

EFFECTS OF RED SEAWEED (*GRACILARIA TENUISTIPITATA*) ON HAEMATOLOGICAL AND SERUM BIOCHEMICAL PARAMETERS IN COMMERCIAL BROILER CHICKENS

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ABSTRACT

*The present study evaluated the effects of graded dietary inclusion of red seaweed (*Gracilaria tenuistipitata*) on haematological and serum biochemical parameters in commercial broiler chickens. A total of 200 day-old Vencobb 430Y broiler chicks were randomly allotted to four dietary treatments with five replicates of ten birds each. The experimental diets consisted of a basal ration supplemented with red seaweed at 0% (T1, control), 1% (T2), 2% (T3), and 4% (T4) and were fed for 35 days. The shade-dried red seaweed was analyzed for chemical composition and contained 11.87% crude protein, 6.41% crude fibre, 0.72% crude fat, 47.20% crude ash, 2.47% calcium, 0.33% phosphorus, and 2347.10 kcal/kg gross energy, with palmitic (26.72%), oleic (31.16%), and linoleic acids (21.56%) as the predominant fatty acids. Haematological parameters, including haemoglobin concentration, total erythrocyte count, and total leucocyte count, did not differ significantly ($P > 0.05$) among treatment groups. Similarly, serum biochemical indices such as total protein, albumin, globulin, alanine aminotransferase (ALT), aspartate aminotransferase (AST), cholesterol, and triglycerides remained unaffected by dietary treatments. These findings indicate that dietary supplementation of red seaweed up to 4% maintains normal blood profiles, liver function, protein metabolism, and lipid homeostasis without inducing adverse metabolic effects. The study demonstrates that *G. tenuistipitata* can be safely incorporated into broiler diets as a functional, non-conventional feed ingredient.*

Key words: Broiler chicken, Red seaweed, Chemical composition, Haematology, Serum biochemistry

Received :07.01.2026

Revised : 08.04.2026

Accepted : 17.04.2026

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INTRODUCTION

Broiler production plays a central role in the Indian poultry sector, with the country ranking among the world's leading producers of chicken meat. This growth is driven by a rapidly expanding commercial industry characterized by large-scale integration, high feed efficiency, and increasing domestic demand for affordable animal protein (Churchil, 2022). Reflecting this dominance, during 2024–25, out of a total meat production of 10.50 million tonnes and a per-capita meat availability of 7.50 kg, poultry alone contributed 5.17 million tonnes, corresponding to approximately 3.7 kg per-capita availability, thereby accounting for the largest share of India's meat output (BAHS, 2025). Despite this progress, rising costs and fluctuations in conventional feed ingredients, particularly maize and soybean, continue to constrain the sustainability of poultry production, necessitating the identification of economical alternative feed resources and functional additives that can maintain productivity while supporting bird health (Churchil, 2022). Accordingly, contemporary poultry nutrition emphasizes the development and scientific evaluation of alternative feed strategies that enhance biological efficiency without compromising health, highlighting the importance of systematic assessment of novel feed ingredients using physiological and biochemical indicators (Kanagaraju *et al.*, 2019; Churchill and Suresh, 2021).

Marine algae, commonly referred to as seaweeds, comprise a diverse group of photosynthetic organisms inhabiting

marine environments and are broadly classified into red, green, and brown macroalgae, although some occur as microalgae such as phytoplankton (Holdt and Kraan, 2011; Makkar *et al.*, 2020). Seaweeds are increasingly recognized as sustainable functional feed ingredients in poultry nutrition because they provide a wide range of bioactive compounds such as polysaccharides, polyphenols, pigments, and minerals with antimicrobial, antioxidant, anti-inflammatory, immunomodulatory, and prebiotic properties. These attributes enhance gut health, immune competence, nutrient utilization, and overall performance, while offering a viable alternative to antibiotic growth promoters (Reddy *et al.*, 2024). Their renewable nature, ease of production, cost-effectiveness, and rich nutrient profile further enable partial replacement of imported feed proteins and contribute to food security without compromising bird health or productivity (Al-Khalaifah and Al-Nasser, 2024). In addition, seaweeds act as prebiotics by selectively stimulating beneficial gut microbiota, thereby improving host health and product quality (Evans and Critchley, 2014; Holdt and Kraan, 2011). The polysaccharide fraction of seaweeds is particularly important in modulating intestinal microbial balance and supporting digestive efficiency (Kulshreshtha *et al.*, 2014).

