Research Article

Serum Mineral Profiles and Testosterone Levels as Integrated Fertility Biomarkers in Boerka Bucks

Tongku Nizwan Siregar[1*], Hafizuddin Hafizuddin[1], Sri Wahyuni[2], Eka Meutia Sari[3], Teuku Armansyah[4], Husnurrizal Husnurrizal[1]

[1]Laboratory of Reproduction, Faculty of Veterinary Medicine, Universitas Syiah Kuala, Banda Aceh, Indonesia; [2]Laboratory of Anatomy, Faculty of Veterinary Medicine, Universitas Syiah Kuala, Banda Aceh, Indonesia; [3]Department of Animal Science, Faculty of Agriculture, Universitas Syiah Kuala, Banda Aceh, Indonesia; [4]Laboratory of Pharmacology, Faculty of Veterinary Medicine, Universitas Syiah Kuala, Banda Aceh, Indonesia.

Abstract | Semen quality in small ruminants is influenced by endocrine and nutritional factors, including mineral availability. Identifying mineral and hormonal biomarkers of fertility is crucial for improving breeding efficiency in Boerka bucks, a composite breed widely used in meat production. This study evaluated the association between serum macro- and micromineral concentrations and semen quality, and identified minerals with potential as fertility biomarkers. Ten Boerka bucks (±3 years old) were categorized into fertile (n = 5) and infertile (n = 5) groups based on semen characteristics. Semen quality parameters, serum testosterone, and concentrations of macro- and microminerals were analyzed. Correlation analyses were performed to determine associations between minerals, testosterone, and semen quality. Fertile bucks showed higher semen volume, sperm concentration, motility, and normal morphology than infertile bucks. Serum testosterone was significantly ($P < 0.05$) higher in fertile animals (25.9 ng/mL) compared with infertile ones (15.5 ng/mL). Concentrations of calcium (Ca), iron (Fe), zinc (Zn), and phosphate (P) were significantly higher in fertile bucks ($P < 0.05$), while cobalt (Co), magnesium (Mg), and manganese (Mo) did not differ between groups. Fe was positively correlated with sperm motility ($r = 0.495$), and Zn was positively correlated with sperm concentration ($r = 0.196$) and viability ($r = 0.390$). Testosterone showed positive correlations with all semen parameters, particularly motility ($r = 0.66$) and viability ($r = 0.70$) ($P < 0.05$). Serum mineral status especially Fe and Zn together with testosterone levels, is strongly associated with semen quality in Boerka bucks. Mineral–hormonal profiling may serve as a useful approach for identifying fertile males and enhancing artificial insemination programs.

Keywords | Boerka bucks, Semen quality, Serum minerals, Testosterone, Fertility biomarkers, Artificial insemination

Received | December 04, 2025; **Accepted** | January 28, 2026; **Published** | March 29, 2026
***Correspondence** | Tongku Nizwan Siregar, Laboratory of Reproduction, Faculty of Veterinary Medicine, Universitas Syiah Kuala, Banda Aceh, Indonesia; **Email:** siregar@usk.ac.id
Citation | Siregar TN, Hafizuddin H, Wahyuni S, Sari EM, Armansyah T, Husnurrizal H (2026). Serum mineral profiles and testosterone levels as integrated fertility biomarkers in Boerka bucks. J. Anim. Health Prod. 14(2): 519-526.
DOI | https://dx.doi.org/10.17582/journal.jahp/2026/14.2.519.526
ISSN (Online) | 2308-2801

INTRODUCTION

The Boerka goat is a composite breed resulting from the crossbreeding of Boer goats with Kacang goats. This breed is characterized by predominantly white body coloration, a two-tone coat pattern with a dark brown head, a convex facial profile, horns, pendulous ears, and a slightly concave rump in both males and females. Boerka goats possess desirable growth performance, weight gain, and strong adaptability to humid tropical environments when provided with adequate nutrition (Nugroho *et al.*, 2019). Efforts to enhance reproductive efficiency can be achieved through the application of artificial insemination (AI) technology, whose success largely depends on semen

quality (Jha *et al*., 2020).

Frozen semen plays a critical role in AI programs, as semen collected from genetically superior males can be used to improve the genetic merit of the population. The production of high-quality frozen semen is closely linked to sperm quality. Previous studies have attempted to improve semen characteristics through the administration of PGF2α (Husnurrizal *et al*., 2021; Panjaitan *et al*., 2024) and GnRH (Syafruddin *et al*., 2020; Sutriana *et al*., 2022). However, these hormonal interventions have limitations, as the enhancement of reproductive performance is temporary and highly dependent on exogenous hormonal stimulation.

Several reports highlight the interaction between nutrition and reproduction in sheep and goats (Vazquez-Armijo *et al*., 2011; Chao *et al*., 2023). Among nutritional factors, macro- and microminerals (trace elements) have received particular attention. In cattle, semen that meets the criteria for successful cryopreservation is associated with high serum concentrations of Zn and Fe, suggesting their potential use as biomarkers for freezability (Pipan *et al*., 2021). To date, no reports are available regarding microminerals as biomarkers of semen quality in goats, despite their potential relevance in predicting post-thaw semen performance.

