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A Comparative Study Between Effects of Ginger, Propolis and Garlic on Semen Quality in Male Rabbits

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Abstract

The present study investigated the effects of propolis, ginger, and garlic supplementation on semen characteristics in male rabbits. A total of four experimental groups were assessed: a control group and three treatment groups receiving propolis, ginger, or garlic. The results demonstrated a significant improvement ($p < 0.05$) in ejaculate volume, sperm concentration, total sperm output, and motility in the treatment groups compared to the control. Specifically, propolis and ginger significantly enhanced sperm concentration (319 ± 7.2 and 321 ± 6.7 $\times 10^6$ ml^{-1}, respectively) compared to the control (264 ± 4.5 $\times 10^6$ ml^{-1}). Additionally, the total functional sperm fraction (TFSF) was highest in the ginger-treated group (192 ± 9.1 $\times 10^6$), indicating improved sperm viability and quality. Reaction time was significantly reduced in the garlic-treated group (2.49 ± 0.152 s), suggesting enhanced sexual performance. The pH levels varied significantly, with the lowest value observed in the garlic group (7.24 ± 0.042), which may indicate an alteration in seminal fluid composition.

Keywords: Propolis, Ginger, Garlic, Semen quality, Sperm motility, Male rabbits, Fertility

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Keywords: Propolis, Ginger, Garlic, Semen quality, Sperm motility, Male rabbits, Fertility

Introduction

Male reproductive health is a crucial aspect of animal physiology, significantly affecting fertility and breeding efficiency [1]. Several natural compounds have been investigated for their potential to enhance semen quality, particularly in male rabbits, which serve as an important model for reproductive studies. Among these natural substances, ginger (Zingiber officinale), propolis, and garlic (Allium sativum) have been extensively studied due to their bioactive compounds and pharmacological properties. These natural products exhibit antioxidant, anti-inflammatory, and immunomodulatory effects, which can potentially improve sperm motility, concentration, and overall reproductive performance [2]. Similarly, propolis, a resinous substance produced by bees, possesses strong antioxidant and antimicrobial properties that contribute to reproductive health improvement [3]. Several studies have evaluated the impact of ginger, propolis, and garlic on reproductive health in rabbits and rodents. Research by [4] demonstrated that dietary supplementation with ginger significantly improved sperm motility, count, and viability in male rabbits, which was attributed to its antioxidant properties. These findings highlight the potential of ginger, propolis, and garlic in enhancing semen quality and reproductive health [5]. However, comparative studies evaluating their relative effectiveness in male rabbits are limited. This study aims to provide a comparative analysis of the effects of these natural supplements on semen quality in male rabbits, contributing to the growing body of research on natural fertility enhancers [6]. Oxidative stress plays a major role in reducing semen quality by causing lipid peroxidation in sperm membranes, leading to reduced motility and viability. Antioxidant-rich supplements such as ginger, propolis, and garlic can counteract this oxidative stress and improve sperm function. Ginger, for instance, contains bioactive compounds like gingerols and shogaols, which have been reported to enhance testosterone levels and sperm parameters in animal models[7]. Similarly, propolis, a resinous substance produced by bees, possesses strong antioxidant and antimicrobial properties that contribute to reproductive health improvement [8]. Additionally, garlic is well known for its sulfur-containing compounds, including allicin, which exhibit protec-

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tive effects against oxidative damage in sperm cells [9].

Materials and Methods

Experimental Materials Propolis, ginger (Zingiber officinale), and garlic were employed in this study. Garlic oil was obtained from the public market for therapeutic herbs in Al-Bayda city. Ginger was purchased from Superior Nutrition and Formulation by Jarrow Formulas, Los Angeles, USA, while propolis was provided by California Health Products, Inc., 11577 W. Olympic Blvd., Los Angeles, CA 90064. All chemicals used in the experiment were of analytical grade.

Experimental Animals and Management The study utilized adult male New Zealand white rabbits aged six months, with an initial body weight of 1.892 ± 27.8 kg. The rabbits were individually housed in separate cages under controlled environmental conditions and were subjected to weekly weigh-ins throughout the three-month experimental period. Food and water were provided ad libitum.

