Original Paper

Effect of Dietary Supplementation of Proanthocyanidin on Semen Quality, Antioxidant Status, and Hormonal Profile in Ageing Broiler Breeder Male

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Fertility in breeder flocks directly affects the number of hatching eggs available for commercial use. However, in broiler breeder males, semen quality declines significantly with age, especially after 50 weeks. The current study was conducted at the poultry facility of the Department of Animal Production, College of Agriculture, Al-Qasim Green University, from November 1, 2023, to January 31, 2024, spanning 13 weeks. The objective was to evaluate the impact of daily proanthocyanidin supplementation on semen quality characteristics of broiler breeder roosters. A total of twenty roosters, aged 54 weeks, were used in this study. The birds were randomly assigned to four treatment groups, each consisting of five replicates with one rooster per replicate. The experimental treatments were as follows: a control group without supplementation, and three experimental groups receiving 20, 30, and 40 mg/day of proanthocyanidin, respectively. The findings demonstrated that proanthocyanidin supplementation positively influenced the semen quality of roosters, particularly at higher doses. Improvements were most evident during the later stages of the experiment, suggesting that continuous supplementation has cumulative benefits. Roosters receiving 20 and 30 mg/day exhibited superior semen motility, sperm concentration, overall semen quality, antioxidant enzyme and testosterone concentration compared to both the control and the 40 mg/day group. These results indicate a dose-dependent effect of proanthocyanidin, with the most pronounced enhancement observed at the highest supplementation level. In conclusion, the study provides strong evidence that dietary oral administration of proanthocyanidin can improve reproductive efficiency in broiler breeder roosters by enhancing semen quality traits.

Keywords: proanthocyanidin, oral administration, semen quality, antioxidant status, breeders

1 Introduction

Fertility in breeder flocks is a critical trait that directly determines the number of hatching eggs available for commercial production. Poultry breeding companies emphasize fertility because it directly affects hatchability rates, chick quality, and ultimately the economic efficiency of poultry operations (Ansari, 2024). However, semen quality in broiler breeder males declines noticeably with advancing age, particularly after 50 weeks. This decline is largely attributed to oxidative stress and an imbalance in the delicate redox system that regulates cellular metabolism (Ali et al., 2017). Oxidative stress results from an overproduction of reactive oxygen species (ROS) combined with a decrease in the efficiency of the body's antioxidant defense systems. This imbalance damages lipids, proteins, and nucleic acids, thereby impairing sperm structure and function. Additionally, oxidative stress disrupts steroidogenesis within the testes and alter testosterone metabolism, the key male reproductive hormone, leading to its conversion into estrogen through the catalytic action of the aromatase enzyme (Al-Zalzaly and Ali, 2024). A reduction in testicular testosterone strongly correlates with diminished sperm concentration, reduced motility, and morphological abnormalities (Jaber, 2023). Older males often exhibit decreased testicular weight, degeneration of seminiferous tubules, and reduced activity of antioxidant enzymes such as superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GPx). This decline in enzymatic defense mechanisms leads to increased

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ROS accumulation in spermatozoa, resulting in lipid peroxidation of membranes, DNA fragmentation, and apoptosis (Asl et al., 2018). These challenges underscore the urgent need for safe, effective, and economically viable strategies to mitigate oxidative stress and preserve semen quality in ageing breeder roosters. Recently, attention has shifted toward natural compounds with strong antioxidant activity as potential dietary supplements in poultry production. Natural phenols and flavonoids are widely recognized for their ability to neutralize free radicals and protect cellular components from oxidative damage (Shakir, 2024).