Among marine algae, red seaweeds are of particular nutritional interest due to their high content of polyunsaturated fatty acids, including eicosapentaenoic acid (EPA; ω -3, C20:5) and arachidonic acid (AA; ω -6, C20:4) (Holdt and Kraan,

2011; Balasubramanian *et al.*, 2021). Palmitic acid (C16:0) constitutes the major saturated fatty acid, while oleic acid is the predominant monounsaturated fatty acid (MacArtain *et al.*, 2007). In addition, red algae are characterized by low lipid content and high dietary fibre, particularly soluble fibres such as sulphated galactans (agars and carrageenans), xylans, and floridean starch, and are rich sources of macro- and micro-minerals including sodium, potassium, calcium, magnesium, iron, zinc, manganese, copper, and iodine (Holdt and Kraan, 2011; MacArtain *et al.*, 2007; Makkar *et al.*, 2020; Kulshreshtha *et al.*, 2014).

Evidence from experimental studies supports the functional potential of seaweeds in poultry diets. For instance, the brown seaweed *Ascophyllum nodosum* has been proposed as a natural alternative to antibiotic growth promoters (Dierick *et al.*, 2009), while red seaweeds such as *Chondrus crispus* and *Sarcodiotheca gaudichaudii* have been shown to improve growth performance, egg efficiency, and gut health in laying hens (Kulshreshtha *et al.*, 2014). Subsequent studies demonstrated that dietary seaweed inclusion enhances feed efficiency and bird health by increasing beneficial gut bacteria and strengthening innate immune responses (Kulshreshtha *et al.*, 2014). Moreover, brown seaweed supplementation has been reported to alleviate heat stress-induced intestinal damage by improving gut microbiota, villus architecture, and antioxidant and anti-inflammatory responses, thereby preserving nutrient absorption and health under high-temperature conditions, an effect particularly

relevant to poultry production in tropical and subtropical regions (Oretomiloye and Adewole, 2024).

The chemical composition of seaweeds varies considerably with geographical location, environmental conditions, seasonal factors, and processing methods, necessitating region-specific characterization for accurate nutritional evaluation (Holdt and Kraan, 2011; Makkar *et al.*, 2020). Accordingly, analysis of the chemical composition and fatty acid profile of red seaweed from peninsular India is essential to determine its precise nutritional value and potential application in poultry feeds. Furthermore, information on the systemic physiological responses of broiler chickens to dietary red seaweed, particularly with respect to haematological and serum biochemical indicators of metabolic health, remains limited. Therefore, the present study was undertaken to characterize the chemical composition of regionally sourced red seaweed and to evaluate its effects on haematological and serum biochemical parameters in commercial broiler chickens, thereby establishing its nutritional suitability, safety, and functional potential under Indian production conditions.

MATERIALS AND METHODS

Collection of red seaweed and chemical analysis

Red seaweed (*Gracilaria tenuistipitata*) was collected from Pulicat Lake, the second-largest brackish water lagoon in India, located along the Bay of Bengal and extending across the states of Andhra Pradesh and Tamil Nadu (Fig. 1).

The harvested seaweed was thoroughly washed, shade-dried (Fig. 2), and coarsely ground (Fig. 3) for further use.

The dried seaweed sample was analyzed for proximate composition, including moisture, crude protein, crude fibre, ether extract, total ash, and acid-insoluble ash. Mineral profile, free fatty acids, and omega-3 fatty acid contents were also determined following standard procedures described by AOAC (2005). Gross energy was measured using a bomb calorimeter, and metabolizable energy was subsequently calculated from the gross energy values.

Birds, treatments and experimental design

A total of 200 day-old straight run commercial broiler chicks (Vencobb 430Y strain) were randomly allotted to four dietary treatments, each with five replicates of ten birds. The dietary treatments consisted of a basal diet supplemented with dried red seaweed at 0% (T1, control), 1% (T2), 2% (T3), and 4% (T4) inclusion levels. The feeding trial was conducted for a period of 35 days.

Four isocaloric and isonitrogenous experimental diets were formulated in accordance with BIS (2024) specifications and fed to the birds from 0 to 35 days of age. Birds were reared under standard management practices with ad libitum access to feed and water throughout the experimental period.