The rabbits were fed pelleted diets composed of 30% berseem (Trifolium alexandrinum) hay, 25% yellow maize, 26.2% wheat bran, 14% soybean meal, 3% molasses, 1% calcium chloride ($CaCl_2$), 0.4% sodium chloride (NaCl), 0.3% mineral and vitamin premix, and 0.1% methionine. The vitamin and mineral premix per kg of diet contained: vitamin A (4000,000 IU), vitamin D3 (5000,000 IU), vitamin E (16.7 g), vitamin K (0.67 g), vitamin B1 (0.67 g), vitamin B2 (2 g), vitamin B6 (0.67 g), vitamin B12 (0.004 g), vitamin B5 (16.7 g), pantothenic acid (6.67 g), biotin (0.07 g), folic acid (1.67 g), choline chloride (400 g), zinc (23.3 g), manganese (10 g), iron (25 g), copper (1.67 g), iodine (0.25 g), selenium (0.033 g), and magnesium (133.4 g). The proximate chemical composition of the pellets, as determined by Smith and Mayer (1955), included 15.8% crude protein, 11.3% crude fiber, 3.7% ether extract, 7.2% ash, 92.9% organic matter, and 62.4% nitrogen-free extract on a dry matter basis.

Experimental Design and Treatments The rabbits were randomly assigned into four groups. The first group served as the control, while the remaining three groups received the following treatments:

Group 2: Garlic at a dose of 40 mg/kg body weight (BW) per day[10]

Group 3: Propolis administered by gavage at a dose of 50 mg/kg BW per day [11]

Group 4: Ginger administered by gavage at a dose of 100 mg/kg BW per day [12]

All treatments were dissolved in corn oil and administered for 12 consecutive weeks.

Semen Collection and Evaluation Semen samples were collected weekly for the duration of the 12-week experiment, yielding a total of 60 ejaculates per treatment group. Ejaculates were collected using a teaser doe and an artificial vagina. The volume of each ejaculate was measured using a graduated collection tube after removing the gel mass.

Sperm concentration was assessed using an improved Neubauer hemocytometer slide (GmbH + Co., Brandstwiete 4, 2000 Hamburg 11, Germany) with a mild eosin solution. Total sperm production was calculated as the product of semen volume and sperm concentration. The percentage of motile sperm was evaluated visually under a light microscope at 10× magnification. The total number of motile sperm was determined by multiplying the proportion of motile sperm by the total sperm count.

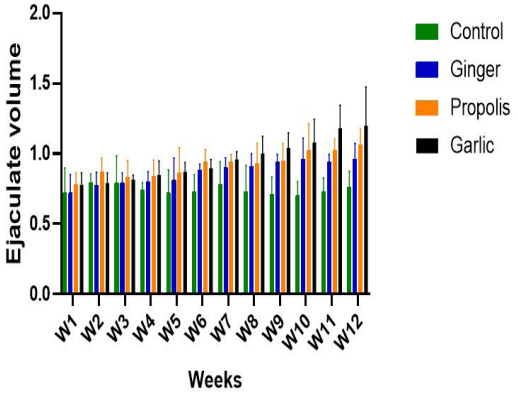
Semen Quality and Biochemical Analysis Reaction time, defined as the time interval from the onset of erection to full erection, was measured in seconds. The initial semen pH was determined immediately after collection using pH indicator paper (Universalindikator pH 0–14, Merck, Merck KGaA, 64271 Darmstadt, Germany). The volume of packed sperm (PSV) was recorded for each sample.

Statistical Analysis Statistical analyses were conducted using GraphPad Prism 8 and Minitab software (version 17). Data normality was assessed before statistical analysis. One-way analysis of variance (ANOVA) followed by Tukey's multiple comparison test was used to evaluate significant differences among groups. A P-value of <0.05 was considered statistically significant.

