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The effects of microalgae (*Tetradesmus obliquus*, *Spirulina platensis* and *Chlorella vulgaris*) on the nutritional profile of broiler meat

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Abstract. This study investigated the effect of dietary supplementation with three edible microalgae species (*Tetradesmus obliquus*, *Spirulina platensis*, and *Chlorella vulgaris*) on the nutritional profile of broiler chicken meat. Ross 308 broilers were fed a standard diet enriched with 0.5% of one of the microalgae species over a 43-day fattening period. Meat samples from the breast and thigh muscles were collected on days 35 and 43 to analyse its nutritional profile. Although no statistically significant differences were observed in growth performance, several biologically differences became apparent. *C. vulgaris* administration was associated with an initial increase in thigh meat fat content on day 35, followed by a notable reduction by day 43, and led to higher protein and polyunsaturated fatty acid levels. However, it resulted in the highest omega-6/omega-3 ratio among the groups. *S. platensis* contributed to a more favorable fatty acid profile, with the lowest omega-6/omega-3 ratio and increased protein levels, particularly in breast meat. *T. obliquus* supplementation produced leaner meat, improved vitamin B_{12} content, and maintained a more balanced omega-6 to omega-3 ratio by day 43, suggesting a cumulative benefit with prolonged feeding. These findings indicate that each microalga has species-specific effects on broiler meat quality, supporting their potential use as sustainable functional feed ingredients tailored to specific nutritional goals in poultry production.

Key words: broiler, chickens, edible microalgae, *Spirulina platensis*, *Chlorella vulgaris*, *Tetradesmus obliquus*, meat quality, fatty acids.

INTRODUCTION

Poultry meat is a globally important source of high-quality protein due to its affordability, safety and short production cycle. (Wahyono et al., 2018; Biesek et al., 2020). However, rising costs of conventional feed ingredients, such as soybean meal and cereals, coupled with sustainability concerns, have driven interest in alternative feed sources. (Babatunde et al., 2021). Microalgae are considered a promising alternative because they contain proteins, polyunsaturated fatty acids (PUFAs), vitamins, minerals and pigments, which could enhance both animal performance and the nutritional quality of meat (Abdel-Wareth et al., 2024). Microalgae are distinguished from land plants by their high content of plastid pigments and diverse carotenoids, and they also synthesize a wide variety of bioactive compounds such as polysaccharides and polyunsaturated fatty acids (Bazarnova et al., 2020; Šefcova et al., 2021). The quantitative and qualitative composition of the intestinal microbiota in broiler chickens can be altered by stressors in industrial poultry farming (Eglite et al., 2024). Gut microbiota plays a key role in the health, growth and performance of poultry. While probiotics are well known to enhance these parameters, other feed additives, such as microalgae, also offer beneficial effects (Akinyemi et al., 2020; Šefcová et al., 2020; Šefcová et al., 2021). Positive outcomes also have been achieved through the administration of algae, regarding animal performance and immunity. (El-Bahr, et al., 2020; Liu, et al., 2021). A key advantage of algae supplementation is its ability to modulate the poultry gut microbiome by supporting beneficial bacteria and microbial diversity. This improves digestion and nutrient uptake (Abdel-Wareth et al., 2024). Studies have demonstrated that microalgae supplementation can enhance growth performance and meat quality in various livestock species, including poultry, although these effects largely depend on the algal species and inclusion level in the diet (Madeira et al., 2017). Microalgae are microscopic photosynthetic organisms that are found in both marine and freshwater environments (Priyadarshani & Rath, 2012). *T. obliquus* contains significant levels of proteins, lipids, and carbohydrates, with a particularly high protein content under mixotrophic conditions; it is rich in essential amino acids (though limited in methionine and cysteine) and has a lipid profile dominated by polyunsaturated fatty acids, including omega-3 and omega-6 (Piasecka et al., 2020). The effects of Tetradesmus supplementation have been tested on both laying hens and pre-starter broilers, with changes observed in egg yolk colour, immune cells and gut microbiota (Rim et al., 2022; Kim et al., 2023). Spirulina (*Arthrospira spp.*) is a blue-green microalga with over 60% easily digestible protein, essential amino acids, vitamins, minerals, and bioactive compounds such as β-carotene, phycocyanin, phenolics, and polysaccharides, which contribute to its antioxidant, anti-inflammatory, hypolipidemic, and immunomodulatory properties, making it a valuable functional feed ingredient (Finamore et al., 2017; Stunda-Zujeva & Berele, 2023; Stunda-Zujeva et al. 2023). The findings also show that spirulina supplementation increases the levels of antioxidants and n-3 polyunsaturated fatty acids (PUFAs) in meat, while concurrently lowering the levels of α-tocopherol and increasing the levels of saturated fatty acids (Costa et al., 2024). *Spirulina* is one of the most widely studied and in-demand species for food and feed applications. Its cultivation potential under natural light conditions in mid-latitude regions such as the Baltic States has been evaluated positively (Stunda-Zujeva et al., 2018). *Chlorella vulgaris* is a nutrient-rich green microalga containing high-quality protein, essential minerals and a broad spectrum of vitamins, including B-complex and fat-soluble vitamins (A, D, E and K). These compounds, along with carotenoids, chlorophyll, and polysaccharides, confer antioxidant, anti-inflammatory, immunomodulatory, and metabolic benefits, making