Among the diverse classes of flavonoids, proanthocyanidins have attracted considerable scientific interest. Proanthocyanidins are oligomeric and polymeric flavonoid compounds commonly found in grape seeds, cranberries, cocoa, pine bark, and other plant-derived materials (Mannino et al., 2021). They are water-soluble pigments responsible for the characteristic colors of many fruits and plants. These compounds exhibit a broad spectrum of biological activities, including antioxidant, anti-inflammatory, antimicrobial, and cardioprotective effects (Rauf et al., 2019). In the context of reproductive physiology, proanthocyanidins act as potent scavengers of free radicals, thereby preventing oxidative damage to spermatozoa and supporting the structural and functional integrity of gametes (Lv et al., 2022). It enhances the endogenous antioxidant defense system by stimulating the expression and activity of enzymatic antioxidants such as SOD, CAT, and GPx. This activation ensures effective neutralization of excessive ROS, thereby maintaining redox balance within cells (Mannino et al., 2021). By mitigating oxidative stress, proanthocyanidins reduce lipid peroxidation in sperm membranes, preserve DNA integrity, and improve mitochondrial function, all of which are vital for sperm motility and fertilizing capacity. Another important aspect is their role in regulating corticosterone secretion under stressful conditions, which helps minimize stress-induced reproductive impairments (Rodríguez et al., 2022). Various additives with antioxidant properties have been investigated for their potential to improve semen quality. Adding 50 mg of camphor per kg of feed to 29-week-old Ross 308 broiler breeder roosters improves seminal characteristics, testosterone concentration, and reproductive performance (Raei et al., 2021). Dietary supplementation with 0.45 mg/day of selenium-enriched yeast enhances sperm motility, plasma membrane function and integrity, as well as ejaculation volume and reproductive performance in 50-week-old Ross 308 roosters (Sabzian-Melei et al., 2022). Breeder diets containing 1% spray-dried plasma reduce late embryonic mortality and sperm cell defects, primarily in 63-week-old broiler breeders (Granghelli et al., 2025). However, little is known about the impact of proanthocyanidins in poultry species, particularly broiler breeder roosters. Therefore, the present study examined the qualitative characteristics of semen from male broiler breeders following daily oral administration of proanthocyanidin at various dosage levels, along with measurements of antioxidant levels and sex hormone concentrations.

2 Material and Methods

2.1 Ethics Approval

All experimental procedures related to animal management and handling were conducted in accordance with the ethical guidelines established by the Scientific Committee in the Department of Animal Production, College of Agriculture, Al-Qasim Green University (Babil, Iraq) under approval number Agri, No. 021,10,24. The welfare of birds was carefully monitored throughout the experimental period, and every effort was made to minimize stress and ensure humane treatment.

2.2 Birds and Housing Management

The experiment was performed from November 1, 2023, to January 31, 2024, spanning 13 weeks using twenty Ross 308 broiler breeder males aged 54 weeks with an average body weight of approximately 5.0 kg. Birds were housed individually in floor-rearing cages measuring 1.5 × 1.5 m. Each pen was bedded with coarse sawdust at a depth of 10 cm to ensure proper insulation and hygiene. A photoperiod of 14 hours light and 10 hours darkness was maintained throughout the study using artificial lighting. Clean drinking water was supplied *ad libitum* via flipped floor nipples. Temperature and ventilation were adjusted regularly to maintain optimal rearing conditions.

2.3 Training and Experimental Design

Prior to data collection, all roosters underwent a two-week training period to accustom them to semen collection procedure. Following this adaptation, the experiment was conducted over six consecutive experimental periods (28 days/period). In each period, semen was collected three times from every rooster to assess its qualitative traits. Between each period, a recovery interval of one week was given to minimize stress and to allow physiological recuperation. The twenty birds were randomly assigned to four experimental treatments in a completely randomized design. Each treatment included five replicates, with one rooster per replicate.

2.4 Experimental Treatments

T1 (control): no supplementation with proanthocyanidin.
T2: daily oral supplementation with 20 mg

proanthocyanidin. T3: daily oral supplementation with 30 mg proanthocyanidin. T4: daily oral supplementation with 40 mg proanthocyanidin. The proanthocyanidin used in this study was a purified commercial preparation (≥ 97% purity) obtained from a certified supplier. Doses were administered individually to ensure accurate intake.

2.5 Nutrition and Feeding

All birds were offered a restricted daily ration of 140 g per bird, formulated in accordance with the manufacturer's recommendations for broiler breeder males. The basal diet was formulated to contain 2800 kcal/kg metabolizable energy and 14.5% crude protein. Feed was provided in mash form once daily to ensure uniform intake of treatment supplements. A group of 20 Ross 308 roosters were fed a standard basal diet supplemented with encapsulated proanthocyanidins at concentrations of 20, 30, and 40 mg per bird per day for 13 weeks (Ali et al., 2017).