Results

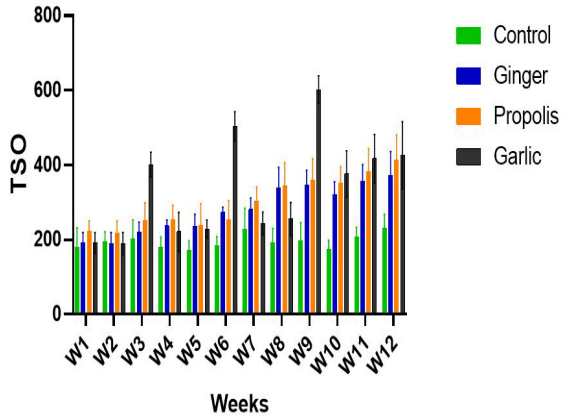
The results indicate that treatment with propolis, ginger, and garlic significantly improved various semen characteristics compared to the control group. Ejaculate Volume & pH: Propolis and ginger significantly increased ejaculate volume compared to the control, while garlic showed a moderate increase. However, pH values were significantly reduced in treated groups, with the lowest pH observed in the garlic group, suggesting a more favorable seminal environment. Reaction Time: All treated groups exhibited a reduced reaction time compared to the control, indicating improved sexual performance. The garlic group showed the shortest reaction time, suggesting a potential enhancement in libido. Sperm Concentration & Motility: Propolis and ginger significantly increased sperm concentration, with both groups showing comparable results, while garlic also enhanced concentration but to a slightly lower extent. Sperm motility was significantly improved in all treated groups compared to the control, with ginger showing the highest percentage. Sperm Viability & Morphology: The percentage of live sperm was significantly increased in all treated groups, with the garlic group showing the highest viability. Conversely, dead sperm percentage decreased, particularly in the garlic-treated group. Normal sperm percentage was significantly higher in treated groups, while abnormal sperm forms were notably reduced. Total Sperm Output (TSO) & Functional Sperm Fraction (TFSF): Propolis, ginger, and garlic treatments significantly increased TSO, with the garlic-treated group showing the highest output. TFSF also improved significantly across all treatments, indicating better overall sperm quality. Seminal Fructose Levels: Fructose concentration was lowest in the propolis-treated group, while ginger and garlic treatments maintained fructose levels closer to the control, suggesting potential differences in energy metabolism.

Figure 1: *Variations in the volume of ejaculate produced by treating*



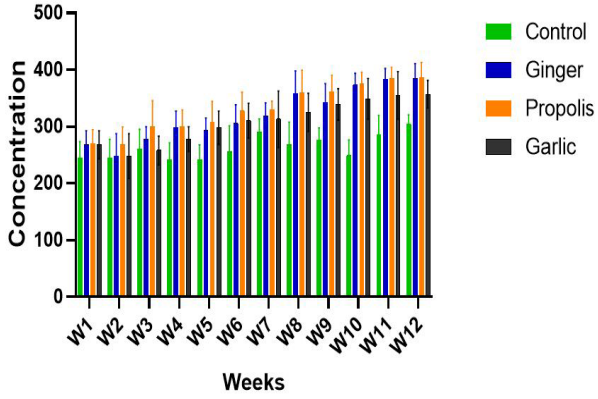
male rabbits with propolis, ginger, and garlic.

Figure 2: *Changes in TSO treatment of male rabbits with garlic , prop-*



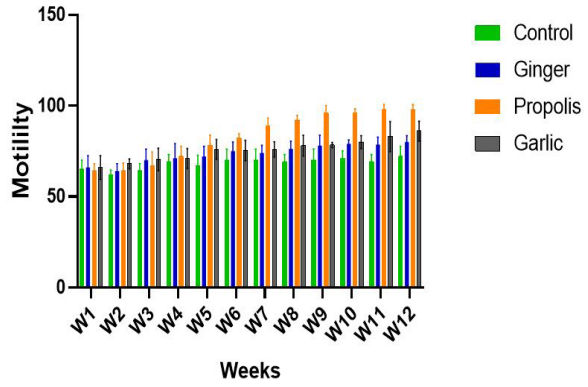
olis and ginger.

Figure 3: *Changes in concentration treatment of male rabbits with*



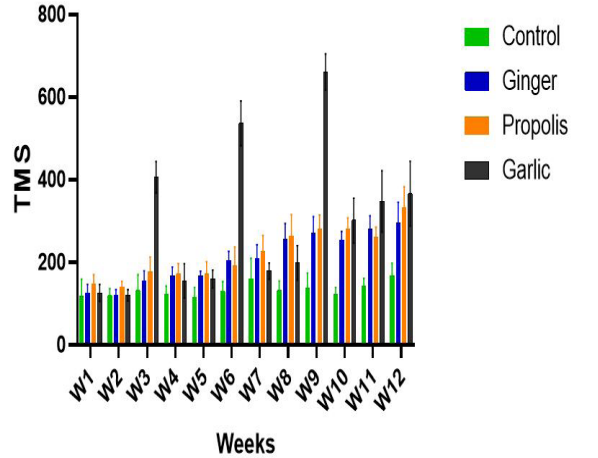
garlic , propolis and ginger.

