Original scientific paper
DOI: /10.5513/JCEA01/27.2.5347
Journal of Central European Agriculture, 2026, 27(2), p.341-353

Effects of fermented feed and pasture access on growth performance, meat quality and fatty acid profile in red broiler chickens

Utjecaj fermentirane hrane i pristupa pašnjaku na proizvodne rezultate, kvalitetu mesa i profil masnih kiselina crvenih brojlera

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Received: April 10, 2026; accepted: June 5, 2026

ABSTRACT

This study evaluated the effects of fermented feed and pasture access on growth performance, feed intake, and meat quality in slow-growing red broiler chickens over a 121-day production period. A total of 125 chickens were assigned to three treatments: fermented feed with pasture access (FerVP_pasture), dry feed with pasture access (VP_pasture), and dry feed under indoor conditions (VP). Growth performance was monitored throughout the experimental period, and meat quality was assessed based on proximate composition, oxidative stability, and fatty acid profile. Chickens reared under indoor conditions (VP) achieved the highest final body weight, whereas the FerVP_pasture group showed reduced feed intake with comparable growth performance, indicating improved feed utilization efficiency. Meat quality parameters were significantly influenced by the feeding regime. The VP group exhibited higher fat content and increased lipid oxidation, while pasture-based systems resulted in leaner meat and improved oxidative stability. Fatty acid analysis revealed significant differences in PUFA composition, with the VP group showing higher n-3 PUFA content, while the FerVP_pasture group achieved a more balanced n-6/n-3 PUFA ratio. All treatments maintained values below the recommended threshold for human health. The results suggest that the combination of fermented feed and pasture access represents a promising strategy for improving feed efficiency and meat quality in alternative poultry production systems, while maintaining acceptable growth performance in male and female animals.

Keywords: broilers, fermented feed, pasture, meat quality, fatty acid, feed

SAŽETAK

U ovom radu ispitivan je utjecaj fermentirane hrane i pristupa pašnjaku na proizvodne rezultate, unos hrane i kvalitetu mesa spororastućih crvenih brojlera tijekom 121-dnevnog tova. U pokus je uključeno ukupno 125 pilića raspoređenih u tri skupine: fermentirana hrana s pristupom pašnjaku (FerVP_pasture), suha hrana s pristupom pašnjaku (VP_pasture) te suha hrana u zatvorenom sustavu (VP). Tijekom pokusa praćen je rast, dok je kvaliteta mesa procijenjena analizom kemijskog sastava, oksidativne stabilnosti i profila masnih kiselina. Pilići uzgajani u zatvorenom sustavu (VP) postigli su najveću završnu tjelesnu masu, dok je skupina FerVP_pasture ostvarila manji unos hrane uz usporediv rast, što upućuje na bolju učinkovitost iskorištenja hrane. Režim hranidbe značajno je utjecao na kvalitetu mesa. Skupina VP imala je veći udio masti i višu razinu lipidne oksidacije, dok su sustavi s pristupom pašnjaku rezultirali mesom s manjim udjelom masnoće i boljom oksidativnom stabilnošću. Analiza masnih kiselina pokazala je značajne razlike u udjelu višestruko nezasićenih masnih kiselina (PUFA), pri čemu je skupina VP imala viši udio n-3 PUFA, dok je skupina FerVP_pasture ostvarila povoljniji omjer n-6/n-3 PUFA. Sve skupine imale su vrijednosti povoljne za ljudsko zdravlje. Dobiveni rezultati ukazuju da kombinacija fermentirane hrane i pristupa pašnjaku predstavlja obećavajuću strategiju za poboljšanje učinkovitosti hranidbe i kvalitete mesa u alternativnim sustavima proizvodnje peradi, te pokazuje prihvatljive rezultate rasta i kod mužjaka i kod ženki.

Ključne riječi: brojleri, fermentirana hrana, pašnjak, kvaliteta mesa, masne kiseline, hranidba