2.6 Semen Collection and Evaluation

Semen was collected using the abdominal massage technique during each sampling period. The collected semen was immediately transferred to pre-warmed tubes and assessed for volume, mass motility, individual motility, concentration, viability, and morphological characteristics (Ali et al., 2017). Microscopic evaluation was carried out using phase-contrast microscopy, while sperm concentration was determined spectrophotometrically. All measurements were repeated three times per period to reduce random error.

2.7 Antioxidant Status and Hormonal Profile

The levels of glutathione, catalase, and malondialdehyde were measured using colorimetric test kits (Elabscience Biotechnology Inc., Houston, TX, USA). Serum testosterone and estrogen levels were determined using a chicken-specific ELISA kit (Elabscience Biotechnology Inc., Houston, TX, USA), following the manufacturer's instructions.

2.8 Statistical analysis

Data were analyzed using a completely randomized design with four treatments and five replicates. The general linear model procedure in SAS software (SAS, 2012) was employed to test treatment effects. Differences among means were compared using Duncan's multiple range test (Duncan, 1955) at a significance level of $P \leq 0.05$.

3 Results and Discussion

3.1 Measurement of Semen Volume (μL)

Table 1 shows the effect of different doses of proanthocyanidin on semen volume, with no significant differences observed during the first, second, and fourth periods of the experiment. In the third period, treatment T3 significantly outperformed the other treatments. In the fifth period, treatments T3 and T2 significantly outperformed treatments T1 and T4. During the sixth period, treatment T3 outperformed all other treatments, while treatment T2 outperformed treatments T1 and T4.

3.2 Total Sperm Motility %

Table 2 shows the effect of different levels of proanthocyanidin on the measurement of collective motility of spermatozoa (%). For the first period, there is a significant advantage for treatment T3 over treatments T1 and T4, and there is no significant difference between treatments T3 and T2. As for the second and fourth periods, there is no significant difference between the treatments. In the third period, treatment 4 outperformed treatment 2, and there was no significant difference between treatments 1, 2, and 3. In the fifth period, treatment 3 outperformed all other treatments in the experiment, and in the sixth period, treatments 2 and 3 outperformed treatments 1 and 4.

Table 1 Effect of daily supplementation of proanthocyanidin on semen volume (μL) of broiler breeder males during six experimental weeks

Treatment	Week1 (μL)	Week2 (μL)	Week3 (μL)	Week4 (μL)	Week5 (μL)	Week6 (μL)
T1	3,500.0 ±28.87	3,500.0 ±50.00	4,000.0 ±40.82^{b}	4,000.0 ±40.82	3,750.0 ±25.00^{b}	3,250.0 ±47.87^{c}
T2	4,250.0 ±25.00	3,500.0 ±28.87	4,500.0 ±28.87^{b}	4,750.0 ±25.00	6,000.0 ±40.82^{a}	5,500.0 ±28.87^{b}
T3	4,000.0 ±40.82	3,750.0 ±47.87	5,500.0 ±28.87^{a}	4,250.0 ±25.00	5,250.0 ±47.87^{a}	7,000.0 ±40.82^{a}
T4	3,250.0 ±25.00	3,750.0 ±47.87	3,500.0 ±28.87^{b}	3,750.0 ±47.87	3,750.0 ±25.00^{b}	3,500.0 ±28.87^{c}
Significance level	0.23	0.46	0.03	0.12	0.04	0.04

NS – no significant; *significant differences at ($P \leq 0.05$); means within the same column bearing different superscript letters differ significantly (*$P \leq 0.05$). Treatments: T1 – control without dosing; T2, T3, T4 – daily dosing of proanthocyanidin at 20, 30, and 40 mg, respectively

Table 2 Effect of daily supplementation of proanthocyanidin on semen motility (%) of broiler breeder males during six experimental weeks