Figure 4: *Changes in Motility treatment of male rabbits with garlic ,*



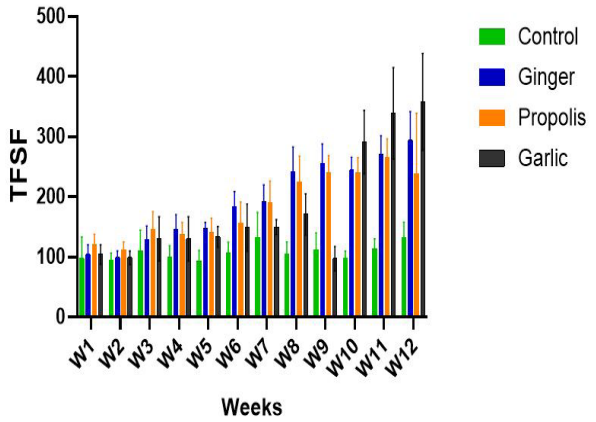
propolis and ginger.

Figure 5: *Changes in TMS treatment of male rabbits with garlic ,*



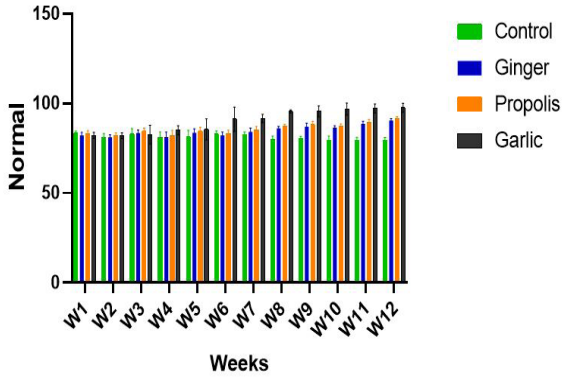
propolis and ginger.

Figure 6: *Changes in TFSF treatment of male rabbits with garlic ,*



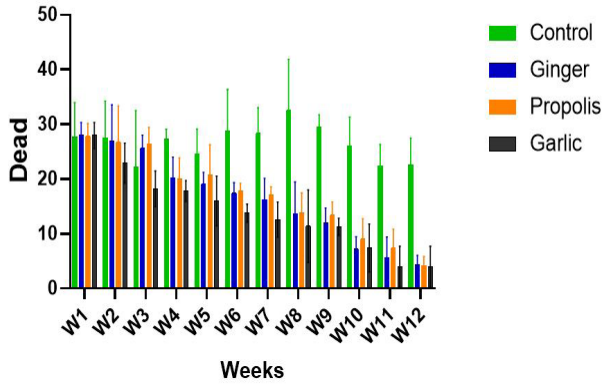
propolis and ginger.

Figure 7: *Changes in Normal treatment of male rabbits with garlic ,*



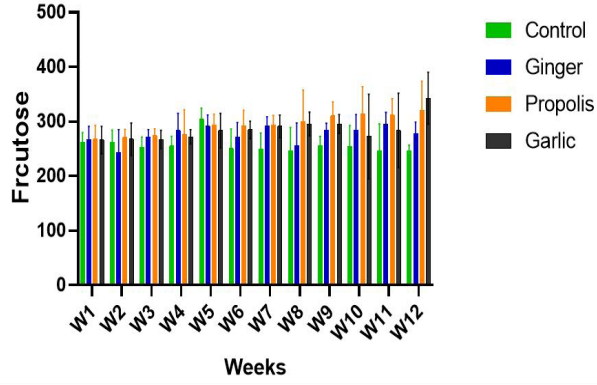
propolis and ginger.

Figure 8: *Changes in Dead treatment of male rabbits with garlic ,*



propolis and ginger.

Figure 9: *Changes in Frcutose treatment of male rabbits with garlic ,*



propolis and ginger.

Figure 10: *Changes in live treatment of male rabbits with garlic , propolis and ginger.*

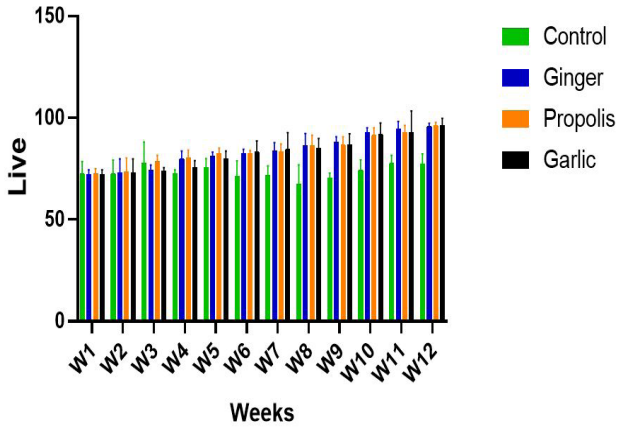


Figure 11: *Changes in Abnormal treatment of male rabbits with garlic , propolis and ginger.*