INTRODUCTION

Increasing consumer awareness has led to a shift in preferences toward products perceived as wholesome, with greater emphasis on production conditions, animal welfare, and product origin (Aertsens et al., 2009; Verbeke et al., 2010; Mijatović et al., 2012; Teixeira and Rodrigues, 2021). In this context, alternative production systems, including outdoor and small-scale poultry farming, are gaining importance. These systems may influence the nutritional quality of meat, particularly its fatty acid composition, including the omega-6 to omega-3 ratio and the content of mono- and polyunsaturated fatty acids (Simopoulos, 2002; Makiš et al., 2024). Outdoor production systems are the most difficult form of poultry production because such production is always associated with increased environmental variability and potential health risks for poultry, including pathogen exposure, which may affect animal performance and product quality (Campbell et al., 2025; Saccavini et al., 2026). In such systems, the feeding strategy is a key factor influencing growth performance and meat quality. Previous studies have demonstrated that housing conditions and diet composition significantly affect parameters such as lipid composition, oxidative stability, and physicochemical properties of meat, although the magnitude of these effects depends on genotype and management conditions (Dal Bosco et al., 2010; Campbell et al., 2025). Research has also evaluated the effects of housing systems and feeding regimes separately, demonstrating that outdoor rearing influences physical activity and fatty acid profile, while diet composition directly affects lipid composition and oxidative stability of meat (Lopez-Ferrer et al., 1999; Simopoulos, 2002; Castellini et al., 2002; Dal Bosco et al., 2010). In addition, feeding strategies such as the use of fermented feed have been shown to improve nutrient digestibility, gut health, and overall feed efficiency in poultry (Missotten et al., 2013; Sugiharto and Ranjiktar, 2019). However, limited information is available on how these factors interact with sex in slow-growing genotypes under small-scale production conditions. Despite existing knowledge, the combined effects of outdoor rearing and different feeding regimes, particularly in slow-growing genotypes under small-scale conditions, remain insufficiently investigated. Therefore, this study aimed to evaluate the effects of outdoor housing and different feeding regimes on growth performance, meat quality, and overall efficiency in R8 red broiler chickens, with particular emphasis on feed consumption, fattening duration, and sex-related differences on growth performance.

MATERIALS AND METHODS

Animals and housing

A total of 125 one-day-old R8 Rosso Super Pesante (Sasso genetics) chickens of mixed sex, sourced from the nearest commercial local hatchery, were included in the experiment. During the first four weeks, the birds were reared in a stationary brooder (2 × 5 × 1.5 m) under controlled environmental conditions. The ambient temperature was initially maintained at 34 °C and gradually reduced by approximately 2 °C per week to ensure optimal thermoregulation. Ventilation and adaptation to external conditions were facilitated by opening the brooder windows during the daytime from the second week onwards. Wood shavings were used as bedding material at a depth of 10 cm and were regularly replenished every two days after the first week to maintain optimal hygiene and health conditions. From week five (day 37), the chickens were allocated to three experimental groups. Two groups were transferred to outdoor mobile poultry cages (chicken tractors), each providing 8 m² per 25 birds. The cages (2 × 4 × 1.5 m) were equipped with waterproof roofing and were moved daily to ensure continuous access to fresh pasture, thereby promoting natural behavior and activity (Dal Bosco et al., 2010). The remaining chickens formed a third group (n = 22), which was maintained in a stationary facility on wood shaving bedding, with open sides allowing natural daylight exposure, air circulation, and ambient temperature variation. During the brooding period (first four weeks, 29 days), a total of seven chickens died due to accidental causes. The fattening period was conducted on a natural, uncultivated meadow without prior seeding. The total fattening period lasted 121 days. Chickens did not receive any vaccines or coccidiostats during the entire fattening period.

Feeding strategy and diet composition

The feeding trial was conducted over a 121-day rearing period using a two-phase feeding program formulated by the animal feed manufacturer. All feeds were soy-free and genetically modified organisms (GMO)-free. Feed and water were provided *ad libitum* throughout the experiment. During the starter phase (days 1–28), chickens were fed a complete diet composed of corn meal, sunflower meal, rapeseed cake, flaxseed, rapeseed oil, dehydrated carrots, and a vitamin–mineral premix. The analytical composition of the starter diet was: crude protein 21.0%, crude fat 7.6%, crude fiber 5.8%, crude ash 6.3%, calcium 0.90%, phosphorus 0.60%, and sodium 0.16%. From day 29 until the end of the trial (day 121), birds were fed a finisher diet of similar composition, adjusted to support slower growth and improved meat quality. The finisher diet consisted of corn, sunflower meal, rapeseed oil, flaxseed, dehydrated carrots, and the same vitamin–mineral premix. Its analytical composition was: crude protein 17.0%, crude fat 7.0%, crude fiber 4.7%, crude ash 5.4%, calcium 0.85%, phosphorus 0.49%, and sodium 0.16%. Feed composition was provided by the feed manufacturer. At 29 days of age, chickens were randomly assigned to three feeding regimes: FerVP_pasture (n = 49), VP_pasture (n = 48), and VP (n = 22; indoor system). During the starter phase, total feed consumption was recorded at the flock level, while from day 29 onward, feed intake was monitored separately for each experimental group. Body weight was measured on a weekly basis. During the starter phase (days 1–28), a subsample of 25 chickens was randomly selected and weighed at each time point. Following allocation to experimental groups (from day 29 onward), all chickens within each group were weighed weekly. However, birds were randomly captured at each sampling point, and individual animals were not tracked over time. Therefore, body weight data collected over thirteen weekly measurements (on 37, 44, 51, 58, 65, 72, 79, 86, 93, 100, 107, 114, 121 days of age) starting on August 29th represent independent cross-sectional observations at each time point rather than repeated measurements of the same individuals.