Treatment	Week1 (%)	Week2 (%)	Week3 (%)	Week4 (%)	Week5 (%)	Week6 (%)
T1	77.50 ±1.44^{bc}	82.50 ±1.44	82.50 ±2.50^{ab}	86.25 ±1.25	83.75 ±1.25^{b}	77.50 ±1.44^{b}
T2	81.25 ±1.25^{ab}	78.75 ±1.25	78.75 ±1.25^{b}	86.25 ±1.25	85.00 ±2.04^{b}	92.50 ±1.44^{a}
T3	86.25 ±2.39^{a}	80.00 ±6.12	85.00 ±3.53^{ab}	87.50 ±1.44	93.75 ±1.25^{a}	92.50 ±1.44^{a}
T4	72.50 ±2.50^{c}	86.25 ±2.39	88.75 ±1.25^{a}	87.50 ±1.44	87.50 ±2.50^{b}	73.75 ±1.25^{b}
Level of significance	0.02	0.67	0.03	0.59	0.03	0.01

N. S. – no significant; *significant differences at ($P \leq 0.05$); means within the same column bearing different superscript letters differ significantly (*$P \leq 0.05$). Treatments: T1 – control without dosing; T2, T3, T4 – daily dosing of proanthocyanidin at 20, 30, and 40 mg, respectively

3.3 Individual Motility %

From Table 3, the effect of dosing different levels of proanthocyanidin compound on the measurement of individual motility of sperm (%) shows that there is no significant difference between the treatments during the second and fourth periods of the experiment. However, in the first and fifth periods, we observe a significant superiority of the third treatment over all other treatments. As for the third period, the fourth treatment outperformed the first and second treatments, and for the sixth period, the second and third treatments outperformed the first and fourth treatments.

3.4 Dead Spermatozoa

Table 4 shows the effect of different levels of proanthocyanidin compound on the percentage of dead spermatozoa. We observe no significant difference between the treatments during the first, second and third periods. However, in the fourth, fifth and sixth periods, there is a significant increase in the percentage of dead spermatozoa in the first treatment (control treatment) compared to the other treatments.

3.5 Antioxidant Enzymes, Malondialdehyde, and Sex Hormone Concentrations

Table 5 illustrates the effect of proanthocyanidin supplementation on oxidative enzymes, malondialdehyde, and sex hormones levels in male broiler breeders. The results showed that the third treatment recorded the highest values compared to all other experimental groups, with significant differences

Table 3 Effect of different levels of proanthocyanidin on individual sperm motility (%) of broiler breeder males during six experimental weeks

Treatment	Week1 (%)	Week2 (%)	Week3 (%)	Week4 (%)	Week5 (%)	Week6 (%)
T1	67.50 ±1.44^{bc}	72.50 ±1.44	72.50 ±2.50^{cb}	76.25 ±1.25	73.75 ±1.25^{b}	67.50 ±1.44^{b}
T2	71.25 ±1.25^{b}	68.75 ±1.25	68.75 ±1.25^{c}	76.25 ±1.25	75.00 ±2.04^{b}	82.50 ±1.44^{a}
T3	77.50 ±1.44^{a}	72.50 ±4.33	77.50 ±1.44^{ab}	78.75 ±1.25	83.75 ±1.25^{a}	82.50 ±1.44^{a}
T4	65.00 ±2.88^{c}	76.25 ±2.39	78.75 ±1.25^{a}	77.50 ±1.44	77.50 ±2.50^{b}	63.75 ±1.25^{b}
Significance level	0.03	0.39	0.04	0.52	0.03	0.01

N. S. – no significant; *significant differences at ($P \leq 0.05$); means within the same column bearing different superscript letters differ significantly (*$P \leq 0.05$). Treatments: T1 – control without dosing; T2, T3, T4 – daily dosing of proanthocyanidin at 20, 30, and 40 mg, respectively

Table 4 Effect of different levels of proanthocyanidin on the percentage of dead spermatozoa (%) of broiler breeder males during six experimental weeks

Treatment	Week1 (%)	Week2 (%)	Week3 (%)	Week4 (%)	Week5 (%)	Week6 (%)
T1	7.25 ±1.31	4.75 ±0.25	6.25 ±0.25	7.50 ±0.96^{a}	10.75 ±0.48^{a}	10.75 ±0.65^{a}
T2	5.75 ±0.48	5.25 ±0.63	5.50 ±0.65	3.75 ±0.75^{b}	4.00 ±0.41^{c}	4.00 ±0.65^{c}
T3	5.25 ±0.63	4.50 ±0.65	4.25 ±1.11	5.25 ±1.03^{ab}	4.75 ±0.25^{c}	4.75 ±0.41^{c}
T4	5.25 ±0.25	5.75 ±0.75	4.75 ±0.48	5.25 ±0.63^{ab}	7.75 ±0.25^{b}	7.75 ±0.65^{b}
Significance level	0.44	0.33	0.51	0.04	0.01	0.01