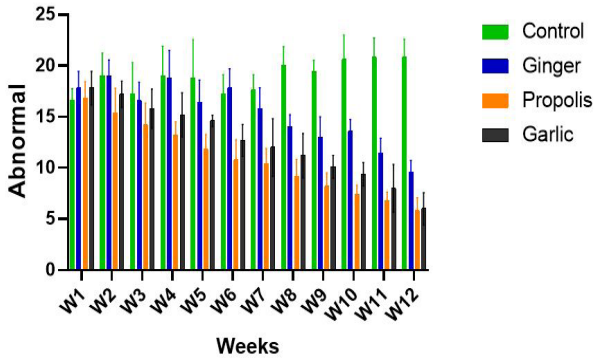


Figure 12: *Changes in PSV treatment of male rabbits with garlic , propolis and ginger.*

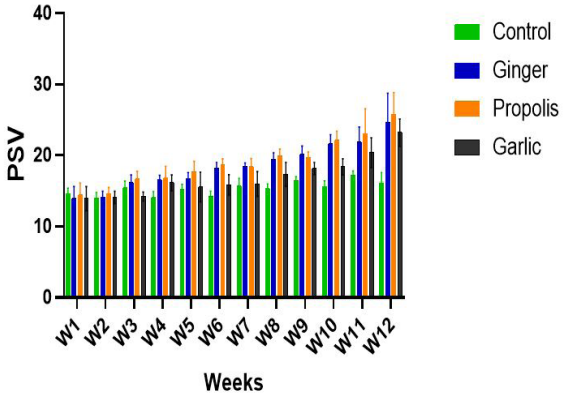


Figure 13: *Changes in PH treatment of male rabbits with garlic , propolis and ginger.*

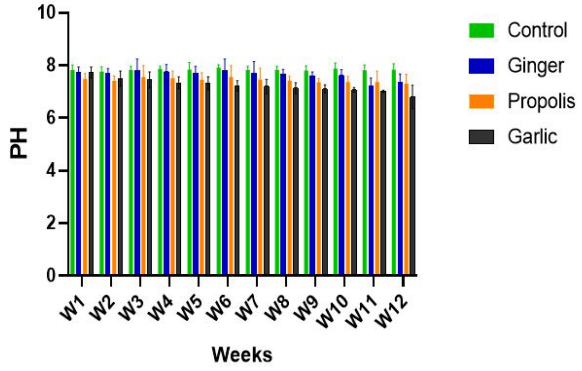
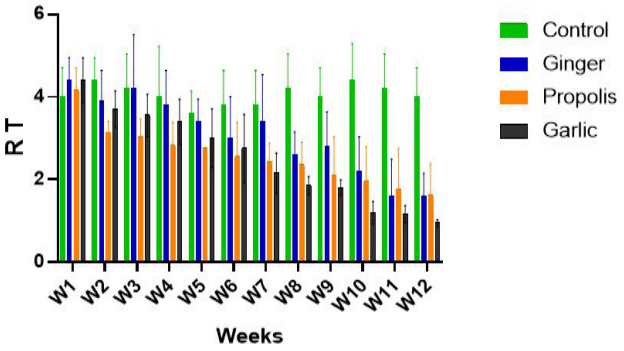


Figure 14: *Changes in RT treatment of male rabbits with garlic , propolis and ginger.*



