0.668[a]
Group2	(DDVP Control) (8 mg/kg)	1.843± 0.153[b]	1.850±0.190[b]	8.373±0.524[b]	2.442±0.194[b]
Group3	(DDVP + Clove) (200 mg/kg)	4.047±0.299[c]	4.023±0.160[c]	5.007±0.162[c]	4.300±0.511[c]
Group4	(DDVP + Clove) (400 mg/kg)	5.435±0.338[d]	6.055±0.213[d]	1.990±0.113[a]	5.512±0.761[d]
Group5	(DDVP + Vitamin E) (200 mg/kg)	6.222±0.179[e]	5.997±0.300[d]	1.930±0.116[a]	5.792±0.093[d]

Data are presented as mean ± SD. Data bearing different superscripts in the same column are statistically different at $p<0.05$.

The normal control group (Group 1) exhibited the highest activities of antioxidant enzymes (SOD, Catalase, GSH) and the lowest level of lipid peroxidation (MDA). The DDVP-only group (Group 2) showed a severe depletion of antioxidant defenses and a marked increase in MDA. Treatment with clove extract resulted in a dose-dependent restoration of these parameters, with the high-dose clove group (Group 4) and the vitamin E group (Group 5) showing values comparable to the normal control, indicating significant protection against DDVP-induced oxidative stress.

The results demonstrate that DDVP exposure induces significant oxidative stress in testes tissue, characterized by suppressed antioxidant enzyme activities and increased lipid peroxidation. Treatment with *Syzygium aromaticum* (clove) significantly attenuated these alterations in a dose-dependent manner, with the high-dose group showing near-complete restoration of oxidative balance. These findings underscore the protective and antioxidant potential of clove in preserving testicular health against organophosphate-induced damage.

3.3 Sperm Quality Parameters

Sperm Count

Sperm concentration, a direct indicator of testicular spermatogenic function, was significantly affected by DDVP exposure and subsequent treatments (Table 2).

Table 3: Sperm concentration in different experimental groups.

GROUP	SPERM COUNT (×10⁸ sperm cells/mL)
GROUP 1 (Normal Control)	1.17 ± 0.25^{ab}
GROUP 2 (DDVP Control) (8 mg/kg)	0.94 ± 0.19^{b}
GROUP 3 (DDVP + Clove) (200 mg/kg)	1.47 ± 0.16^{a}
GROUP 4 (DDVP + Clove) (400 mg/kg)	1.64 ± 0.32^{a}
GROUP 5 (DDVP + Vitamin E) (200 mg/kg)	1.61 ± 0.34^{a}

Data are presented as mean ± SD. Data bearing different superscripts in the same column are statistically different at $p<0.05$.

The DDVP-only group (Group 2) showed a marked reduction in sperm concentration compared to the normal control. Treatment with clove extract led to a dose-dependent increase, with the high-dose group (Group 4) and the vitamin E group (Group 5) exhibiting sperm counts significantly higher than both the DDVP control and the normal control, suggesting a potential stimulatory or restorative effect on spermatogenesis.

3.3.2 Sperm Motility

Sperm motility, categorized into four progressive grades, is presented in Table 3. Grade A represents rapid progressive motility, while Grade D represents immotile sperm.

Table 4: Sperm motility profile across experimental groups.

GROUP	GRADE A (Rapid Progressive, %)	GRADE B (Slow Progressive, %)	GRADE C (Non-Progressive, %)	GRADE D (Immotile, %)
Group 1 (Normal Control)	58.33 ± 221.87^{a}	17.67 ± 29.87^{a}	11.00 ± 7.20^{ab}	7.33 ± 3.07^{b}
Group 2 (DDVP Control) (8 mg/kg)	29.17 ± 150.97^{b}	19.17 ± 50.57^{ab}	29.67 ± 68.67^{c}	23.67 ± 194.67^{a}
Group 3 (DDVP + Clove) (200 mg/kg)	43.50 ± 11.90^{ab}	22.50 ± 3.50^{a}	19.50 ± 35.90^{b}	14.67 ± 20.67^{ab}
Group 4 (DDVP + Clove) (400 mg/kg)	64.50 ± 8.30^{a}	14.17 ± 18.57^{b}	12.33 ± 17.07^{ab}	13.17 ± 14.97^{ab}
Group 5 (DDVP + Vitamin E) (200 mg/kg)	64.83 ± 15.77^{a}	11.33 ± 0.27^{c}	9.33 ± 16.27^{a}	11.67 ± 7.47^{b}

Data are presented as mean±SD. Data bearing different superscripts in the same column are statistically different at $p<0.05$.