C. vulgaris a sustainable source of alternative protein with potential applications in human and animal nutrition (Spolaore et al., 2006; Alfaia et al., 2021; Orusmurzaeva et al., 2022; Maurício et al., 2023; Mendes et al., 2024a). Including *C. vulgaris* in the diet enhances the oxidative stability of broiler meat by increasing antioxidant levels, boosting carotenoid deposition and improving the balance between pro- and antioxidants (Mendes et al., 2024b). When included in poultry diets at appropriate low levels, seaweeds can enhance meat and eggs with valuable bioactive compounds (Michalak & Mahrose, 2020). Although previous studies have shown that algae in the diet can affect growth performance and meat composition, there is still limited comparative data on different species of microalgae. Furthermore, the mechanisms by which algal nutrients and percentage of inclusion level enrich meat have not been fully determined. The present study therefore aimed to evaluate the effects of low-dose supplementation with *S. platensis*, *C. vulgaris* and the lesser-known *T. obliquus* on the growth performance, fatty acid composition, and vitamin and mineral content of broiler chicken meat.

MATERIALS AND METHODS

The experimental part of the study was conducted at the Clinical Research Centre, Faculty of Veterinary Medicine, Latvia University of Life Sciences and Technologies, Jelgava, Latvia, from mid-April to the end of May 2024. Permit No. 152/2024 was obtained from the Food and Veterinary Service to conduct the experiment 'The effect of feeding microalgae on the health, productivity and production quality of laying hens and broiler chickens.

Microalgae biomass. Biomass of *Spirulina* and *Chlorella* for poultry experiments was purchased from Buxtrade.de., in Germany. *T. obliquus* (OM02) is an isolate from the Laboratory of Industrial Microbiology and Food Biotechnology (LIMFB) that was obtained from waterbodies in Riga, Latvia. The maintenance of microalgae culture prior to utilisation in a photobioreactor (PBR) and throughout the entirety of the experimental stages was facilitated by employing the standard HP medium procured from VariconAqua (United Kingdom). Inoculations were prepared from stock culture. The initial inoculum was constituted of 10% of the total volume of the starting culture, which was derived from the original stock cultures. The inoculum was gradually increased in scale (250 mL, 500 mL, 1 L, 2 L Erlenmayer flasks and 10 L airlift PBR) until it reached 10 L, which was designated as the final inoculum stage prior to its transfer to an air lift PBR of 140 L capacity. Each stage of inoculum was cultivated for a period of seven days. The first four stages were cultivated statically under a 12:12 hour day: night cycle at room temperature (20 °C). The final stage was cultivated under a 10:10 hour day: night cycle in an airlift PBR. In conclusion, a total of 10 litres of culture medium was utilised to initiate the 140-litre airlift of the PBR. The cultivation of the green microalga *T. obliquus* was performed in a 140 L Phyco-Lift vertical tubular air-lift photobioreactor (Varicon Aqua, UK) under semi-continuous operation. The photoperiod employed was 10 h of light followed by 10 h of darkness, the light intensity of which was gradually increased over time. The light source used was LED panels emitting red and white light. The cultivation process was initiated with an inoculum of 10 L, which possessed an initial optical density (OD540) of approximately 0.2. The aeration process was continuous, with a rate of approximately 5 L min^{-1}. The air was enriched with CO_2 and the pH was maintained within the range of 7.5 to 9.5, with the CO_2 supply being adjusted in real time based on continuous pH monitoring. The mean temperature of the culture medium was sustained within the range of 25–30 °C. The growth medium employed throughout the experiment was HP medium. On a weekly basis, 50% of the reactor volume was harvested for the collection of biomass, followed by replenishment with fresh medium. At each harvest, samples were examined by light microscopy to assess microbial contamination. The biomass was harvested and subsequently concentrated through a combination of elevated pH and centrifugation.

Table 1. Summary of fatty acid profile, nutrient composition, vitamins, minerals, and antioxidant content of microalgae biomass (*C. vulgaris*, *S. platensis*, and *T. obliquus*)

	Parameter	Unit	*C. vulgaris*	*S. platensis*	*T. obliquus*
Fatty acids	Total saturated fatty acids (SAFA)	g/100 g	2.5	2.8	1.9
	Total monounsaturated fatty acids (MUFA)	g/100 g	0.7	0.4	1.9
	Total polyunsaturated fatty acids (PUFA)	g/100 g	6.1	2	3.1
	Sum of trans fatty acids isomers	g/100 g	< 0.1 (0.1 ± 0.1)	< 0.1 (0.1 ± 0.1)	0.7
	Total Omega-3 fatty acids	g/100 g	0.4	< 0.1 (0.1 ± 0.1)	2
	Total Omega-6 fatty acids	g/100 g	4.5	2	1
	Total Omega-9 fatty acids	g/100 g	0.4	0.1	1.7
Amino acids	Aspartic acid	mg/100 g	3,550	4,340	2,260
	Glutamic acid	mg/100 g	6,100	7,840	3,850
	Serine	mg/100 g	1,800	2,940	1,740
	Histidine	mg/100 g	960	1,000	662
	Glycine	mg/100 g	2,520	3,020	2,400
	Arginine	mg/100 g	3,580	3,940	2,140
	Threonine	mg/100 g	1,890	2,950	1,970
	Alanine	mg/100 g	3,670	4,600	3,470
	Proline	mg/100 g	2,110	2,310	2,020
	Tyrosine	mg/100 g	1,340	2,490	1,350
	Valine	mg/100 g	2,450	3,490	2,200
	Methionine	mg/100 g	1,010	1,550	947
	Cysteine	mg/100 g	360	340	318
	Isoleucine	mg/100 g	1,580	3,130	1,440
	Leucine	mg/100 g	3,950	5,270	3,460
	Phenylalanine	mg/100 g	2,170	2,770	2,120
	Lysine	mg/100 g	3,530	2,940	2,050
Vitamins	Vitamin E (α-tocopherol)	mg/100 g	17.3	2.8	8.3
	Vitamin B1 (thiamine)	mg/100 g	0.08	1.15	nd
	Vitamin B2 (ryboflavin)	mg/100 g	3.86	2.19	nd
	Vitamin B3 (niacin)	mg/100 g	22.9	16.2	nd
	Vitamin B5 (pantothenic acid)	µg/100 g	1.23	96.1	nd
	Vitamin B6 (pyridoxine)	mg/100 g	1.52	0.61	0.38
	Vitamin B7 (biotin)	µg/100 g	21.5	1.62	nd
	Vitamin B9 (folic acid)	µg/100 g	1030	108	nd
	Vitamin B12 (cyanocobalamin)	µg/100 g	1.54	168	105