Fermented feed preparation and chemical analysis

Fermented feed was prepared by soaking the finisher diet in water and allowing spontaneous fermentation for 72 h under controlled conditions in a separate room with a stable temperature of 22 °C. The feed was kept in clean containers and was mixed three times a day. This method, described by Missotten et al. (2013), was used to be repeatable for small farmers without any special equipment. Only feed with acceptable sensory characteristics (absence of mold and off odors) was used. Dry matter content was determined according to HRN ISO 6496:2001, ash content according to HRN ISO 5984:2023, crude protein according to HRN EN ISO 5983-2:2010 (Kjeldahl method), crude fat according to HRN ISO 6492:2001, and crude fiber according to HRN EN ISO 6865:2001.

Chemical analysis of meat composition

A total of 30 chicken breast samples, five male and five female per each of the three feeding groups, were analysed. Samples were taken after a cooling period of 24 hours and transported to the laboratory in plastic packaging (approximately 500 g) and stored under controlled conditions before analysis. Moisture content was determined according to ISO 1442, ash content according to ISO 936, protein content according to ISO 937 (Kjeldahl method), and fat content according to ISO 1443. Lipid oxidation was determined as malondialdehyde (MDA) content using the Thiobarbituric Acid Reactive Substances (TBARS) method (Grotto et al., 2009). Sample extraction and derivatization were performed according to Bertolín et al. (2019), while High-Performance Liquid Chromatography (HPLC) analysis was conducted following Agarwal and Chase (2002) and Botsoglou et al. (1994). Meat color was determined using the CIE Lab (1976) system.

Total lipids from breast muscle samples of 30 male chickens (10 per feeding treatment) were extracted according to Folch et al. (1957) using a chloroform–methanol mixture (2:1, v/v). Total lipid analyses were conducted exclusively in male chickens to reduce variability associated with sex differences in carcass composition. The extracted lipids were transesterified according to standard procedures to obtain Fatty Acid Methyl Esters (FAME). FAMEs were analysed by gas chromatography with flame ionization detection (GC-FID). Individual fatty acids were identified by comparing their retention times with those of commercial standards. Results were expressed as a percentage (%) of total identified fatty acids. Based on these data, total proportions of n-3 polyunsaturated fatty acids (n-3 PUFA) and n-6 polyunsaturated fatty acids (n-6 PUFA) were calculated, and the n-6/n-3 ratio was determined.

Statistical analysis

All statistical analyses were performed using R software (version 4.5.2; R Core Team, 2025), with the packages rstatix (Kassambara, 2023), psych (Revelle, 2025), and WRS2 (Mair and Wilcox, 2020). Data were tested for normality using the Shapiro–Wilk test (Shapiro and Wilk, 1965) and for homogeneity of variances using Levene's test, which is robust to deviations from a normal distribution (Levene, 1960) and the Bartlett's test when data were distributed normally. Statistical significance was set at $P < 0.05$. Differences in individual chicken weight between feeding treatments in five weekly measurements during the early growth phase, when sex was not yet determined (days 37 to 65), were analysed using one-way ANOVA followed by the Tukey–Kramer post hoc test (Tukey, 1949). When the assumption of homogeneity of variances was violated, Welch's one-way ANOVA followed by the Games–Howell post hoc test was applied (in the first weekly chicken measurement). After sex determination (69^{th} day onward over seven weekly measurements), the effects of feeding treatment and sex on chicken weight were analysed using a robust two-way ANOVA based on 5% trimmed means, including interaction effects, the method robust to heteroscedasticity, unbalanced groups, and large sample differences. Post hoc comparisons were performed using a one-step M-estimator with Huber's Psi function (WRS2 package), and results are presented as mean ± standard deviation (SD) or trimmed mean and Winsorised standard deviations (WSD) where appropriate. The effect of dietary treatments on n-6 and n-3 polyunsaturated fatty acids (PUFA) and n-6/n-3 ratio in breast meat of broiler chickens was analysed using one-way ANOVA followed by Tukey-Kramer post hoc test after testing for normality and homogeneity of variances.