A variety of accurate methods exist for identifying fertility biomarkers in males; however, many of these methods are time-consuming, expensive, and inefficient because they can only be applied to a limited number of animals at one time (Singh *et al*., 2018). Therefore, simple, affordable, accurate, and reliable biomarkers are needed to assess semen freezability based on fresh semen or blood analysis. Biomarkers based on macro- and micromineral concentrations (Pipan *et al*., 2021) and testosterone levels (Hafizuddin *et al*., 2023) offer promising avenues for application in Boerka goats.

Beyond predicting semen quality, mineral-based biomarkers can also be used to improve semen processing techniques to maintain high post-thaw performance. Optimization may be achieved by supplementing extenders with minerals identified as biomarkers. This supplementation can be performed in the form of mineral nanoparticles (Orzolek *et al*., 2021). Nanoparticles, defined as particles measuring 1–100 nm, exert effects similar to antibiotics during semen processing by protecting sperm against bacterial contamination and enhancing motility and viability in a dose-dependent manner (Hill and Li, 2017). Previous studies have demonstrated the antioxidant properties of nanoparticles in semen from rams, bulls, rats, and poultry (Singh *et al*., 2018). The present findings could be further developed for producing mineral nanoparticles identified as biomarkers to support improved semen cryopreservation procedures in Boerka goats. Therefore, this study aimed to identify macro- and micromineral profiles associated with semen quality and freezability in Boerka goats, evaluate the potential of these minerals as biomarkers for predicting post-thaw semen performance, and provide a scientific basis for the future development of mineral-based strategies, including nanoparticle supplementation, to improve frozen semen quality in Boerka goats.

MATERIALS AND METHODS

This study was conducted from May to December 2025. The Boerka goats used for semen collection and evaluation were obtained from the UPT for Ruminant Breeding, Lobusona, under the Department of Plantation and Livestock Services of North Sumatra Province. Analyses of macro- and microminerals, including Fe, Zn, Mn, Mg, Ca, and Co, were performed at the Chemical Engineering Laboratory, Faculty of Engineering, Syiah Kuala University, Banda Aceh, while testosterone analysis was carried out at the Physiology Laboratory, Faculty of Veterinary Medicine, Syiah Kuala University.

Research procedures

Semen collection and quality evaluation

A total of 10 semen plasma samples and 10 blood serum samples were collected from Boerka bucks (n= 10), all approximately 3 years of age. The animals were maintained at the UPT for Ruminant Breeding, Lobusona, under good management and feeding practices. Semen collection was performed using an electroejaculator. Immediately after collection, semen was evaluated both macroscopically and microscopically. Macroscopic parameters included volume, color, pH, and consistency, while microscopic assessments covered sperm concentration, motility, viability, and abnormalities. Semen samples were classified into fertile (F) and infertile (IF) groups. The fertile group met the criteria of ≥75% motility, ≥60% progressive motility, and ≥75% normal morphology (n= 5). Samples failing to meet at least one of these criteria were categorized as infertile (n= 5).

Blood sample collection and preparation

Blood samples were collected from the jugular vein into EDTA-containing vacutainer tubes, homogenized, and placed in a chilled cool box. Samples were centrifuged at 3000 rpm for 15 minutes, and the resulting plasma was transferred into microtubes and stored at –20°C until analysis.

Mineral concentration measurement

The concentrations of macro- and microminerals (Fe, Zn, Mn, Mg, Ca, and Co) were measured using spectrophotometry and Atomic Absorption

Spectrophotometry (AAS; Shimadzu 700).

Testosterone measurement

Testosterone concentrations were measured on the day of semen collection using an ELISA kit. The testosterone antibody showed 0.8% cross-reactivity with 5α-dihydrotestosterone, 0.9% with androstenedione, 3.3% with 11β-hydroxytestosterone and 19-nortestosterone, and <0.1% with epitestosterone, estrogens, progesterone, cortisol, and danazol. The mean inter-assay coefficient of variation for duplicate samples was 6.71%, and the intra-assay coefficient was 3.28%.

Data analysis

Semen characteristics and macro- and micromineral concentrations in semen plasma and blood serum were presented descriptively. Statistical comparisons between fertile (F) and infertile (IF) groups for fresh semen, post-thaw semen, and mineral concentrations were analyzed using the Wilcoxon test. Correlations between mineral concentrations in blood serum and semen plasma were assessed using Spearman's correlation coefficient, and adjusted P-values were used to determine statistical significance.

RESULTS AND DISCUSSION

Semen quality characteristics of fertile and infertile Boerka bucks

An evaluation of semen characteristics was performed to determine differences in ejaculate quality between fertile and infertile Boerka bucks. The assessment included macroscopic parameters (volume, color, odor, and consistency) and microscopic parameters (sperm concentration, mass motility, individual motility, viability, and morphological abnormalities). These parameters were used to assess the physiological capability of spermatozoa to support the fertilization process. Overall, the findings revealed clear variations in semen quality between the two groups. The fertile Boerka bucks exhibited higher mean semen volume and sperm concentration compared with the infertile bucks. Individual motility and sperm abnormalities also differed markedly between groups, whereas mass motility and viability were relatively similar (Table 1).