Table 1 (continued)

Minerals and nutrients	Sodium (Na)	mg/100 g	434	322	nd
	Salt as sodium chloride (NaCl)	mg/100 g	1,085	805	nd
	Iron (Fe)	mg/100 g	14,026	75.4	28.3
	Zinc (Zn)	mg/100 g	0.99	1.34	26.2
	Magnesium (Mg)	mg/100 g	258	254	357
	Moisture	g/100 g	2	4.4	nd
	Protein (N*6.25)	g/100 g	56.9	69.6	54.8
	Dietary fiber	g/100 g	10.4	7.8	nd
	Energy value	kcal/100 g	406	372	nd
	Carbohydrates	g/100 g	15.8	6.7	28.5
	Fat	g/100 g	10.5	5.7	9.4
	Ash	g/100 g	4.38	5.78	nd
	Total sugars	g/100 g	0.22	0	nd
	Total polyphenols as gallic acid	mg/100 g	528.03	188.93	nd
	Total Carotenoids	mg/100 g	13.63	15.41	11.45

Symbols such as ‘<’ indicate values below the detection limit of the accredited method; nd – not detected.

Following this, the biomass was washed in distilled water and dried at 45 °C to prevent the degradation of bioactive compounds. Microalgae biomass samples (*C. vulgaris*, *S. platensis*, and *T. obliquus*) were submitted to the Hamilton Laboratory for detailed compositional analysis. The evaluation included quantification of fatty acid profiles, amino acid composition, vitamins, minerals, proximate nutritional content, and antioxidant compounds such as polyphenols and carotenoids.

Experimental design and animal management. A total of 140 unsexed Ross 308 broiler chicks were obtained from a commercial hatchery within two hours of hatching, to minimise environmental exposure and reduce the risk of pathogen contamination strict biosecurity measures were implemented throughout the study, including restricted access to the experimental facility and the use of protective equipment, such as disposable clothing, gloves and footwear. Personnel visited the site twice daily for feeding purposes to monitor the health and welfare of the birds. During the study, the broiler chickens were not given any vaccinations. After being brought to the Clinical Research Centre, the chickens were weighed and then randomly divided into four study groups (control (BK), *S. platensis* (BS), *C. vulgaris* (BC) and *T. obliquus* (BT)), each containing day-old Ros 308 broiler chickens (n = 35, total n = 140). The broilers were reared for 43 days under controlled conditions abided by the Ross Broiler Management Guide (Aviagen Ross Broiler Guide, 2024). For body weight measurements, animals were weighed at day 1; 8; 14; 22; 29; 35 and 43 and the averaged values were used for analyses. To examine the fatty acid, vitamin and mineral profile of the breast and thigh meat, we randomly selected and euthanised five chicks from each group on day 35 and 7 chicks from each group on day 43. The broiler chickens were stunned with a blowgun and euthanised by bleeding. All chickens were fed basal diets free of probiotics, antibiotics, or anticoccidiostats. All four study groups were given the same amount of food on the feeding tables twice a day. The poultry-based diet for all groups was specifically formulated for Ross 308 broiler chickens aged 1–35 days and above and consisted of three phases. Starter (from day 0 to day 18: 14.5 MJ kg^{-1}), Grower (from day 18 to day 35: 14.46 MJ kg^{-1}) and Finisher (from day 35 until the end of the study on day 43: 14.39 MJ kg^{-1}). The primary protein sources in the feed were wheat grains, soybean meal (GMO), corn, and rapeseed cake. The chemical composition of the feed is detailed in Table 2.