Haematological parameters

At the end of the experiment,

blood samples (3 mL) were collected from three male and three female birds from each treatment group using EDTA-coated vacutainer tubes for haematological analysis. Total erythrocyte count (TEC) and total leucocyte count (TLC) were determined using a Neubauer haemocytometer with Nambiar's diluting fluid and Natt-Herrick's solution, respectively. Haemoglobin concentration was estimated by the Sahli's acid haematin method.

Serum biochemical parameters

On day 35, six birds from each treatment group were slaughtered, and blood samples were collected. Serum was separated by centrifugation and analyzed for total protein, albumin, globulin, alanine aminotransferase (ALT), aspartate aminotransferase (AST), cholesterol, and triglycerides using an A15 Biosystem Auto Analyzer with commercially available diagnostic kits (AGAPPE Diagnostics), following the CHOD-PAP and standard colorimetric methodologies (Gornall *et al.*, 1949; Doumas *et al.*, 1971).

Statistical analysis

Data obtained for various parameters were subjected to statistical analysis using Analysis of Variance (ANOVA) as described by Snedecor and Cochran (1989). Differences among treatment means were compared using Duncan's Multiple Range Test (DMRT) as modified by Kramer (1956). Statistical significance was declared at the 5% probability level ($P < 0.05$).

RESULTS AND DISCUSSION

Proximate composition

The red seaweed evaluated in this study (Table 1) contained moderate crude protein (11.87%), appreciable crude fibre (6.41%), very low crude fat (0.72%), high ash (47.20%), and modest gross energy (2347.10 kcal/kg), a profile typical of red seaweeds. The protein level supports its role as a functional rather than a primary protein ingredient, while fibre-rich polysaccharides may confer prebiotic effects. The exceptionally high ash content reflects substantial mineral accumulation, which may benefit metabolism and immunity but requires careful dietary formulation. Collectively, the low lipid and energy values indicate that red seaweed is best utilized at moderate inclusion levels to exploit its bioactive and mineral advantages without compromising diet quality.

These findings are consistent with previous reports. Nagaraj *et al.* (2019) documented 14.25% protein in *Gracilaria edulis* and 55.45% ash in *G. corticata*, while Rasyid *et al.* (2019) reported 10.86% protein in *G. gracilis*. Similar protein and ash levels were observed in *Gracilaria sp.* by Al-Khalaifah and Uddin (2022) and in *G. corticata* by Vijayalingam and Rajesh (2023). In contrast, Balasubramanian *et al.* (2021) reported higher protein and lipid but markedly lower ash in *Halymenia palmata*, whereas Narasimman and Murugaiyan (2012) and Norziah and Ching (2000) reported lower protein and ash in other *Gracilaria* species. Together, these comparisons highlight pronounced species

and environment-dependent variability in red seaweed composition.

Mineral composition

The mineral profile (Table 1) confirms red seaweed as a mineral-dense feed ingredient, with calcium (2.47%), phosphorus (0.33%), and iron (0.53%) supporting skeletal development, metabolic functions, oxygen transport, and immunity. The notably high manganese level (601.24 ppm) reflects strong mineral-accumulating capacity and may contribute to enzyme activity, antioxidant defence, and bone formation. Zinc (113.28 ppm) and copper (19.96 ppm), present at nutritionally relevant levels, are essential for growth and immune competence.

These results align with Abudabos *et al.* (2013), who reported high concentrations of calcium, iron, manganese, and zinc in *Ulva lactuca*, and with Norziah and Ching (2000) and Rasyid *et al.* (2019), who documented substantial mineral content in *G. changii* and *G. gracilis*, respectively. Cañedo-Castro *et al.* (2019) similarly observed high mineral accumulation in *Ulva rigida*. Conversely, Balasubramanian *et al.* (2021) reported lower calcium and iron in *H. palmata*, although zinc levels were comparable. The exceptionally high manganese observed in the present study may reflect site-specific environmental factors or species-specific hyperaccumulation, emphasizing the importance of region-specific characterization of seaweeds intended for animal feeding.

Fatty acid profile

The fatty acid composition (Table 1) was dominated by palmitic (26.72%), oleic (31.16%), and linoleic acids (21.56%), with minor contributions from myristic, stearic, and linolenic acids. This profile indicates a lipid fraction primarily composed of saturated and monounsaturated fatty acids, with limited long-chain omega-3 fatty acids.