The evaluation of semen characteristics revealed variations between fertile and infertile Boerka goats in both macroscopic and microscopic parameters. Overall, the semen volume of fertile Boerka goats (1.66±0.90 mL) was higher than that of infertile goats (1.45±1.20 mL) (Table 1). This difference indicates that the secretory activity of the accessory glands is more optimal in fertile males, contributing to an increased ejaculate volume. The uniform color and odor of semen in both groups (creamy and typical) indicate no abnormalities in the composition of seminal plasma, while the thick consistency suggests relatively high sperm concentrations in both groups.

Microscopically, the sperm concentration of fertile goats (208.20±142.07 × 10^6/mL) was significantly ($P < 0.05$) higher than that of infertile goats (180.20±103.05 × 10^6/mL) (Table 1). This elevated concentration reflects more efficient spermatogenesis, particularly in the proliferation and differentiation stages of spermatids into mature spermatozoa. This process is strongly influenced by intratesticular testosterone levels, Sertoli cell activity, and adequate testicular blood perfusion (Hafez and Hafez, 2016). Meanwhile, individual motility in fertile goats (80.52±0.85%) was also higher than in infertile goats (72.18±17.05%), indicating that spermatozoa from fertile animals possess better progressive motility. Sperm motility is a key parameter influenced by molecular factors such as proAKAP4 and supplementation treatments such as EGF. Good motility is essential for supporting successful fertilization (Ribas-Maynou *et al*., 2023).

In contrast to these parameters, mass motility and viability did not show clear differences between the groups (Table 1). The relatively similar values of mass motility (2.40±0.55 vs. 2.50±0.76) indicate that the collective movement of spermatozoa in both groups remains within the normal range, while comparable viability (66.91±16.96% vs. 68.75±13.54%) suggests that membrane integrity is still well-maintained in both groups. Nevertheless, morphological abnormalities were lower in the fertile group (2.22±1.49) compared with the infertile group (12.65±5.92).

Reproductive hormones exert differential impacts on semen quality. Elevated LH and FSH levels tend to reduce sperm parameters, while testosterone plays a supportive but not strongly dominant role. Thus, endocrine evaluation combined with semen analysis is essential in assessing infertility (Long *et al*., 2025). Therefore, the higher concentration and individual motility observed in the fertile group are likely related to more stable reproductive hormone levels and a better physiological response of testicular tissues. This finding aligns with previous studies reporting that males with higher sperm motility and membrane integrity have higher conception success rates (Jalius and Depison, 2008). Consequently, superior semen characteristics in fertile Boerka goats may serve as physiological indicators in selecting high-quality breeding males.

However, this study has several limitations, including a limited sample size and the absence of hormonal profiling and pregnancy tests to confirm the direct association

between semen quality and fertilization ability. Further studies integrating semen quality parameters with testosterone levels, sperm DNA integrity, and conception rates are required to obtain a more comprehensive understanding of the factors determining fertility in Boerka bucks.

Testosterone concentration in fertile and infertile Boerka goats

The results showed that testosterone levels in fertile Boerka goats (25.9±9.23 ng/mL) were significantly ($P < 0.05$) higher than in infertile goats (15.5±7.83 ng/mL), as presented in Table 1. The testosterone concentration in fertile Boerka goats is relatively high compared to other goat breeds. In Gembrong goats, testosterone levels were reported at 13.30±3.90 ng/mL. In Kacang goats, the mean (± SD) testosterone concentrations in adult males and females were 7.25±1.45 ng/mL and 0.95±0.77 ng/mL, respectively (Gholib *et al.*, 2016).

Table 1: Semen characteristics and testosterone levels of fertile and infertile Boerka bucks.

Semen characteristics	Treatment groups	
	Fertile Boerka goats (n=5)	Infertile Boerka goats (n=5)
Macroscopic		
Volume (mL)	1.66±0.90[b]	1.45±1.20[a]
Color	Cream	cream
Odor	Characteristic	characteristic
Consistency	Thick	thick
Microscopic		
Concentration (x10^6 sperms/mL)	208.20±142.07[b]	180.20±103.05[a]
Mass motility (%)	2.40±0.55	2.50±0.76
Individual motility (%)	80.52±0.85[b]	72.18±17.05[a]
Viability (%)	66.91±16.96	68.75±13.54
Abnormalities (%)	2.22±1.49[b]	12.65±5.92[a]
Testosterone (ng/mL)	25.9±9.23[b]	15.5±7.83[a]

[a,b] Different superscript letters in a row showing significant difference ($P<0.05$).