N.S. – no significant; *significant differences at ($P \leq 0.05$); means within the same column bearing different superscript letters differ significantly (*$P \leq 0.05$). Treatments: T1 – control without dosing; T2, T3, T4 – daily dosing of proanthocyanidin at 20, 30, and 40 mg, respectively

Table 5 Effect of proanthocyanidin supplementation on oxidative parameters, antioxidant enzymes, malondialdehyde concentration (µmol/mol), and sex hormones (ng/ml) in blood serum of male broiler breeders

Treatment	Glutathione (µmol/mol)	Catalase (µmol/mol)	MDA (µmol/mol)	Testosterone (ng/ml)	Estrogen (ng/ml)
T1	13.82 ±1.63^{b}	26.80 ±5.87^{b}	41.42 ±7.27^{a}	7.90 ±0.90^{c}	327.90 ±1.25^{a}
T2	17.35 ±6.87^{a}	28.13 ±5.06^{ab}	27.12 ±1.20^{c}	11.66 ±0.84^{b}	280.20 ±1.65^{c}
T3	18.59 ±3.55^{a}	31.90 ±2.48^{a}	26.27 ±0.78^{c}	13.86 ±1.52^{a}	185.50 ±2.91^{d}
T4	12.52 ±4.78^{b}	20.94 ±5.20^{c}	31.88 ±0.93^{b}	8.67 ±0.39^{c}	291.63 ±0.90^{b}
Significance level	0.04	0.02	0.02	0.01	0.01

N.S. – no significant; *significant differences at ($P \leq 0.05$); means within the same column bearing different superscript letters differ significantly (*$P \leq 0.05$). Treatments: T1 – control without dosing; T2, T3, T4 – daily dosing of proanthocyanidin at 20, 30, and 40 mg, respectively

($P \leq 0.05$). Regarding testosterone concentration, the third treatment (30 mg/day) was significantly higher than the other treatments, with a clear difference ($P \leq 0.05$). At the same time, no significant difference was observed between the first and fourth treatments in testosterone levels. For malondialdehyde concentration (MDA), the first treatment demonstrated superiority over all other groups, whereas no significant differences were detected among the remaining treatments.

3.6 Discussion

The findings of this study demonstrated that daily supplementation with proanthocyanidin led to marked improvements in several semen quality parameters of ageing broiler breeder males. The elevated MDA concentration observed in the control group of this study provides evidence of oxidative stress. Increased oxidative stress and lipid peroxidation disrupt sperm membranes, DNA, and proteins, leading to reduced motility and viability (Ghadimi et al., 2024). In contrast, the reduced levels of oxidative stress in proanthocyanidin-treated groups confirm its protective action. These positive changes may be explained by the antioxidant properties of proanthocyanidins, which act by scavenging ROS, stabilizing sperm cell membranes, and enhancing mitochondrial functionality (Mannino et al., 2021). It is well established that mitochondria are central to sperm motility and viability through their role in ATP production, and any intervention that protects mitochondrial integrity contributes directly to semen quality (Liu and Bolling, 2024). Similar findings were reported in bovine semen studies, where antioxidants preserved DNA integrity and reduced cryodamage (AE et al., 2022).

Enzymatic antioxidant activity was also enhanced by proanthocyanidin in this study. Glutathione and catalase are key defenses against oxidative stress, and their increased activity indicates a strengthened antioxidant system capable of detoxifying hydrogen peroxide and other peroxides in poultry (Luo et al., 2007; Panda et al., 2008a,b; Ognik and Krauze, 2016). Proanthocyanidin supplementation enhances semen quality in ageing broiler breeder males by reducing oxidative stress, enhancing enzymatic antioxidant activity, and restoring hormonal balance (Studer et al., 2022). From a practical standpoint, improvements in semen quality in breeder males have direct implications for fertility, hatchability, and the efficiency of artificial insemination programs (Bakst and Dymond, 2013). By integrating proanthocyanidin supplementation into management systems, fertility rates and reproductive lifespan of breeder flocks can be extended, ensuring higher hatching egg production and economic sustainability.