RESULTS AND DISCUSSION

Feed analysis

Table 1 presents the chemical composition of fermented feed expressed on both a dry matter (DM) and as-fed basis. Values on a DM basis represent nutrient composition independent of moisture content, whereas as-fed values reflect the composition of the feed as consumed by the chickens. Lower nutrient concentrations observed on an as-fed basis are primarily the result of high moisture content in fermented feed. This dilution effect is a typical characteristic of wet fermentation processes and has been reported previously (Canibe and Jensen, 2012; Missotten et al., 2013).

Table 1. Chemical composition (%) of dry and fermented feed on an as-fed and dry matter basis

Component	Dry feed	Fermented feed (as feed basis)	Fermented feed (DM basis)
Crude protein	17.0	6.4	21.3
Crude fat	7.0	2.8	9.2
Crude fiber	4.7	1.7	5.6
Ash	5.4	1.6	5.3

Values are expressed as a percentage (%) on either a dry matter (DM) or as-fed basis, depending on the column. Values for dry feed represent the original unfermented diet, while values for fermented feed are presented on both an as-fed and DM basis.

When expressed on a DM basis, fermented feed exhibited higher crude protein (21.3% vs. 17.0%), crude fat (9.2% vs. 7.0%), and crude fiber (5.6% vs. 4.7%) compared to dry feed. This apparent increase does not reflect net nutrient synthesis but rather a relative concentration effect caused by microbial utilization of readily fermentable substrates, mainly carbohydrates, during fermentation. This process results in dry matter losses and enrichment of the remaining nutrients (Canibe and Jensen, 2012; Missotten et al., 2013; Niba et al., 2009). In contrast, ash content remained relatively stable (5.3% vs. 5.4%), indicating that mineral composition was not substantially affected by fermentation. This is consistent with previous findings showing that mineral fractions are less susceptible to microbial degradation during fermentation (Canibe and Jensen, 2012).

Influence of feed on growth performance

Total feed intake during the starter phase (days 1 to 28) amounted to 125 kg, corresponding to an average of 1.0 kg per chicken for the entire flock. Following the allocation of chickens to experimental groups on day 29, differences in finisher feed intake were observed among treatments. The FerVP_pasture group (n = 49) consumed a total of 680 kg of fermented feed, corresponding to 13.88 kg per chicken, which was 17.3% lower compared to the VP_pasture group. The VP_pasture group consumed 789 kg of dry feed (16.79 kg per chicken), while the VP (indoor) group consumed 417 kg (18.95 kg per chicken). Growth performance followed a three-phase pattern over the early growth phase and the sex-feed interaction phase. During the early growth phase (up to day 72), the effect of the feeding regime on body weight was inconsistent across sampling points. Significant differences between treatments were observed on specific days in the early phase, including day 37 (Welch's ANOVA, F (2, 51, 61) = 6.996 P = 0.002), day 44 (F (2, 115) = 3.81, P = 0.025), and day 72 (F(2, 115) = 3.59, P = 0.031), whereas no significant differences were detected at intermediate sampling points (days 51, 58 and 65). Post hoc comparisons for the first sampling day (day 37) indicated that chickens from the VP_pasture group were significantly heavier than both the FerVP_pasture and VP groups (P = 0.01 and P = 0.040, respectively), while no significant difference was found between FerVP_pasture and VP (P = 0.832. The opposite was found on day 72, the last sampling of the early growth phase, with no sex information. The post hoc Tukey-Kramer test revealed that the VP_pasture group had significantly lower values compared to the FerVP_pasture group (P = 0.028), while all other pairwise comparisons were non-significant. Descriptive weight statistics per group over the sampling events are shown in Table 2.

From day 72 onward, corresponding to the uniform response phase, differences between treatments became more consistent, indicating a gradual stabilization of growth patterns and increasing dietary influence. In the final phase of the experiment (days 93, 100, 107, 114 and 121), the effect of the feeding regime became more pronounced and consistent. Robust two-way ANOVA revealed a significant main effect of feeding treatment and effect of sex across all sampling points, with males being heavier than females (Tables 2 and 3), and the VP_pasture showing less growth than the VP and FerVP_pasture groups. The interaction between feeding regime and sex was not significant at earlier time points but became significant in the (days 114 and 121), with post hoc tests indicating that males responded more than females in treatments (Tables 2 and 3).