Correlation analysis demonstrated that testosterone levels were positively associated with all semen quality parameters in Boerka goats, although the strength of the correlations varied across variables. Testosterone showed a moderate positive correlation with sperm concentration ($r = 0.42$) and a strong, significant correlation with motility ($r = 0.66$; $P < 0.05$) and sperm viability ($r = 0.70$; $P < 0.05$), as presented in Table 2. These findings confirm that increases in testosterone levels tend to be followed by improvements in semen performance, particularly sperm motility and survival, which are key indicators of male fertility.

Table 2: Correlation between testosterone concentration and semen quality in Boerka goats.

Variable	Semen quality		
	Concentration	Motility	Viability
Testosterone	0.42	0.66*	0.70*

* $P < 0.05$

The significant differences observed between the fertile and infertile groups indicate that fertility status is strongly associated with testosterone concentration. These findings are consistent with the report of Bodu *et al.* (2025), who stated that fertility in male livestock is influenced by testosterone as a biomarker, although it remains interconnected with other reproductive parameters. Testosterone is produced by Leydig cells in response to luteinizing hormone (LH) secretion and plays a critical role in the development of male characteristics. It has been reported that men and male dogs with poor semen quality exhibit lower testosterone concentrations. In circulation, testosterone is largely bound to proteins primarily albumin and, more specifically, sex hormone-binding globulin (SHBG) while only the free fraction exerts biological activity (Laurent, 2016; Almeida *et al.*, 2017; Holst and Nilsson, 2023).

Testosterone plays an essential role in maintaining spermatogenesis, the function of accessory reproductive glands, and libido (Rojas-Zambrano *et al.*, 2025). Therefore, the lower testosterone levels in the infertile group indicate impaired testicular function or endocrine regulation, which subsequently affects semen quality and quantity. The significant differences between the fertile and infertile bucks further underscore the central role of testosterone in supporting the reproductive function of Boerka goats.

These results align with the findings of Baharun *et al.* (2022), Rajak *et al.* (2014), and Dasrul *et al.* (2020). Testosterone concentration shows a positive correlation ($P < 0.01$) with semen characteristics such as sperm motility (0.813), normal morphology (0.639), and libido (0.952) (Baharun *et al.*, 2022). Rajak *et al.* (2014) and Dasrul *et al.* (2020) also reported a relationship between testosterone levels and semen characteristics particularly motility and sperm morphology in crossbred bulls (Friesian Holstein × Tharparkar) and Aceh cattle. Similarly, Gulia *et al.* (2010) reported that testosterone deficiency can reduce semen quality.

The findings of this study support the potential use of testosterone concentration as a fertility biomarker in Boerka bucks. Physiologically, the strong positive correlation between testosterone and both motility and viability indicates that this androgenic hormone is a key determinant of semen quality. Motility and sperm

morphology are considered the most important factors in determining semen quality, as only motile spermatozoa are capable of fertilizing the oocyte (Morrell and Rodriguez-Martinez, 2009). In addition to motility, the ability of spermatozoa to fertilize the oocyte depends on normal morphology and intact chromatin (Nagy *et al*., 2013).

In this study, motility and viability showed a significant correlation with testosterone ($P < 0.05$), whereas sperm concentration exhibited a moderate but non-significant correlation with testosterone ($P > 0.05$). This finding is consistent with the report of Alba *et al*. (2024), which states that testosterone enhances membrane stability, acrosomal integrity, and motility capacity through the modulation of key proteins in the sperm tail, collectively influencing sperm motility and morphology. Testosterone affects the expression of oxidative enzymes, antioxidants, and structural proteins that maintain motility and reduce oxidative damage. The moderate and non-significant correlation between testosterone and sperm concentration suggests that other factors such as ejaculation frequency, nutrition, or physiological status may influence sperm numbers.

These findings provide practical implications, indicating that testosterone measurement may serve as a fertility biomarker for selecting Boerka bucks for semen freezing programs. However, testosterone alone cannot serve as the sole indicator of fertility, as several semen parameters are influenced by management, health, age, and environmental factors. Therefore, combining testosterone evaluation with semen quality analysis is necessary for more accurate reproductive assessments. Overall, the significant correlations between testosterone, motility, and viability underscore the central role of this hormone in supporting the reproductive performance of Boerka bucks. These conclusions are consistent with the literature, which emphasizes that the stability of androgenic hormones is crucial for successful spermatogenesis and male fertility.

Serum macro- and micromineral concentrations

Minerals are essential components for maintaining physiological balance and reproductive function in males. The availability of macro- and microminerals in the blood strongly influences spermatogenesis, semen quality, and the fertilizing capacity of spermatozoa. Minerals such as calcium (Ca), phosphorus (P), zinc (Zn), iron (Fe), and magnesium (Mg) play important roles in enzymatic regulation, membrane stability, and mitochondrial activity in testicular and sperm cells. Imbalances whether deficiencies or excess can impair sperm quality and ultimately lead to infertility (Fallah *et al*., 2018).

To determine the relationship between mineral status and male fertility, serum macro- and micromineral levels were measured in fertile and infertile Boerka bucks on the day of semen collection. The results of the mineral concentration analysis are presented in Table 3.