Regarding hormonal responses, the third treatment (30 mg/day) produced the highest testosterone concentration, which is crucial for spermatogenesis, libido, and semen production. This result is consistent with evidence showing that antioxidants inhibit aromatase activity, thereby reducing the conversion of testosterone to estrogen and restoring androgen balance (Ali et al., 2017; Al-Zalzaly and Ali, 2024). Phytogenic antioxidants have also been shown to elevate luteinizing hormone (LH) and improve fertility traits, reinforcing the link between oxidative balance and endocrine function (Adel dust et al., 2017). Abbas and Ali (2023) demonstrated that dietary proanthocyanidin supplementation improved physiological and reproductive traits in laying hens. Abdul Amir and Ali (2024) confirmed that açaí supplementation enhanced semen motility, ejaculate volume, and fertility in roosters, emphasizing the consistent effects of plant-derived antioxidants across avian species. Al-Hassani et al. (2023) investigated both nutritional and molecular approaches complementary roles in poultry productivity, suggesting that nutritional antioxidants may influence gene expression patterns associated with oxidative balance and steroidogenesis. Alani and Al-Bayati (2021) showed that vitamin E and selenium improved semen quality and fertility in roosters. Shakir and Ali (2025) also confirmed that nano-curcumin supplementation enhanced semen traits in breeder males, indicating the wide applicability of natural antioxidants in reproductive management. Hussein et al. (2025) found similar antioxidative effects of curcumin and nano-curcumin in broilers under toxic dietary stress. Collectively, these studies emphasize the consistent pattern of improved semen quality through antioxidant supplementation.

4 Conclusions

The results of this study indicate that administering proanthocyanidin at doses of 20 mg/day and 30 mg/day to male broiler breeders significantly improves semen quality. This is evidenced by increases in ejaculate volume, overall semen motility, individual sperm motility, sperm concentration, antioxidant levels, and testosterone hormone concentration. Future research could investigate the long-term effects of proanthocyanidin supplementation on reproductive performance and overall health in broiler breeders. Additionally, studies could assess the potential benefits of proanthocyanidin supplementation in other poultry species or livestock to determine its broader applicability in animal husbandry.

Conflict of Interest

The authors declare that there is no conflict of interest.

Author Contributions

Emad Abdulgabbar Ali: Conceptualization, writing-original draft, supervision, methodology. Nihad Abdul-Lateef Ali: Data curation, investigation. Tahreer M. Al-Thuwaini: Data curation, investigation. In addition, all authors reviewed and approved the final manuscript.

AI and AI-Assisted Technologies use Declaration

No generative AI tools/AI-assisted technologies were used during the preparation of the manuscript.

References

Abbas, N. K., & Ali, N. A.-L. (2023). Physiological changes caused by dietary proanthocyanidin supplementation at varying doses in laying hens. *IOP Conference Series: Earth and Environmental Science*, 1262(7), 072040. https://doi.org/10.1088/1755-1315/1262/7/072040

Abdul Amir, O. H., & Ali, E. A. (2024). Effect of adding different levels of Euterpe oleracea (açaí) fruit to the diet of laying hens on the volume of ejaculate, sperm motility and some histological characteristics. *IOP Conference Series: Earth and Environmental Science*, 1371(7), 072023. https://doi.org/10.1088/1755-1315/1371/7/072023

Adel dust, H., Ghanbari, A., & Hassanpour, H. (2017). Effect of medicinal plants containing bioactive antioxidants on testosterone and fertility in ageing males. *Iranian Journal of Applied Animal Science*, 7(4), 633–639.