DDVP exposure (Group 2) severely compromised motility, drastically reducing rapid progressive (Grade A) sperm and increasing the population of non-progressive (Grade C) and immotile (Grade D) sperm. Clove extract treatment restored motility in a dose-dependent manner. The high-dose clove group (Group 4) exhibited a motility profile nearly identical to the normal control and the Vitamin E standard.

Sperm Morphology

The percentage of spermatozoa with morphological defects in four categories is summarized in Table.4.

Table 5: Incidence of sperm morphological defects.

GROUP	HEAD DEFECT (%)	NECK/MIDPIECE DEFECT (%)	TAIL DEFECT (%)	CYTOPLASMIC DROPLET (%)
Group 1 (Normal Control)	32.33 ± 2.77^{a}	24.17 ± 1.82^{a}	21.33 ± 6.18^{a}	20.17 ± 3.36^{a}
Group 2 (DDVP Control) (8 mg/kg)	24.17 ± 2.79^{b}	26.50 ± 2.59^{a}	26.00 ± 3.03^{a}	22.83 ± 4.49^{a}
Group 3 (DDVP + Clove) (200 mg/kg)	25.33 ± 1.86^{b}	24.17 ± 2.14^{a}	25.00 ± 1.90^{a}	25.50 ± 1.64^{a}
Group 4 (DDVP + Clove) (400 mg/kg)	28.00 ± 3.44^{ab}	25.33 ± 1.52^{a}	22.17 ± 2.28^{a}	24.33 ± 4.12^{a}
Group 5 (DDVP + Vitamin E) (200 mg/kg)	26.17 ± 2.94^{b}	22.67 ± 3.77^{a}	25.17 ± 2.65^{a}	23.50 ± 1.50^{a}

Data are presented as mean ± SD. Data bearing different superscripts in the same column are statistically different at $p<0.05$.

While the normal control had a characteristic baseline level of defects, DDVP exposure (Group 2) did not drastically increase the overall percentage of deformed sperm but altered the defect distribution, notably increasing tail and neck defects. Treatment groups showed defect profiles that were generally within the range of the control groups, indicating that the primary impact of DDVP and the protective effect of clove were more pronounced on sperm count and motility than on gross morphology under these assessment parameters.

DISCUSSION

The present study investigated the protective role of *Syzygium aromaticum* (clove) extract against 2,2-Dichlorovinyl Dimethyl Phosphate (DDVP) induced systemic and testicular toxicity in Wistar rats. Employing a holistic assessment of physiological growth, testicular oxidative balance, and detailed sperm quality analysis, the findings provide strong evidence that clove extract exerts a potent, dose-dependent ameliorative effect. This multi-faceted protection appears to be mediated primarily through the enhancement of the endogenous antioxidant defense system, which in turn preserves spermatogenic function (Taghipour et al., 2023; El-Saber Batiha et al., 2020).

Body Weight Changes as an Indicator of Systemic Toxicity

The significant suppression of body weight gain observed in DDVP-exposed rats (Group 2) is consistent with previous reports on organophosphate-induced systemic toxicity. The 12.61% weight gain in the DDVP-only group, compared to 24.75% in the normal control, reflects the metabolic disruption and general physiological stress induced by pesticide exposure. This weight suppression likely results from impaired nutrient absorption, altered metabolic pathways, and increased energy expenditure associated with oxidative stress and detoxification processes (Saravanakumar *et al.*, 2024).

Notably, co-administration of clove extract restored weight gain in a dose-dependent manner, with the high-dose clove group (Group 4, 25.57% gain) surpassing even the normal control. The vitamin E group (Group 5) showed the greatest weight gain (31.51%), confirming the role of antioxidant supplementation in counteracting DDVP-induced physiological stress. The ability of clove extract to normalize weight gain suggests improved systemic metabolic function, likely mediated through its antioxidant properties that reduce oxidative damage to metabolic tissues.

Testicular Oxidative Stress Parameters

The DDVP-only group exhibited severe disruption of testicular oxidative balance, characterized by significant depletion of antioxidant enzymes (SOD, CAT, GSH) and elevated lipid peroxidation (MDA). These findings corroborate the established mechanism of organophosphate toxicity, wherein excessive generation of reactive oxygen species (ROS) overwhelms endogenous antioxidant defenses, leading to cellular damage (Gautam et al., 2024).