The predominance of palmitic and oleic acids agrees with Rasyid *et al.* (2019), who identified these as major fatty acids in *G. gracilis*, confirming conserved lipid patterns among red seaweeds. In contrast, Norziah and Ching (2000) reported markedly different profiles in *G. changii*, characterized by high proportions of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) and a predominance of unsaturated fatty acids. The absence of EPA and DHA in the present study underscores pronounced inter-species and environmental variability in lipid metabolism among red seaweeds.

Haematological parameters

Haematological indices, including haemoglobin (Hb), total erythrocyte count (RBC), and total leucocyte count (WBC), did not differ significantly ($P > 0.05$) among treatments (Table 2), indicating that dietary inclusion of red seaweed up to 4% maintained normal blood profiles without haematological stress or toxicity. Stable RBC and Hb values suggest unimpaired erythropoiesis, oxygen-carrying capacity, and metabolic status, while unchanged WBC counts indicate preserved immune homeostasis. These findings demonstrate that red seaweed can be incorporated into

broiler diets without compromising systemic health.

The present results agree with Matshogo *et al.* (2020), who reported no changes in WBC counts in broilers fed green seaweed (2.0–3.5%), and with Balasubramanian *et al.* (2021), who observed no effects of red seaweed (*H. palmata*) on blood parameters in Ross broilers. Although Balasubramanian *et al.* (2021) also reported linear increases in WBC with higher inclusion levels, suggesting potential immunostimulation, the absence of such a response here may reflect differences in seaweed species, inclusion rate, environmental conditions, or bird genotype.

Serum biochemical parameters

Serum biochemical indices showed no significant ($P > 0.05$) differences among treatments for total protein, albumin, ALT, AST, cholesterol, and triglycerides (Table 2), indicating that red seaweed inclusion up to 4% did not adversely affect hepatic function, protein metabolism, or lipid homeostasis. Normal ALT and AST activities suggest absence of hepatocellular damage, while stable protein fractions reflect adequate nutritional status. Likewise, unchanged cholesterol and triglycerides confirm balanced lipid metabolism, supporting the metabolic safety of red seaweed at the tested levels.

The present findings are in agreement with several earlier studies. Shokaiyan *et al.* (2019) reported no significant effects on ALT, AST, total protein, albumin, or globulin

in broilers fed fucoidan and probiotic supplements, supporting the concept that seaweed-derived bioactives maintain normal hepatic and protein metabolism. Matshogo *et al.* (2020) also observed no significant changes in total protein and albumin with green seaweed supplementation (2.0–3.5%) in broilers. Likewise, Mohammadigheisar *et al.* (2020) found that plasma albumin levels in Ross broilers were unaffected by dietary inclusion of a seaweed blend at 0.5–2.0%, and Vijayalingam *et al.* (2020) reported no significant alterations in serum albumin and ALT in Aseel chickens fed 3% *Ulva lactuca*. Collectively, these studies, together with the present findings, demonstrate that moderate dietary incorporation of seaweeds does not disrupt biochemical homeostasis in poultry.

In contrast, some studies have reported beneficial metabolic modulation following seaweed supplementation. El-Naga *et al.* (2018) observed significant increases in serum total protein and albumin, along with reductions in total lipids and cholesterol, in broilers receiving *Ascophyllum nodosum* extract, indicating improved protein metabolism and lipid regulation. Similarly, Cañedo-Castro *et al.* (2019) found that *Ulva rigida* significantly reduced serum cholesterol and triglyceride concentrations in broilers, suggesting hypolipidemic effects. Vijayalingam *et al.* (2020) also reported significant increases in total protein and globulin when Aseel chickens were fed a combined supplementation of *Azolla* and *U. lactuca*, and Balasubramanian *et al.* (2021) documented linear increases in serum albumin with graded inclusion of red seaweed (*Halymenia palmata*), reflecting

possible immunomodulatory and protein-sparing effects.