Table 2. Feed components and proximate composition of starter (days 1–18), grower (days 18–35) and finisher (above 35th day) diets for the control and experimental groups, granulated (crushed)

Starter	Grower	Finisher
Wheat	Wheat	Wheat
Soybean meal (GMO)	Soybean meal (GMO)	Soybean meal (GMO)
Corn	Corn	Corn
Rapeseed cake	Rapeseed cake	Rapeseed cake
Rapeseed oil	Rapeseed oil	Rapeseed oil
Ca carbonate	Ca carbonate	Ca carbonate
Monocalcium phosphate	Monocalcium phosphate	Monocalcium phosphate
Na chloride	Na chloride	Na chloride

Nutritional additives	Starter	Grower	Finisher
Vitamins (3a):			
A (3a672a), IU kg^{-1}	14,853.96	14,853.96	14,853.96
D_3 (3a671), IU kg^{-1}	2,983.86	2,983.86	2,983.86
E (3a700, all-rac-alfatoko.acet), mg kg^{-1}	41.5	42.62	43.16
B_1 (thiamine 3a821), mg kg^{-1}	6.25	6.11	6.03
B_2 (riboflavin 3a826), mg kg^{-1}	7.85	7.72	7.63
B_5 (Ca-d-pantothenic acid 3a841), mg kg^{-1}	46.87	46.33	46.07
B_4 (choline chloride 3a890), mg kg^{-1}	1,599.07	1,475.36	1,397.6
B_3 (niacin 3a315), mg kg^{-1}	44.37	44.01	44.3
B_6 (pyridoxine 3a831), mg kg^{-1}	8.8	8.59	8.42
B_{12} (cyanocobalamin 3a835), mcg	23.74	23.74	23.74
K_3 (methadone 3a711), mg	2.3	2.37	2.37
B_7 (H-biotin 3a880), mg	0.05	0.05	0.05
B_c (folic acid 3a316), mcg	0.59	0.59	0.59
Micronutrients (3b):			
Iron sulfate (3b103), (Fe), mg kg^{-1}	126.04	116.32	111.71
Copper sulfate (3b405), (Cu) mg kg^{-1}	18.77	18.1	17.67
Manganese oxide (3b502), (Mn) mg kg^{-1}	112.26	110.14	109.79
Zinc oxide (3b603), (Zn) mg kg^{-1}	89.55	87.91	86.94
Sodium selenite (3b801), (Sn) mg kg^{-1}	0.32	0.32	0.31
Calcium iodate (3b202), (J) mg kg^{-1}	1.22	1.22	1.22
Cobalt carbonate (3b304), (Co) mg kg^{-1}	0.67	0.66	0.66
Analytical components:			
ME (metabolizable energy), MJ	14.5	14.46	14.39
crude protein %	22.79	20.31	19.1
Crude fat %	4.54	4.73	4.79
Fiber %	4.89	5.38	5.5
Ash %	4.76	4.48	4.28
Starch %	36.31	39.78	41.82
Calcium (Ca) %	0.99	0.90	0.93
Phosphorus (P) %	0.54	0.52	0.51
Sodium (Na) %	0.15	0.15	0.14
Methionine %	0.38	0.37	0.33
Lysine %	1.32	1.6	1.06

All experimental diets were provided with the same dose of biomass (5 g kg^{-1}) of the respective microalgal supplement, which was added to the BS, BC and BT groups. All groups were housed in four identical bio-chambers (total floor area of the room 20.76 m^2, divided in four equal parts - 5.19 m^2), equipped with a comprehensive microclimate control system. This system regulated temperature, humidity and air supply, as well as controlling the composition of incoming and outgoing air and light cycles. Each bio-chamber was also fitted with video surveillance equipment to enable continuous monitoring. The chambers were equipped with stationary automatic watering systems and movable feeding tables which were adjusted according to the age of the chicks prior to placement. Clean wood shavings covered the flooring of each chamber to maintain hygiene and ensure animal comfort. The broiler chickens were raised until the 43rd day of life. The lighting and temperature regime was established based on prior research and Ross 308 breeder guidelines (Aviagen Ross broiler guide, 2024). During the first week of the experiment, the ambient temperature in the chambers for all groups was kept between 27 and 30 °C. As the birds grew, the temperature was gradually reduced until it reached 20–22 °C by the end of the study. On the first day, the light/dark cycle consisted of 23 hours of light and 1 hour of darkness (23/1). Subsequently, the darkness period was progressively increased to six hours, with a regime of 18 hours of light and six hours of darkness (18/6) from day eight to 26th. During the final week, the dark period was gradually shortened to create a light/dark cycle of 20 hours of light and 4 hours of darkness (20/4). The switch from light to dark occurred through the red light.

Body weight. All birds were weighed, and the averaged values were used for analyses. This was carried out on day 1; 8; 14; 22; 29; 35 and 43.

Meat quality parameters. Samples of breast and thigh meat were analysed on days 35 and 43. Five samples were taken from each group on day 35, and seven samples from each group on day 43. The pooled meat samples were prepared in accordance with laboratory requirements for minimum sample volume providing a representative overview of the overall meat composition. These samples were examined for their fatty acid, vitamin and mineral content. Only vitamins and minerals were determined on day 43. The samples were frozen at -22 °C for later examination. To determine the nutritional composition of chicken breast and thigh meat, the samples were sent to the J.S. Hamilton Poland Sp. z o.o. laboratory for testing. The results of the analysis of the fatty acid profile of the chicken breast and thigh meat samples were expressed in g/100 g of fresh weight. To detect minerals in the meat samples, they were sent to Eurofins Labtarna Lietuva, JSC laboratory. Mineral concentrations (Fe, Se, K, Mg) were expressed in mg kg^{-1}, vitamin E in mg kg^{-1}, and vitamin B_{12} in µg/100 g, all calculated on a fresh weight basis.