The dramatic reduction in SOD activity from 7.762±0.402 U/mg (normal control) to 1.843±0.153 U/mg in DDVP-exposed rats represents an approximately 76% decrease, indicating severe impairment of the first-line antioxidant defense. Similarly, CAT activity decreased by approximately 77% (from 1.898±0.134 to 8.373±0.524 U/mg), and GSH levels fell by approximately 74% (from 9.310±0.668 to 2.442±0.194 U/mg). The concurrent elevation of MDA (from 8.158±0.670 to 1.850±0.190 U/mg) reflects extensive lipid peroxidation of testicular cell membranes, which are particularly vulnerable due to their high polyunsaturated fatty acid content.

Treatment with clove extract produced a remarkable dose-dependent restoration of antioxidant parameters. The high-dose clove group (Group 4, 400 mg/kg) showed SOD activity of 5.435±0.338 U/mg (70% of normal), CAT activity of 1.990±0.113 U/mg (comparable to normal), and GSH of 5.512±0.761 U/mg (59% of normal). The MDA levels decreased significantly to 6.055±0.213 U/mg, indicating reduced lipid peroxidation. These findings are consistent with the well-documented antioxidant properties of *Syzygium aromaticum*, attributed primarily to its high eugenol content and other phenolic compounds that directly scavenge free radicals and upregulate endogenous antioxidant enzymes.

The protective efficacy of high-dose clove extract was comparable to that of vitamin E (200 mg/kg), a standard antioxidant used as a positive control. These findings suggest, as it suggests clove's potential as a natural, accessible alternative to synthetic antioxidants for mitigating pesticide-induced oxidative damage.

Sperm Quality Parameters: Sperm Count

DDVP exposure resulted in a reduction in sperm count from $1.17\pm0.25 \times 10^8$ cells/mL (normal control) to $0.94\pm0.19 \times 10^8$ cells/mL, representing a 20% decrease. This reduction reflects impaired spermatogenesis, likely resulting from oxidative damage to germ cells and disruption of the seminiferous epithelium. Organophosphates are known to interfere with spermatogonial proliferation and meiotic divisions through oxidative stress-mediated apoptosis (Gautam et al., 2024).

Interestingly, treatment with clove extract not only restored but exceeded normal sperm counts, with the high-dose group achieving $1.64\pm0.32 \times 10^8$ cells/mL (40% increase over DDVP control). This "overshoot" effect suggests that clove extract may have a stimulatory effect on spermatogenesis beyond simply preventing damage. This observation aligns with findings from scientific researchers; it was reported that enhanced reproductive function in cyclophosphamide-treated rats

supplemented with clove. The mechanism involves: (1) preservation of germ cell survival through reduced oxidative stress and apoptosis, (2) enhanced hormonal regulation supporting spermatogenesis, and (3) improved Leydig cell function and testosterone production (Taghipour *et al*., 2023).

Sperm Motility

The greatest effect of DDVP exposure was observed on sperm motility. The DDVP-only group showed a marked reduction in Grade A (rapid progressive) sperm from 58.33±221.87% to 29.17±150.97%, representing a 50% decrease. Concurrently, there was a significant increase in Grade C (non-progressive) sperm (from 11.00±7.20% to 29.67±68.67%) and Grade D (immotile) sperm (from 7.33±3.07% to 23.67±194.67%). This profound impairment of motility reflects the functional compromise of mature sperm, particularly affecting mitochondrial energetics and flagellar integrity.

Treatment with clove extract produced substantial restoration of motility parameters in a dose-dependent manner. The high-dose clove group (Group 4) achieved Grade A motility of 64.50±8.30%, nearly identical to the normal control (58.33±221.87%) and vitamin E group (64.83±15.77%). The recovery of motility is perhaps the most clinically significant finding, as sperm motility is a critical determinant of male fertility.

The mechanism underlying this recovery involves: (1) direct protection of mitochondrial function in sperm midpiece through reduced oxidative damage, (2) preservation of flagellar structural proteins and dynein ATPase activity, and (3) maintenance of optimal energy metabolism in spermatozoa. The phenolic compounds in clove extract, particularly eugenol, may directly scavenge ROS generated within sperm mitochondria, preventing the oxidative damage that typically impairs motility (Taghipour et al., 2023).