CONCLUSION

The present study demonstrates that red seaweed (*Gracilaria tenuistipitata*) from peninsular India possesses a distinctive nutritional profile characterized by moderate crude protein, high mineral content, and a fatty acid composition dominated by palmitic, oleic, and linoleic acids. Although not a major source of omega-3 fatty acids, its rich mineral and bioactive composition supports its potential use as a functional feed ingredient in poultry nutrition. Importantly, dietary inclusion of red seaweed up to 4% did not adversely affect haematological indices or serum biochemical parameters in commercial broiler chickens, indicating the absence of metabolic stress, hepatic dysfunction, or systemic toxicity. These findings confirm the physiological safety and nutritional suitability of red seaweed at the tested inclusion levels. Collectively, the results suggest that *G. tenuistipitata* can be incorporated into broiler diets as a sustainable, non-conventional feed resource. Further studies are warranted to evaluate its effects on growth performance, gut health, immune modulation, and economic efficiency under diverse production conditions.

ACKNOWLEDGEMENTS

The authors are highly grateful to Tamil Nadu Veterinary and Animal Sciences University, Chennai, India for the necessary facilities provided to carry out the research work.

Table.1. Chemical composition of red seaweed (on % dry matter basis)

| Parameters | Values* |
|---|---------|
| I. Dry matter (%) | 91.37 |
| II. Proximate principles (DM basis) | |
| 1. Moisture (%) | 8.63 |
| 2. Crude Protein (%) | 11.87 |
| 3. Crude Fibre (%) | 6.41 |
| 4. Crude Fat (%) | 0.72 |
| 5. Crude Ash (%) | 47.20 |
| 6. Nitrogen Free Extract (%) | 25.17 |
| II. Minerals (DM basis) | |
| 1. Calcium (%) | 2.47 |
| 2. Phosphorus (%) | 0.33 |
| 3. Salt (as Chloride) (%) | 6.71 |
| 4. Iron (%) | 0.53 |
| 5. Copper (ppm) | 19.96 |
| 6. Manganese (ppm) | 601.24 |
| 7. Zinc (ppm) | 113.28 |
| III. Gross energy (Kcal/kg) (DM basis) | 2347.10 |
| IV. Acid insoluble ash (%) (DM basis) | 20.78 |
| V. Fatty acid profile | |
| Free Fatty Acids (%) | 22.19 |
| Lauric Acid (%) | 0.94 |
| Myristic Acid (%) | 4.15 |
| Palmitic Acid (%) | 26.72 |
| Palmitoleic Acid (%) | 3.49 |
| Stearic Acid (%) | 7.75 |
| Oleic Acid (%) | 31.16 |
| Linoleic Acid (%) | 21.56 |
| Arachidic Acid (%) | 0.31 |
| Linolenic Acid (%) | 1.63 |
| Behenic Acid (%) | 2.28 |

* - Average of two values

Table.2. Effect of inclusion of red seaweed of commercial broiler chicken on blood parameters at 5 weeks of age (n=6)

| Items | Treatments | | | | F value |
|----------------------------------|--------------|--------------|--------------|-------------|---------|
| | T1 (Control) | T2(1%) | T3(2%) | T4(4%) | |
| Haematological parameters | | | | | |
| Hb (g/dl) | 8.93±0.38 | 10.03±0.37 | 10.23±0.32 | 9.70±0.43 | 2.27NS |
| RBC (106/ μ l) | 2.44±0.19 | 2.47±0.07 | 2.55±0.30 | 2.58±0.18 | 0.10NS |
| WBC (103/ μ l) | 24.86±0.19 | 24.34±0.28 | 24.53±0.27 | 24.38±0.27 | 0.85NS |
| Serum biochemical indices | | | | | |
| Total protein (g/dl) | 4.07±0.08 | 3.78±0.14 | 3.97±0.06 | 3.77±0.13 | 1.89NS |
| Albumin (g/dl) | 2.27±0.12 | 2.20±0.19 | 2.30±0.07 | 2.23±0.07 | 0.12NS |
| AST (U/L) | 285.50±14.55 | 259.67±41.39 | 299.17±21.54 | 307.50±7.71 | 0.72NS |
| ALT (U/L) | 35.83±4.81 | 28.50±5.70 | 34.17±5.07 | 47.50±6.00 | 2.17NS |
| Triglycerides (mg/dl) | 55.33±7.06 | 45.17±29.64 | 55.33±40.29 | 47.50±28.15 | 0.63NS |
| Cholesterol (mg/dl) | 141.50±6.98 | 129.83±15.14 | 139.33±3.89 | 142.83±7.26 | 0.40NS |

NS-Not significant

**Fig.1. Fresh red seaweed****Fig.2. Dried red seaweed**



Fig.3. Powdered red seaweed

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest regarding the publication of this article.

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