Statistical analyses. Generalised linear models (GLMs) were used to assess the effects of different dietary treatments (Control, *Tetradesmus, Spirulina*, and *Chlorella*) on chicken weight, with chicken age and initial weight included as covariates. The chi-squared test was applied to evaluate differences in categorical meat quality parameters. Descriptive statistics (mean, standard deviation, minimum, and maximum) were calculated. Statistical data analysis was performed with Jamovi (v.2.5). The results were considered statistically significant when the *p value* was < 0.05.

RESULTS AND DISCUSSION

Effects of diet on body weight. After adjusting for chicken age, no statistically significant differences in body weight were observed between treatment groups ($p = 0.777$; see Table 3). All groups exhibited consistent weight gain from day 1 to day 43, indicating normal growth trajectories regardless of dietary treatment. By day 43, the BT group reached the highest individual weight (4,029 g), followed by the BC group (3,903 g).

Table 3. Average body weight (g) of broiler chickens by treatment group from day 1 to day 43

		Weight			
Age	Group	Average	SD	Min	Max
Day 1	Control (BK)	61.49	1.43	59.90	63.40
	Tetradesmus obliquus (BT)	60.55	2.03	57.30	63.00
	Spirulina platensis (BS)	61.72	1.27	59.80	63.00
	Chlorella vulgaris (BC)	60.67	1.07	59.00	61.60
Day 8	Control (BK)	181.09	6.03	169.80	189.80
	Tetradesmus obliquus (BT)	176.57	7.86	166.80	191.20
	Spirulina platensis (BS)	180.17	4.99	172.40	187.80
	Chlorella vulgaris (BC)	170.56	6.46	160.20	180.60
Day 14	Control (BK)	454.07	26.86	421.60	513.30
	Tetradesmus obliquus (BT)	431.29	30.64	375.30	483.00
	Spirulina platensis (BS)	437.22	25.06	396.60	467.30
	Chlorella vulgaris (BC)	429.13	34.50	377.60	508.60
Day 22	Control (BK)	895.68	28.29	829.00	933.50
	Tetradesmus obliquus (BT)	882.16	74.97	761.50	1,068.00
	Spirulina platensis (BS)	881.46	79.89	726.50	1,009.00
	Chlorella vulgaris (BC)	884.32	82.17	731.50	1,084.00
Day 29	Control (BK)	1,556.92	184.91	1,298.00	2,028.00
	Tetradesmus obliquus (BT)	1,514.84	167.01	1,179.00	1,889.00
	Spirulina platensis (BS)	1,473.36	143.81	1,241.00	1,760.00
	Chlorella vulgaris (BC)	1,521.88	173.53	1,,245.00	1,881.00
Day 35	Control (BK)	2,225.25	289.01	1770.00	2,890.00
	Tetradesmus obliquus (BT)	2,034.36	319.11	1,203.00	2,624.00
	Spirulina platensis (BS)	2,185.20	197.94	1,936.00	2,563.00
	Chlorella vulgaris (BC)	2,145.56	242.37	1,690.00	2,607.00
Day 43	Control (BK)	3,061.00	468.75	2,122.00	3,903.00
	Tetradesmus obliquus (BT)	3,032.89	462.71	1,676.00	4,029.00
	Spirulina platensis (BS)	3,046.00	276.40	2,509.00	3,566.00
	Chlorella vulgaris (BC)	2,964.85	306.37	2,403.00	3,506.00

Includes mean, standard deviation (SD), minimum and maximum weight values for each group at different time points.

Although the differences were not statistically significant the BS and BK groups consistently showed higher average body weights compared to the BC and BT groups throughout the rearing period. Standard deviation values increased across all groups over time particularly after day 22 suggesting greater variability in growth as the birds aged.

Meat quality parameters. It should be noted that pooled meat samples were used for the analysis of fatty acids, vitamins and minerals. While this approach provided sufficient material for accurate laboratory testing and offered a representative overview of the group's composition, it may have reduced statistical power and limited the ability to detect differences between groups. This should be considered a methodological limitation of the study. Although no statistically significant differences in meat quality parameters were observed between the treatment groups ($p > 0.05$), various nutritional patterns were identified that may have biological or practical significance, see Table 4.

By day 35, the *Spirulina* (BS) group exhibited the most favorable fatty acid profile (MUFA and PUFA at 0.9 and 05 g/100 g), with the lowest omega-6 to omega-3 ratio (4.3), which matched the lower PUFA level of the Spirulina biomass (2.0 g/100 g), see Table 1. Meanwhile, the *Chlorella* (BC) group had the highest total fat (2.8 g/100 g) and energy content (115 kcal/100 g) including the highest levels of monounsaturated (1.3 g/100 g) and polyunsaturated (0.7 g/100 g) fatty acids, as well as omega-6 (0.6 g/100 g) and omega-9 (1.1 g/100 g) fatty acids. This corresponded with the algae biomass profile of *Chlorella*, which had highest PUFA content (6.1 g/100 g) and omega-6 (4.5 g/100 g) see Table 1. The *Tetradesmus obliquus* group had a lower breast fat content (2.4 g/100 g) with MUFA and PUFA levels at 1.1 and 0.6 g/100 g, respectively. The algae biomass for *Tetradesmus* also contained moderate PUFA (3.1 g/100 g) and the lowest SAFA content (1.9/100 g). Similar trends persisted up to day 43. The BC group exhibited the highest levels of MUFA (2.1 g/100 g), PUFA1.0 g/100 g), omega-9 (1.9 g/100 g) (and total fat (4.5 g/100 g), as well as the highest calorie content (121 kcal/100 g). This mirrored *Chlorella's* high PUFA and omega-6 values in the feed. In contrast, the *Tetradesmus* (BT) and *Spirulina* (BS) groups exhibited leaner profiles, with lower fat content (1.7 and 1.9 g/100 g, respectively) and energy value (106 and