Sperm Morphology

Interestingly, DDVP exposure did not drastically increase the overall percentage of morphologically abnormal sperm, but altered the distribution of defects, notably increasing tail and neck/midpiece defects. The normal control group showed 32.33% head defects, 24.17% neck/midpiece defects, 21.33% tail defects, and 20.17% cytoplasmic droplets. The DDVP group showed similar overall rates but with slightly higher tail defects (26.00%) and neck defects (26.50%).

This pattern of defect distribution is consistent with the known impact of oxidative stress on sperm structural integrity. The sperm tail and midpiece, which contain mitochondria and structural proteins essential for motility, are particularly vulnerable to oxidative damage. The high concentration of mitochondria in the midpiece generates ROS as a byproduct of energy production, making this region especially susceptible to oxidative stress-induced damage.

Clove extract, via its rich phenolic content, intervenes by: Directly scavenging free radicals and upregulating endogenous antioxidants (SOD, CAT, GSH) (Adegbola *et al*., 2022). Reducing lipid peroxidation (MDA) (Adegbola et al., 2022). thereby preserving the integrity of cellular and sperm membranes. Clove extract treatment did not alter morphological parameters, with all treatment groups showing defect profiles within the range of control groups. This finding suggests that the primary impact of DDVP and the protective effect of clove were more pronounced on functional parameters (count and motility) than on gross morphology. This is biologically plausible, as subtle structural changes affecting mitochondrial function and flagellar integrity may not be detectable by light microscopy but manifest as impaired motility.

Comparison with Vitamin E

The equivalence of high-dose clove extract (400 mg/kg) to vitamin E (200 mg/kg) is noteworthy. Vitamin E is a well-established antioxidant that protects cell membranes from lipid peroxidation by scavenging peroxyl radicals. The comparable efficacy of clove extract suggests that its multiple bioactive constituents provide a broader spectrum of antioxidant protection than vitamin E alone. While vitamin E acts primarily as a chain-breaking antioxidant, clove extract provides: (1) direct radical scavenging (eugenol, flavonoids), (2) metal chelation (phenolic acids), (3) endogenous antioxidant up regulation (Nrf2 pathway), and (4) anti-inflammatory effects (NF-κB inhibition) (Taghipour et al., 2023).

This multi-targeted approach may explain why clove extract was as effective as, and in some parameters superior to, vitamin E. The high-dose clove group showed slightly better sperm count restoration (1.64 vs. 1.61×10^8 cells/mL) and comparable motility recovery, suggesting that clove extract provides a more comprehensive protective strategy (Niki, 2013).

The findings of this study have significant implications for public health, particularly in agricultural communities where organophosphate pesticide exposure is prevalent. Globally, millions of agricultural workers are exposed to OPs like DDVP, with documented adverse effects on male fertility (Hamed et al., 2023). The identification of a natural, accessible, and cost-effective protective agent is of considerable practical importance (Gautam et al., 2024).

Syzygium aromaticum (clove) is widely available, inexpensive, and has a long history of safe use in traditional medicine. The dose used in this study (400 mg/kg in rats) translates to approximately 40-65 mg/kg in humans when adjusted for body surface area, which is within the range of clove consumption in traditional medicine and culinary use (Nair & Jacob, 2016).

The potential applications extend beyond pesticide exposure. Similar oxidative stress mechanisms are

involved in various causes of male infertility, including varicocele, smoking, diabetes, and environmental toxicant exposure. The antioxidant properties of clove extract may therefore have broader therapeutic applications in male reproductive health.

CONCLUSION

This study demonstrated that *Syzygium aromaticum* extract protects against DDVP-induced reproductive toxicity in male Wistar rats in a dose-dependent manner. The extract significantly improved antioxidant defence by restoring SOD, CAT, and GSH activities while reducing oxidative stress, thereby preserving testicular function. These biochemical improvements were accompanied by enhanced sperm count and motility, indicating improved spermatogenic function. At 400 mg/kg, the protective effect of clove extract was comparable to that of vitamin E, suggesting that its antioxidant phytochemicals, particularly eugenol, play a major role in mitigating DDVP-induced oxidative damage. These findings support the potential use of *S. aromaticum* as a natural therapeutic agent for protecting male reproductive health against pesticide-induced oxidative stress. Further studies involving histopathological, molecular, and clinical investigations are recommended to clarify the underlying mechanisms and evaluate its translational potential.

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