The analysis revealed significant ($P < 0.05$) differences in the concentrations of several macro- and microminerals between fertile and infertile Boerka bucks. Levels of calcium (Ca), iron (Fe), zinc (Zn), and phosphate (P) were significantly higher in fertile bucks than in infertile bucks ($P < 0.05$), whereas cobalt (Co), magnesium (Mg), and manganese (Mo) did not differ markedly between the groups (Table 3).

Table 3: Concentrations of macro- and microminerals in blood serum on the day of semen collection.

Minerals	Treatment groups	
	Fertile Boerka goats (n=5)	Infertile Boerka goats (n=5)
Calcium (Ca)	0.62±0.13^{b}	0.30±0.11^{a}
Cobalt (Co)	0.01±0.01	0.01±0.01
Iron (Fe)	2.63±2.1^{b}	0.74±0.11^{a}
Magnesium (Mg)	0.42±0.16	0.42±0.16
Manganese (Mn)	0.22±0.28	0.34±0.41
Zinc (Zn)	0.76±0.23^{b}	0.18±0.01^{a}
Phosphate (P)	0.99±0.06^{b}	0.87±0.05^{a}

^{a,b} Different superscript letters in a row showing significant difference ($P<0.05$).

The higher Ca concentration in fertile bucks indicates its role in supporting spermatogenesis and sperm motility. Calcium functions as a key regulator of sperm physiology, playing essential roles in capacitation, hyperactivation, the acrosome reaction, and fertilization (Garriga *et al*., 2026). Reduced Ca levels in infertile bucks may reflect impaired ionic metabolism affecting germ cell function. In males, calcium deficiency disrupts the regulation of sperm motility and capacitation, both of which are critical for successful fertilization (Yahyavi *et al*., 2024). However, Ca exhibited a negative correlation with all semen parameters, particularly sperm viability ($r= -0.474$; $P= 0.420$). This suggests that excessively high Ca concentrations do not necessarily improve semen quality. Ca^{2+} supplementation increases metabolic activity but may also trigger capacitation-like changes that impair sperm functionality. Intracellular Ca^{2+} accumulation may reduce motility and disrupt metabolic balance, potentially due to H^{+} ion accumulation in the mitochondrial intermembrane space (Garriga *et al*., 2026).

The higher Fe concentration in fertile bucks highlights its role in enzymatic systems and testicular cellular respiration. Fe showed a moderate positive correlation with sperm motility ($r= 0.495$; $P= 0.096$), indicating that increased serum Fe tends to enhance sperm motility (Table 4). This aligns with Fe's involvement in mitochondrial oxidative

Table 4: Correlation analysis between semen quality and blood concentrations of Ca, Fe, Zn, and P in Boerka bucks.

Variable	Blood serum minerals							
	Ca		Fe		Zn		P	
Semen characteristics	r	P-value	R	P-value	r	P-value	r	P-value
Concentration	-0.023	0.970	0.075	0.018	0.196	0.042	0.226	0.774
Motility	-0.205	0.741	0.495	0.096	0.012	0.985	0.182	0.818
Viability	-0.474	0.420	-0.145	0.816	0.390	0.046	-0.444	0.556

processes that generate energy for sperm movement. Fe also acts as an antioxidant, serving as a cofactor for catalase, which protects spermatozoa from oxidative damage. However, excessive Fe may induce lipid peroxidation and membrane damage, negatively affecting motility. Overall, higher Fe levels are associated with improved sperm motility and fertility potential (Rodriguez *et al*., 2021). Iron is an essential component of cytochrome and catalase, contributing to energy metabolism in Sertoli and Leydig cells. Iron deficiency increases oxidative stress, reduces antioxidant enzyme activity, and ultimately impairs reproductive function (Tsao *et al*., 2022).

Zinc concentration in semen is strongly associated with semen quality parameters, and Zn deficiency is a major risk factor for reduced semen quality (Osadchuk *et al*., 2021). This finding is consistent with the present study, in which fertile bucks displayed significantly higher Zn levels than infertile bucks ($P < 0.05$; Table 3). Zn showed positive correlations with sperm concentration ($r = 0.196$; $P = 0.042$) and viability ($r = 0.390$; $P = 0.046$), as shown in Table 4. Numerous studies confirm that zinc ions (Zn^{2+}) play a major role in male fertility. Zn influences multiple sperm functions, including motility, capacitation, and acrosomal exocytosis processes essential for fertilization. Across vertebrate and invertebrate species, Zn^{2+} modulates sperm motility. In humans, high seminal Zn^{2+} concentrations reduce motility, whereas lowering Zn^{2+} enhances motility. During epididymal transit, reduced intracellular Zn^{2+} enables the development of progressive and hyperactivated motility during capacitation. Mechanistically, Zn^{2+}signals through the zinc-sensing receptor GPR39 located in the sperm tail and acrosome. Activation of GPR39 triggers intracellular cascades involving increased cAMP, activation of CatSper Ca^{2+} channels, elevated intracellular Ca^{2+}, PKA activation, and the Src–EGFR–PLC pathway. These pathways induce hyperactivated motility and regulate F-actin remodeling required for capacitation and acrosomal exocytosis (Allouche-Fitoussi and Breitbart, 2020). Conversely, Zn deficiency impairs spermatogenesis, increases sperm abnormalities, and reduces serum testosterone levels (Fallah *et al*., 2018).