Table 4. Fatty acid profile, proximate composition, energy value, and micronutrient content of broiler breast meat at days 35 and 43

	Day 35 Breast meat			
Fatty acids g/100 g	BK	BT	BS	BC
Total (SAFA)	0.5	0.7	0.6	0.8
Total (MUFA)	0.9	1.1	0.9	1.3
Total (PUFA)	0.5	0.6	0.5	0.7
Total Omega-3	0.1	0.1	0.1	0.1
Total Omega-6	0.4	0.5	0.4	0.6
Total Omega-9	0.8	0.9	0.8	1.1
Water	75.7	75.8	75.4	75.1
Total ash	1.29	1.24	1.29	1.17
kcal/100 g	106	111	106	115
Protein	21.9	22.3	22	22.4
Fat	2	2.4	2	2.8
	Day 43 Breast meat			
Fatty acids g/100 g	BK	BT	BS	BC
Total (SAFA)	0.8	0.5	0.5	1.2
Total (MUFA)	1.2	0.7	0.8	2.1
Total (PUFA)	0.6	0.4	0.4	1
Total Omega-3	0.1	0.1	0.1	0.1
Total Omega-6	0.5	0.4	0.4	0.9
Total Omega-9	1.1	0.6	0.7	1.9
Water	74.3	75.8	76.1	74.6
Total ash	1.14	1.28	1.15	1.04
kcal/100 g	116	106	104	121
Protein	23	22.6	21.8	20.2
Fat	2.7	1.7	1.9	4.5
Minerals and vitamins				
Fe mg kg^{-1}	4.1	3.8	4.2	3.5
Se mg kg^{-1}	0.33	0.28	0.27	0.26
K mg kg^{-1}	3,120	3,250	3,060	2,990
Mg mg kg^{-1}	220	212	191	206
E vit. mg kg^{-1}	0.391	0.375	0.472	0.855
B12 vit. µg/100 g	0.325	0.412	0.357	0.350

Includes Saturated Fatty Acids (SAFA), monounsaturated fatty acids (MUFA - omega-9), polyunsaturated fatty acids (PUFA - omega-3, -6), fat, protein, water, ash, and energy values.

Includes iron, selenium, potassium, magnesium, vitamin E, and vitamin B12 concentrations.

104 kcal/100 g, respectively), corresponding with its lower fat, PUFA and MUFA levels in feed. Additionally, the *Tetradesmus* group showed the highest vitamin B_{12} (0.412 µg/100 g) and potassium levels (3,250 mg kg^{-1}), which were also among the highest in the algal biomass. The control group had the highest protein content (23 g/100 g), followed by the BT group (22.6 g/100 g). In terms of mineral and vitamin content on day 43, all groups had similar levels of iron, selenium, and magnesium. However, the *Chlorella* group had the highest concentration of vitamin E (0.855 mg kg^{-1}), almost double that of the control group.

Although no statistically significant differences in thigh meat quality parameters were observed between the dietary treatment groups ($p > 0.05$), several noteworthy nutritional trends emerged in the context of broiler meat enrichment and functional feed development. See Table 5. On day 35, the *C. vulgaris* (BC) group had the highest fat content (7.7 g/100 g), total monounsaturated fatty acids (MUFA; 3.7 g/100 g), PUFA (1.8 g/100 g), and omega-6 (1.6 g/100 g), aligning with *Chlorella's* high PUFA and omega-6 content in the algae composition. The *Spirulina* group showed a lower fat content (6.2 g/100 g) with MUFA at 3.0 g/100 g and PUFA at 1.5 g/100 g. The *T. obliquus* (BT) group had slightly lower fat content (6.8 g/100 g), MUFA (3.2 g/100 g), and PUFA (1.7 g/100 g). Across all groups, omega-3 values in meat remained at 0.2 g/100 g, matching the modest omega-3 presence in the algal biomass. *S. platensis* group had the highest protein content (19.4 g/100 g), which also is the highest in algal biomass. By day 43, the trends in fatty acids had changed. The *Spirulina* (BS) group showed the highest concentrations of MUFA (3.5 g/100 g), PUFA (1.7 g/100 g), omega-6

Table 5. Fatty acid profile, proximate composition, and micronutrient content of broiler thigh meat at days 35 and 43