AE, A. K., Dowidar, Y., El-Nagar, H., Wafa, W., El-Ratel, I. & Mousbah, A. (2022). A review on various antioxidants utilized in bovine semen extenders. *Journal of Applied Veterinary Sciences*, 7(2), 13–24. https://doi.org/10.21608/javs.2022.115377.1120

Alani, A. M., & Al-Bayati, H. A. (2021). Effect of adding vitamin E and selenium to rooster diets on semen quality and fertility traits. *Anbar Journal of Agricultural Sciences*, 19(3), 145–153. https://doi.org/10.32649/ajas.2021.176250

Al-Bayar, M. A., & Attallah, M. H. (2024). Effect of centrifugation and addition of melatonin hormone to semen on characteristics of seminal plasma in cooled semen of Iraqi local roosters. *Anbar Journal of Agricultural Sciences*, 22(1), 517–527. https://doi.org/10.32649/ajas.2024.183750

Al-Hassani, A. S. A. (2011). Effect of dietary supplementation with different levels of arginine on some blood traits of laying hens. *International Journal of Poultry Science*, 10(10), 705–709. https://doi.org/10.3923/ijps.2011.705.709

Al-Hassani, A. S., Al-Hassani, D. H., & Abdul-Hassan, I. A. (2023). PCR-RFLP analysis of insulin-like growth factor 2 gene polymorphisms in two commercial broiler chicken strains (Cobb 500 and Hubbard F-15) and their associations with performance traits. *Archives of Razi Institute*, 78(3), 1153–1157. https://doi.org/10.22092/ARI.2022.359894.2504

Ali, E. A., Zhandi, M., Towhidi, A., Zaghari, M., Ansari, M., Najafi, M., & Deldar, H. (2017). Letrozole, an aromatase inhibitor, reduces post-peak age-related regression of rooster reproductive performance. *Animal Reproduction Science*, 183, 110–117. https://doi.org/10.1016/j.anireprosci.2017.05.010

Al-Zalzaly, A. H. N., & Ali, E. A. J. (2024). Effect of adding different levels from *Brassica oleracea* var. *caulerpa* L. in some semen quality traits of the broiler breeder males (Ross 308). *Euphrates Journal of Agricultural Science,* 16(1).

Ansari, M. (2024). Recent strategies to mitigate reproductive ageing in male broiler breeders: a review. *Animal Reproduction Science*, 107570. https://doi.org/10.1016/j.anireprosci.2024.107570

Asl, R. S., Shariatmadari, F., Sharafi, M., Torshizi, M. A. K., & Shah Verdi, A. (2018). Improvements in semen quality, sperm fatty acids, and reproductive performance in aged Ross breeder roosters fed a diet supplemented with a moderate ratio of n-3: n-6 fatty acids. *Poultry Science*, 97(11), 4113–4121. https://doi.org/10.3382/ps/pey314

Bakst, M. R., & Dymond, J. S. (2013). Artificial insemination in poultry. *Success in Artificial Insemination-Quality of Semen and Diagnostics Employed* (pp. 722–723).

Bansal, A. K., & Bilaspuri, G. S. (2010). Impacts of oxidative stress and antioxidants on semen functions. *Veterinary Medicine International*, 2010, 686137. https://doi.org/10.4061/2011/686137

Duncan, D.B. (1955). Multiple Rang and Multiple *F*-test. *Biometrics*, 11, 4–42.

Ghadimi, M., Najafi, A., Sharifi, S. D., Mohammadi-Singhateh, A., & Mehr, M. R. A. (2024). Effects of dietary Moringa oleifera leaf extract on semen characteristics, fertility, and hatchability in aged broiler breeder roosters. *Poultry Science*, 103(4), 103491. https://doi.org/10.1016/j.psj.2024.103491

Granghelli, C. A., Lopes, M. H. S., Rangel, L., Campbell, J., Polo, J., Crenshaw, J., Jorge-Neto, P.N., Ferreira, H.L., Moura, V.S., & Araujo, L. F. (2025). Effect of dietary supplementation of spray-dried plasma on performance and semen quality in aging broiler breeders. *Animal Reproduction*, 22(2), e20240111. https://doi.org/10.1590/1984-3143-AR2024-0111

Hussein, S. S., Mousa, B. H., Nafea, H. H., & Al-Thuwaini, T. M. (2025). Impact of curcumin and nano-curcumin supplementation in lead acetate-contaminated diets on the productive performance and oxidative status of broilers. *Journal of Animal Health and Production*, 13(3), 521–530. https://doi.org/10.17582/journal.jahp/2025/13.3.521.530

Jaber, A. S. (2023). *The effect of adding the aqueous extract of Moringa oleifera leaves to drinking water on the reproductive and physiological characteristics of laying mother cocks* (*Lohmann Brown*). MSc thesis, College of Agriculture, Al-Qasim Green University.