Similarly, higher P concentrations in fertile bucks support the energetic metabolism of sperm, particularly ATP production, which is essential for motility and viability (Yousef *et al*., 2017). Suwimonteerabutr *et al*. (2023) reported that supplementing semen extenders with 0.04% phosphorus and vitamin B12 improved sperm motility, kinetic parameters, and overall sperm quality. In the present study, phosphorus showed a positive but nonsignificant correlation with semen quality, particularly sperm concentration ($r = 0.226$; $P = 0.774$), as presented in Table 4. Phosphate plays a key role in sperm energy metabolism, especially in ATP formation required for motility and the acrosome reaction (Yousef *et al*., 2017). Although nonsignificant, this positive trend suggests a potential contribution of phosphorus to efficient sperm function.

In general, these results reinforce the view that blood mineral status particularly Ca, Fe, Zn, and P can serve as physiological indicators for assessing the reproductive status of male animals. Deficiencies in these minerals have the potential to disrupt spermatogenesis and reduce fertility performance (Yimer *et al*., 2023). Conversely, the absence of significant differences in Mg and Co levels indicates that these two minerals remain relatively stable in the basal metabolism of goats, with no evident functional differences related to fertility under these physiological conditions. Correlation analysis was conducted to determine the relationship between macro-minerals (Ca and P) and micro-minerals (Fe and Zn) in blood serum with semen quality characteristics of Boerka goats, including sperm concentration, motility, and viability. These minerals were selected because they are known to exert significant influences distinguishing fertile and infertile males.

CONCLUSIONS AND RECOMMENDATIONS

This study demonstrates that semen quality in Boerka bucks is closely linked to their mineral and hormonal profiles. Fertile males exhibited superior semen characteristics and significantly higher serum testosterone, along with elevated concentrations of Ca, Fe, Zn, and P. Correlation analyses showed that Fe was positively associated with sperm motility, while Zn correlated with sperm concentration and viability. Testosterone exhibited strong positive correlations with all semen parameters. These findings indicate that

serum Fe, Zn, and testosterone are key physiological indicators of fertility in Boerka bucks. Mineral–hormonal profiling thus offers a promising strategy for identifying fertile males and improving the efficiency of breeding and artificial insemination programs.

ACKNOWLEDGEMENTS

The author expresses gratitude to the Rector of Universitas Syiah Kuala for funding support through the Penelitian Professor for the fiscal year 2025, with contract number 347/UN11.L1/PG.01.03/14138-PTNBH/2025.

NOVELTY STATEMENT

This study provides the first evidence that integrated serum mineral–hormonal profiling, particularly involving iron, zinc, and testosterone levels, can reliably differentiate fertile and infertile Boerka bucks. By demonstrating strong mineral–semen quality relationships in this composite meat breed, the research introduces Fe and Zn as potential physiological biomarkers for buck fertility, offering a novel diagnostic approach to enhance breeding selection and artificial insemination efficiency.

AUTHOR'S CONTRIBUTION

Conceptualization: TNS, SW and HH. Methodology, formal analysis, and investigation: EMS, TA, and HH. Writing original draft, review and editing: TNS, HH, EMS, and SW. All authors have read and agreed to the published version of the manuscript.

Ethical statement

Ethical approval was not required for this study based on the recommendation of the Ethical Feasibility Team of the Faculty of Veterinary Medicine, Universitas Syiah Kuala, as no experimental or invasive procedures were applied to the animals and semen collection was conducted using a routine and standardized procedure.

Generative AI and AI-assisted technology statement

The authors declare that generative AI and AI-assisted technologies were used solely to support the development of the manuscript framework and to improve the clarity, grammar, and academic quality of the English language. No AI tools were used for data analysis, data interpretation, figure generation, or drawing scientific conclusions. All scientific content, analyses, and interpretations remain the sole responsibility of the authors.

Conflict of interest

The authors have declared no conflict of interest.

REFERENCES

Alba AC, Nguyen L, O'Flaherty C (2024). Testosterone regulates sperm maturation and membrane stability through androgen-mediated signaling pathways. Reprod. Biol. Endocrinol., 22(1): 45–56.