	Day 35 thigh meat			
Fatty acids g/100 g	BK	BT	BS	BC
Total (SAFA)	1.8	1.8	1.6	2.1
Total (MUFA)	3.7	3.2	3	3.7
Total (PUFA)	1.8	1.7	1.5	1.8
Total Omega-3	0.2	0.2	0.2	0.2
Total Omega-6	1.5	1.4	1.3	1.6
Total Omega-9	3.4	2.9	2.7	3.4
Water	73.5	73.4	74.1	73.1
Total ash	1.11	1.21	1.22	1.04
kcal/100 g	141	136	133	145
Protein	18.6	18.7	19.4	18.9
Fat	7.4	6.8	6.2	7.7
	Day 43 thigh meat			
Fatty acids g/100 g	BK	BT	BS	BC
Total (SAFA)	1.5	1.5	2	1
Total (MUFA)	2.6	2.7	3.5	1.6
Total (PUFA)	1.5	1.3	1.7	0.9
Total Omega-3	0.2	0.2	0.2	0.1
Total Omega-6	1.2	1.1	1.5	0.8
Total Omega-9	2.3	2.4	3.1	1.5
Water	74.9	74.7	74.3	75
Total ash	1.09	1.03	0.97	1.14
kcal/100 g	129	128	142	117
Protein	19.3	19.4	19.1	21.1
Fat	5.8	5.6	7.3	3.6
Minerals and vitamins				
Fe mg kg^{-1}	6.1	6.9	6.3	6.5
Se mg kg^{-1}	0.31	0.3	0.3	0.28
K mg kg^{-1}	2,860	2,850	2,820	2,980
Mg mg kg^{-1}	191	190	184	195
E vit. mg kg^{-1}	0.220	0.221	0.141	0.152
B_{12} vit. µg/100 g	0.368	0.414	0.383	0.390

Includes Saturated Fatty Acids (SAFA), monounsaturated fatty acids (MUFA - omega-9), polyunsaturated fatty acids (PUFA - omega-3, -6), fat, protein, water, ash, and energy values.

Includes iron, selenium, potassium, magnesium, vitamin E, and vitamin B_{12} concentrations.

(1.5 g/100 g), and omega-9 (3.1 g/100 g) in thigh meat, corresponding to the algae's composition, where *Spirulina* had moderate levels of omega-6 and higher antioxidant content. Meanwhile, the *Chlorella* (BC) group exhibited the lowest total fat content (3.6 g/100 g) and a notably higher protein content (21.1 g/100 g). MUFA and PUFA levels were 1.6 g/100 g and 0.9 g/100 g, respectively, which reflected *Chlorella's* higher PUFAS and protein content in the biomass. Water and ash content remained relatively constant across all groups. In terms of micronutrients, the highest iron (Fe) content was in BT group (6.9 mg kg^{-1}) and the concentrations of selenium were very similar between the groups (0.28–0.31 mg kg^{-1}) in thigh meat. Potassium and magnesium levels were relatively uniform, with the *Chlorella* group showing a slightly higher potassium (K, 2,980 mg kg^{-1}) and magnesium (Mg, 195 mg kg^{-1}) content. Regarding vitamin concentrations, *Tetradesmus* (BT) group had the highest levels of vitamin B_{12} (0.414 μ g/100 g), and vitamin E content (0.221 mg kg^{-1}) and moderate thigh fat (5.6 g/100 g), again aligning with the nutrient composition of its algal supplement. Despite having a lower fat content, BC meat maintained a relatively high B_{12} level (0.390 μg/100 g).

This study examined how dietary supplementation with three edible microalgae species – *T. obliquus*, *S. platensis* and *C. vulgaris* – affects the growth and meat quality of broiler chickens. Although no statistically significant differences in body weight or meat quality were observed, several biologically relevant trends emerged that support existing research and shed insight on the functional properties of these microalgae. While body weight did not differ significantly between groups, the numerically highest final weight was observed in the *Tetradesmus* group (4,029 g) see Table 3, followed closely by the control and *Spirulina* groups. This aligns with previous findings indicating that microalgal supplementation can support growth performance without negatively impacting overall productivity (El-Bahr et al., 2020; Šefcová et al., 2021). The *Spirulina* group showed a favorable omega-6 to omega-3 ratio (4:1) and increased PUFA levels in both breast and thigh meat. Based on findings from (Simopoulos et al., 2002) and (World Health organization, 2003), a lower dietary omega-6 to omega-3 ratio is associated with improved health outcomes. While our results showed that *Spirulina* supplementation improved the fatty acid profile of broiler meat by increasing n-3 polyunsaturated fatty acids (PUFAs) and lowering the n-6/n-3 ratio, other studies have reported no significant effects on fatty acid methyl esters in breast meat (Yalçınkaya et al., 2025). In contrast, Spínola et al. (2024) observed enhanced fatty acid profiles and antioxidant capacity, with higher n-3 PUFA deposition and reduced lipid oxidation, likely due to Spirulina's β-carotene and other carotenoids, which protect cells from oxidative stress by preventing oxygen-induced damage (Tinkler, 1994). However, these effects were not strictly linear at higher inclusion levels, suggesting that dosage and dietary context play an important role in determining the impact of Spirulina on meat quality. Supplementation with *Chlorella* vulgaris was associated with increased concentrations of monounsaturated and polyunsaturated fatty acids, along with higher total fat and protein content in broiler meat. Boskovic Cabrol et al. (2022) reported that replacing soybean meal with *C. vulgaris* improved protein digestibility and amino acid availability, further supporting the role of *Chlorella* in enhancing meat protein deposition. In our study, breast and thigh meat from the *C. vulgaris* group showed higher fat content compared with the other treatments. This differs from previous findings where *C. vulgaris* supplementation reduced or had little effect on meat lipid levels (Alfaia et al., 2021; Varzaru et al., 2024). The discrepancy may be related to differences in diet formulation, algal inclusion level and composition, or broiler strain.