Liu, C., & Bolling, B. W. (2024). Dietary proanthocyanidins for improving gut immune health. *Current Opinion in Food Science*, 56, 101133. https://doi.org/10.1016/j.cofs.2024.101133

Luo, X. G., Li, S. F., Lu, L., Liu, B., Kuang, X., Shao, G. Z., & Yu, S. X. (2007). Gene expression of manganese-containing superoxide dismutase as a biomarker of manganese bioavailability for manganese sources in broilers. *Poultry Science*, 86(5), 888–894. https://doi.org/10.1093/ps/86.5.888

Lv, J. M., Gouda, M., Bekhit, A. E. D., He, Y. K., Ye, X. Q., & Chen, J. C. (2022). Identification of novel bioactive proanthocyanidins with potent antioxidant and anti-proliferative activities from kiwifruit leaves. *Food Bioscience*, 46, 101554. https://doi.org/10.1016/j.fbio.2022.101554

Merzah, L. H., & Ali, N. A.-L. (2023). Different diets of maca roots (Lepidium meyenii) affect several physiological blood characteristics of broiler chickens under oxidative stress. *IOP Conference Series: Earth and Environmental Science*, 1259(1), 012071. https://doi.org/10.1088/1755-1315/1259/1/012071

Ognik, K., & Krauze, M. (2016). The potential for using enzymatic assays to assess the health of turkeys. *World's Poultry Science Journal*, 72(3), 535–550. https://doi.org/10.1017/S0043933916000246

Panda, A. K., Ramarao, S. V., Raju, M. V. L. N., & Chatterjee, R. N. (2008b). Responses in production performance, immunity and antioxidant status of White Leghorn layers under tropical summer conditions to supplementation with vitamins E and C. *British Poultry Science*, 49(5), 592–599. https://doi.org/10.1080/00071660802290588

Panda, A. K., Rao, S. V. R., Raju, M. V. L. N., & Sharma, S. R. (2008a). The role of dietary antioxidants in poultry. *World's Poultry Science Journal*, 64(2), 151–163. https://doi.org/10.1017/S0043933908000027

Raei, H., Torshizi, M. A. K., Sharafi, M., & Ahmadi, H. (2021). Improving seminal quality and reproductive performance in male broiler breeder by supplementation of camphor. *Theriogenology*, 166, 1–8. https://doi.org/10.1016/j.theriogenology.2021.02.002

Rodríguez, R. M., Cortés-Espinar, A. J., Soliz-Rueda, J. R., Feillet-Coudray, C., Casas, F., Colom-Pellicer, M., et al. (2022). Time-of-day circadian modulation of grapeseed procyanidin extract (GSPE) in hepatic mitochondrial dynamics in cafeteria-diet-induced obese rats. *Nutrients*, 14(4), 774. https://doi.org/10.3390/nu14040774

Sabzian-Melei, R., Zare-Shahneh, A., Zhandi, M., Yousefi, A. R., & Rafieian-Naeini, H. R. (2022). Effects of dietary supplementation of different sources and levels of selenium on the semen quality and reproductive performance in aged broiler breeder roosters. *Poultry science*, 101(10), 101908. https://doi.org/10.1016/j.psj.2022.101908

SAS. (2018). Statistical Analysis System, User's Guide. Statistical. Version 9. 4th ed. SAS. Inst. Inc. Cary. N.C. USA.

Shakir, A. R., & Ali, E. A. (2025). Effect of supplementing different levels of nano-curcumin on broiler breeder males' semen quality. *IOP Conference Series: Earth and Environmental Science*, 1487(1), 012151. https://doi.org/10.1088/1755-1315/1487/1/012151

Shakir, A. R. (2024). *Effect of dosing different levels of nano-curcumin carried by nano zinc in some physiological and reproductive traits of broiler breeder chickens*. MSc thesis, College of Agriculture, Al-Qasim Green University.

Studer, J. M., Schweer, W. P., Gabler, N. K., & Ross, J. W. (2022). Functions of manganese in reproduction. *Animal Reproduction Science*, 238, 106924.