Allouche-Fitoussi D, Breitbart H (2020). The role of zinc in male fertility. Int. J. Mol. Sci., 21(20): 7796. https://doi.org/10.3390/ijms21207796

Almeida S, Rato L, Sousa M, Alves MG, Oliveira PF (2017). Fertility and sperm quality in the aging male. Curr. Pharm. Des., 23: 4429–4437. https://doi.org/10.2174/1381612823666170503150313

Baharun A, Said S, Arifiantini RI, Karja NWK (2022). Karakteristik semen dan korelasi antara konsentrasi testosteron dengan libido pejantan sapi Simental. Acta Vet. Indones., 10(1): 1–7. https://doi.org/10.29244/avi.10.1.1-7

Bodu M, Hitit M, Memili E (2025). Harnessing the value of fertility biomarkers in bull sperm for buck sperm. Anim. Reprod. Sci., 272: 107643. https://doi.org/10.1016/j.anireprosci.2024.107643

Chao HH, Zhang Y, Dong PY, Gurunathan S, Zhang XF (2023). Comprehensive review on regulators of spermatogenesis and male fertility. Front. Nutr., 9: 1–15. https://doi.org/10.3389/fnut.2022.1063510

Dasrul D, Wahyuni S, Sugito S, Hamzah A, Zaini Z, Haris A, Gholib G (2020). Correlation between testosterone concentration with scrotal circumference and semen characteristic in Aceh bulls. E3S Web of Conf., 151: 1–5. https://doi.org/10.1051/e3sconf/202015101015

Fallah A, Mohammad-Hasani A, Colagar AH (2018). Zinc as an essential element for male fertility: A review. J. Reprod. Infertil., 19(2): 69–81.

Garriga F, Codina J, Benaiges J, Yeste M, Llavanera M (2026). Calcium homeostasis in preserving sperm function during liquid storage of pig semen. Theriogenology, 249: 117638. https://doi.org/10.1016/j.theriogenology.2025.117638

Gholib, Wahyuni S, Kadar OH, Adam M, Lubis TM, Azhar, Akmal M, Siregar TN, Armansyah T, Nugraha TP (2016). Measurement of serum testosterone in Kacang goat using ELISA: Importance of kit validation. J. Kedokt. Hewan., 10(1): 32–36. https://doi.org/10.21157/j.ked.hewan.v10i1.3367

Gulia S, Sarkar M, Kumar V, Meyer HHD, Prakash BS (2010). Testosterone secretion patterns in zebu, crossbred cattle and buffaloes during growth. Trop. Anim. Health Prod. 42: 1143–1148. https://doi.org/10.1007/s11250-010-9538-x

Hafez ESE, Hafez B (2016). Reproduction in farm animals. Wiley-Blackwell.

Hafizuddin H, Husnurrizal H, Siregar TN, Eriani K, Wahyuni S, Ahsan MM, Sutriana A, Anwar A, Aliza D (2023). Amelioration of seminal plasma testosterone in Gembrong goats after $PGF_2\alpha$ administration. J. Medik. Vet., 6(2): 256–261. https://doi.org/10.20473/jmv.vol6.iss2.2023.256-261

Hill EK, Li J (2017). Current and future prospects for nanotechnology in animal production. J. Anim. Sci. Biotechnol., 8: 26. https://doi.org/10.1186/s40104-017-0157-5

Holst BS, Nilsson S (2023). Associations between age,

weight, testosterone, and prostate size in dogs. Theriogenology, 198: 356–360. https://doi.org/10.1016/j.theriogenology.2022.12.047

Husnurrizal H, Aritonang AS, Siregar TN, Armansyah T, Hafizuddin H (2021). $PGF_2\alpha$ addition in semen extenders improves post-thaw sperm motility in Waringin sheep. Livest. Anim. Res. 19(2): 210–216. https://doi.org/10.20961/lar.v19i2.49186

Jalius, Depison (2008). Korelasi motilitas progresif dan keutuhan membran sperma semen beku sapi Ongole terhadap keberhasilan inseminasi. J. Ilm. Ilmu-Ilmu Peternak., 11(2): 75–80. https://doi.org/10.22437/jiiip.v11i2.752

Jha PK, Alam MGS, Al-Mansur MD, Talukder MRI, Naher N, Rahman AKMA, Hall DC, Bari FY (2020). Effects of frozen–thawed ram sperm number on fertility in synchronized ewes. J. Anim. Reprod. Biotechnol., 35: 190–197. https://doi.org/10.12750/JARB.35.2.190

Laurent MR (2016). Sex hormone-binding globulin regulation of androgen bioactivity: Validation of the free hormone hypothesis. Sci. Rep., 6: 35539. https://doi.org/10.1038/srep35539

Long TQT, Tu NTD, Anh ND (2025). Semen quality and reproductive hormone profiles in male patients. Tap. Chi Nghien Cuu. Y Hoc., 196(11E17): 230–239. https://doi.org/10.52852/tcncyh.v196i11E17.4169

Morrell JM, Rodriguez-Martinez H (2009). Biomimetic techniques for improving sperm quality in animal breeding. J. Androl., 30: 1–9.