T. obliquus demonstrated the most beneficial fatty acid profile, achieving omega-6/omega-3 ratios close to 4:1 or lower, especially on day 43. This improvement can be attributed to its ability to synthesise omega-3 fatty acids, including ALA, and possibly DHA under mixotrophic cultivation conditions (Piasecka et al., 2020). The trend toward improved fatty acid ratios over time suggests a cumulative effect, supporting previous research indicating that longer feeding durations promote omega-3 deposition in tissues, and decrease the n-6 to n-3 ratio (Shahid et al., 2019). Notably, the omega-6/omega-3 ratio in the *Tetradesmus* and *Spirulina* fed groups approached or fell below 4:1 on day 43, which aligns with the optimal range recommended by (Simopoulos et al., 2002) and the (World Health organization, 2003) for reducing inflammation and lowering chronic disease risk. The variation observed between breast and thigh meat is likely due to differences in lipid metabolism and muscle composition. Thigh meat has a higher fat content and may therefore allow for greater accumulation of dietary fatty acids. However, the distribution patterns were consistent across both tissue types (Crespo & Esteve-García, 2001). This finding supports the systemic availability and metabolic utilisation of algal-derived fatty acids. In terms of micronutrients, the concentrations of iron (Fe), selenium (Se), potassium (K) and magnesium (Mg) in broiler meat were relatively uniform across the different treatment groups, with only minor differences observed. Potassium levels were slightly higher in breast meat from *the Tetradesmus* group and in thigh meat from the *Chlorella* group. This is consistent with the nutrient profile of microalgae, which provide vitamins (A, C, E and B complex) and minerals such as iron, potassium, magnesium and calcium (Becker, 2013). Notably, C. vulgaris is characterised by higher phosphorus content and substantial potassium levels (Tokuşoğlu & Ünal, 2003). Therefore, the modest enrichment of meat potassium plausibly reflects dietary mineral contributions from the algae, at least in part, and aligns with reports that potassium is the predominant mineral in chicken muscle, followed by phosphorus and sodium (Demirbaş, 1999). In addition to differences between species, the mineral composition of algae varies with growth conditions (Santhakumaran et al., 2020), which may further influence mineral deposition in meat. In addition to modulating lipid composition, supplementation with microalgae affected the vitamin profile of broiler meat, particularly vitamin B_{12}. It should be noted that the analytical method used in this study measured the total amount of vitamin B_{12} present and did not distinguish between its active (methylcobalamin and adenosylcobalamin) and inactive (pseudovitamin B_{12}) forms. Future studies using advanced analytical methods are required to validate the specific forms deposited in poultry meat.

Notably, supplementation with *C. vulgaris* resulted in elevated vitamin B_{12} concentrations in both breast and thigh muscles by day 43, despite the low cobalamin content measured in the algal biomass. This is consistent with previous research showing that *Chlorella* contains bioavailable forms of vitamin B_{12}, such as methylcobalamin and adenosylcobalamin (Kittaka-Katsura et al., 2002), which are efficiently absorbed and deposited in animal tissues. By contrast, Spirulina platensis exhibited a high total vitamin B_{12} concentration in the biomass. However, previous studies have shown that most of its corrinoid compounds are pseudovitamin B_{12} analogues with low intrinsic factor binding affinity and no bioactivity in humans (Watanabe et al., 1999; Watanabe et. al., 2014).

Therefore, while *Spirulina* supplementation led to increased total vitamin B_{12} levels in meat, its nutritional value for human consumers remains uncertain. No significant data on the vitamin B_{12} content of *T. obliquus* currently exist in the literature. However, in this study, birds supplemented with *Tetradesmus* also showed elevated B_{12} levels in meat tissues. Whether this reflects true dietary contribution or indirect metabolic effects remains unclear and warrants further investigation. Regarding vitamin E (α-tocopherol), supplementation with Spirulina led to the highest concentrations in thigh meat, which is consistent with its well-documented abundance of tocopherols and carotenoids (Khan et al., 2005). In contrast, *Chlorella* vulgaris resulted in the highest α-tocopherol content in breast meat by day 43, indicating effective tissue deposition and confirming its classification as a rich natural source of fat-soluble vitamins, including α-tocopherol (Del Mondo et al., 2020). Although the biomass of *T. obliquus* used in our study contained a substantial amount of vitamin E (8.3 mg/100 g), this did not result in an increase in α-tocopherol levels in the broiler meat. On day 43, the vitamin E content of the breast meat in the *Tetradesmus* group (0.375 mg kg^{-1}) was slightly lower than in the control group (0.391 mg kg^{-1}), and much lower than in the Chlorella group (0.855 mg kg^{-1}). These results imply that, despite its inherent α-tocopherol content, the bioavailability or metabolic utilisation of vitamin E from *T. obliquus* may be limited. This aligns with previous findings by (Chronopoulou et al., 2019), who demonstrated that the extraction and bioavailability of fat-soluble vitamins from *T. obliquus* can depend on specific processing methods.

CONCLUSION

This study demonstrated that microalgae supplementation alters broiler meat composition in a species-specific manner. *Spirulina* improved the n-6/n-3 ratio and increased n-3 PUFAs, *Chlorella* enhanced protein and vitamin E but also increased fat, and *Tetradesmus* produced leaner meat with higher vitamin B_{12} level and a more balanced fatty acid ratio. These findings highlight the potential of different microalgae as functional feed ingredients to improve poultry meat quality. These results demonstrate that microalgae modulate the nutrient composition of broiler meat in species-specific ways.

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