Discussion

The present study evaluated the effects of dietary supplementation with propolis, ginger, and garlic on semen characteristics in male rabbits. The findings indicate significant improvements in various semen parameters across the treatment groups compared to the control, suggesting the potential of these natural supplements in enhancing male reproductive performance [13]. The increase in ejaculate volume observed in the propolis and ginger groups aligns with previous research indicating that dietary supplementation with these substances can enhance reproductive parameters[14]. For instance, a study by [14] reported that propolis supplementation improved semen quality in rabbit bucks during hot seasons, which could be attributed to its antioxidant properties that mitigate oxidative stress in reproductive tissues. Similarly, ginger has been shown to enhance semen quality, possibly due to its antioxidative effects[3]. The reduction in semen pH in the treated groups may indicate an improved seminal environment, as a slightly acidic to neutral pH is conducive to sperm viability and motility[15]. The decreased reaction time in the garlic-supplemented group suggests an enhancement in sexual desire and performance [16]. This finding is consistent with previous studies that have demonstrated the aphrodisiac properties of garlic, which may be linked to its ability to increase testosterone levels and improve blood circulation [17]. The significant increase in sperm concentration and total sperm output in the propolis and ginger groups corroborates findings from earlier studies [18]. [19] observed that propolis supplementation led to higher sperm concentrations in rabbit bucks, likely due to its role in enhancing testicular function and reducing oxidative damage. Ginger's positive impact on sperm concentration has also been documented, with improvements attributed to its antioxidative and androgenic activities [3]. Enhanced sperm motility and viability in the ginger and garlic groups are in agreement with previous research [20]. Ginger supplementation has been reported to improve sperm motility, possibly through its antioxidant properties that protect spermatozoa from oxidative damage [21]. Garlic's beneficial effects on sperm viability may be due to its antioxidant compounds, which help maintain sperm membrane integrity and function [22]. The improvement in normal sperm morphology observed in the treated groups aligns with findings from previous studies [23]. Propolis has been shown to reduce sperm abnormalities, likely due to its antioxidative effects that protect developing sperm cells from oxidative stress [24]. Similarly, ginger supplementation has been associated with a higher percentage of normal spermatozoa, which may be attributed to its role in enhancing spermatogenesis and protecting sperm from oxidative damage. The increase in the total functional sperm fraction in the ginger and propolis groups suggests an overall enhancement in semen quality[25]. This is consistent with previous findings that have reported improvements in sperm functionality with the supplementation of these natural products, likely due to their combined antioxidative, anti-inflammatory, and androgenic effects[3]. The observed decrease in initial fructose con-

Table 1: The overall means (±SE) of semen characteristics during treatment of male rabbits with propolis, ginger and garlic.
[abc]Within row, means with different superscript letters differ significantly ($p < 0.05$).

Parameters	Animal Groups			
	Control	**Propolis**	**Ginger**	**Garlic**
Ejaculate volume (ml)	0.74±0.017[c]	0.92 ± 0.018[a]	0.831.8[a]	75.7±1.0[a]
PH	7.83±0.022[a]	7.49±0.038[c]	7.64±0.042[b]	7.24±0.042[d]
Reaction time (s)	4.05±0.099[a]	2.98±0.145c[b]	3.08±0.154[b]	2.49±0.152[c]
Packed sperm volume (%)	15.3±0.16[c]	18.11±0.38[a]	18.5±0.45[ab]	16.9±0.39[b]
Sperm concentration ($\times 10^6$ ml^{-1})	264±4..5[b]	319±7.2[a]	321±6.7[a]	308±6.1[a]
Total sperm output ($\times 10^6$)	195±5.1[c]	299±10.3[ab]	280±9.3[b]	338±17.8[a]
Sperm motility (%)	68.2±0.7[b]	71.3±0.9[a]	73.6±0.9[a]	70.2± 0.8[a]
Total motile sperm ($\times 10^6$)	199±5.0[c]	220±8.8[ab]	280±9.3[b]	297±22.3[a]
Live sperm (%)	73.4± 0.8[b]	82.1±1.1[a]	81.7±1.1[a]	82.8±1.2[a]
Dead sperm (%)	26.6±0.82[a]	17.9±1.07[b]	16.3±1.10[b]	14.2±1.01[b]
Normal sperm (%)	81± 0.3[c]	86± 0.4[b]	90± 0.9[cb]	89± 0.89[cb]
Abnormal (%)	19± 0.3[a]	14±0.4[cb]	15± 0.4[b]	13± 0.5[c]
Total functional sperm fraction ($\times 10^6$)	108±3.4[c]	184±8.3[b]	192±9.1[b]	179±12.7[a]
Initial fructose (mg/dl)	257±3.9[b]	200±4.01[a]	266±3.7[a]	285±5.1a[b]

centration in the propolis group may indicate alterations in seminal plasma composition. Fructose serves as an energy source for sperm motility, and its concentration can reflect seminal vesicle function. The reduction in fructose levels could be due to increased utilization by the more active spermatozoa or alterations in seminal vesicle secretion dynamics induced by propolis supplementation [26].

Conclusion

The supplementation of male rabbit diets with propolis, ginger, and garlic has demonstrated significant improvements in semen characteristics, including ejaculate volume, sperm concentration, motility, viability, and morphology. These findings suggest that these natural supplements can be beneficial in enhancing male reproductive performance, potentially through their antioxidative and androgenic properties. Further research is warranted to elucidate the underlying mechanisms and to determine the optimal dosages for these supplements in rabbit diets.

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