Nagy S, Johannisson A, Wahlsten T, Ijäs R, Andersson M, Rodriguez-Martinez H (2013). Sperm chromatin and morphology in relation to fertility in dairy sires. Theriogenology, 79: 1153–1161. https://doi.org/10.1016/j.theriogenology.2013.02.011

Nugroho T, Kustiyani C, Ratriyanto A, Widyas N, Prastowo S (2019). Reproductive rate performance of Boer goat and its F1 cross. IOP Conf. Ser. Earth Environ. Sci., 334(1): 012008. https://doi.org/10.1088/1755-1315/334/1/012008

Orzołek A, Rafalska KT, Otowska WA, Kordan W, Korzekwa AJ (2021). Influence of zinc and manganese nanoparticles on turkey semen stored at 4°C. Animals, 11: 3289. https://doi.org/10.3390/ani11113289

Osadchuk L, Kleshchev M, Danilenko A, Osadchuk A (2021). Impact of seminal and serum zinc on semen quality in young Russian men. J. Trace Elem. Med. Biol., 68: 126855. https://doi.org/10.1016/j.jtemb.2021.126855

Panjaitan B, Husnurrizal H, Edward VY, Nishcaya DA, Armansyah T, Siregar TN, Adam M, Sayuti A, Sutriana A, Hafizuddin H (2024). Effects of interval between $PGF_2\alpha$ injection and semen collection on sperm and testosterone levels in Aceh bulls. Adv. Anim. Vet. Sci., 12(2): 337–341. https://doi.org/10.17582/journal.aavs/2024/12.2.337.341

Pipan MZ, Zrimšek P, Strajn BJ, Vrtač KP, Knific T, Mrkun J (2021). Macro- and microelements in serum and seminal plasma as biomarkers for bull sperm cryotolerance. Acta Vet. Scand., 63(1): 1–10. https://doi.org/10.1186/s13028-021-00590-2

Rajak SK, Kumaresan A, Gaurav MK, Layek SS, Mohanty TK, Aslam KMM, Tripathi UK, Prasad S, De S (2014). Testicular cell indices and testosterone in relation to age and semen quality in crossbred bulls. Asian-Australas. J. Anim. Sci., 27(11): 1554–1561. https://doi.org/10.5713/ajas.2014.14139

Ribas-Maynou J, Barranco I, Salas-Huetos A (2023). Sperm quality and fertility of livestock animals. Animals, 13: 604. https://doi.org/10.3390/ani13040604

Rodrigue RDAlcaide-Ruggiero L, Zamora RB, Rodríguez JG, Paz S, Hardisso A, Torre DL, Gutierrez A, Rubio C, Hess-Medler S (2021). Association between seminal Fe levels and semen quality. Hum. Reprod., 36(Suppl 1): deab130.058. https://doi.org/10.1093/humrep/deab130.058

Rojas-Zambrano JG, Rojas-Zambrano A, Rojas-Zambrano AF (2025). Impact of testosterone on male health: A systematic review. Cureus, 17(4): e82917. https://doi.org/10.7759/cureus.82917

Singh AK, Rajak SK, Kumar P, Kerketta S, Yogi RK (2018). Nutrition and bull fertility: A review. J. Entomol. Zool. Stud., 6: 635–643.

Sutriana A, Siregar TN, Sirait FC, Melia J, Hafizuddin H (2022). Effect of GnRH on sperm quality of Bali cattle. J. Sain Vet., 40(3): 307–313. https://doi.org/10.22146/jsv.65067

Suwimonteerabutr J, Ketkaew P, Netiprasert G, Khaopong C, Osiriphan B, Sriamornrat P, Nuntapaitoon M (2023). Supplementing semen extenders with a combination of phosphorus and vitamin B12 Improves post-thawed cryopreserved rooster semen quality. Front. Vet. Sci., 10: 1301186. https://doi.org/10.3389/fvets.2023.1301186

Syafruddin S, Iryandi F, Rahmi RAS, Husnurrizal H, Armansyah T, Panjaitan B, Sayuti A, Sutriana A, Aliza D, Hafizuddin H (2020). Effect of GnRH on semen quality and testosterone in Nubian goats. Vet. Zootech., 77(99): 19–21.

Tsao CW, Liao YR, Chang TC, Liew YF, Liu CY (2022). Effects of iron supplementation on testicular function of iron-deficient rats. Nutrients, 14: 2063. https://doi.org/10.3390/nu14102063

Vázquez-Armijo JF, Rojo R, López D, Tinoco JL, González A, Pescador N, Domínguez-Vara IA (2011). Trace elements in sheep and goat reproduction: A review. Trop. Subtrop. Agroecosyst., 14: 1–13. https://doi.org/10.1007/s12011-011-9113-y

Yahyavi SK, Boisen IM, Cui Z, Jorsal MJ, Kooij I, Holt R, Juul A, Jensen MB (2024). Calcium and vitamin D homeostasis in male fertility. Proc. Nutr. Soc., 83(2): 95–108. https://doi.org/10.1017/S002966512300486X

Yimer A, Forsido SF, Addis G, Ayelign A (2023). Nutritional composition of some wild edible plants consumed in Southwest Ethiopia. Heliyon 9(6): e16541. https://doi.org/10.1016/j.heliyon.2023.e16541

Yousef MI, Kamel KI, Esmail AM (2017). Influence of phosphorus deficiency on semen quality and fertility in rabbits. Anim. Reprod. Sci., 185: 94–101.