Table 2. Descriptive statistics of body weight (g) of broiler chickens in three feeding treatments during the experimental period

Days of age	FerVP_pasture n = 49 Mean ± SD (5% trimmed mean ± WSD)	VP n = 22 Mean ± SD (5% trimmed mean ± WSD)	VP_pasture n = 47 Mean ± SD (5% trimmed mean ± WSD)
37	984^{a} ± 126	1008^{a} ± 179	1144^{b} ± 265
44	1255^{a} ± 193	1406^{b} ± 281	1314^{ab} ± 201
51	1716 ± 237	1752 ± 267	1638 ± 268
58	2061 ± 312	2023 ± 341	1901 ± 325
65	2333 ± 360	2328 ± 413	2182 ± 370
72	2791^{a} ± 436	2740^{ab} ± 500	2547^{b} ± 463
79	3017 ± 491	3025 ± 569	2813 ± 490
	(3017^{a} ± 486)	(3019^{a} ± 560)	(2810^{b} ± 470)
86	3274 ± 559	3303 ± 635	2939 ± 522
	(3276^{a} ± 549)	(3302^{a} ± 626)	(2937^{b} ± 508)
93	3545 ± 649	3522 ± 715	3148 ± 550
	(3549^{a} ± 640)	(3537^{a} ± 669)	(3148^{b} ± 526)
100	3693 ± 691	3895 ± 700	3380 ± 571
	(3696^{a} ± 677)	(3896^{a} ± 696)	(3379^{b} ± 554)
107	3902 ± 689	4186 ± 716	3500 ± 624
	(3905^{a} ± 674)	(4187^{a} ± 712)	(3493^{b} ± 597)
114	4141 ± 798	4428 ± 808	3615 ± 560
	m: 4925 ± 447, n = 21	m: 5032 ± 476, n = 12	m: 4034 ± 399, n = 21
	f: 3553 ± 383, n = 28	f: 3703 ± 421, n = 10	f: 3277 ± 428, n = 26
	(4110^{A} ± 756	(4433^{A} ± 798	(3617^{B} ± 550
	m: 4931^{c} ± 429	m: 5032^{d} ± 476	m: 4036^{d} ± 382
	f: 3556^{a} ± 365)	f: 3703^{a} ± 421)	f: 3263^{b} ± 394)
121	4278 ± 820	4527 ± 750	3803 ± 508
	m: 5082 ± 461, n = 21	m: 5088 ± 430, n = 12	m: 4194 ± 302, n = 21
	f: 3675 ± 397, n = 28	f: 3853 ± 402, n = 10	f: 3506 ± 426, n = 26
	(4261^{A} ± 768	(4509^{A} ± 717	(3799^{B} ± 500
	m: 5080^{b} ± 393	m: 5088^{b} ± 430	m: 4194^{c} ± 302
	f: 3671^{a} ± 381)	f: 3853^{a} ± 402)	f: 3507^{a} ± 425)

Values represent the mean body weight (g) of broiler chickens for each feeding treatment at each sampling point, with standard deviation (SD). FerVP_pasture: fermented feed with pasture access; VP: dry feed (indoor system); VP_pasture: dry feed with pasture access; m: male; f: female. Different superscript letters (A, B, and a, b, c, d) within a row indicate statistically significant differences between treatment means based on post hoc tests ($P < 0.05$). During the early growth phase (days 37 to 65), differences were analysed using one-way ANOVA followed by the Tukey–Kramer post hoc test, or Welch's ANOVA with the Games–Howell test when assumptions were violated. From day 72 onward, differences were evaluated using robust two-way ANOVA based on 5% trimmed means. For the trimmed mean ANOVA applied at later sampling points, the trimmed mean, Winsorised standard deviation (WSD), and sample size (n) are given in brackets. For days 114 and 121, interaction was significant and basic statistics are shown by sex.

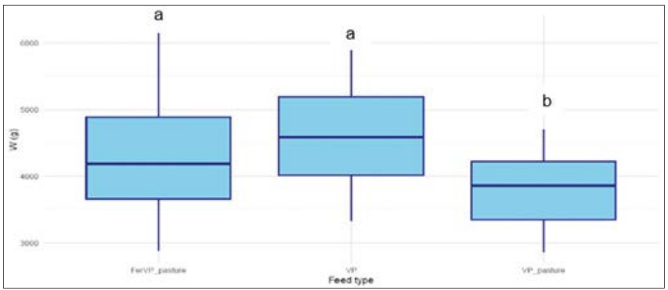
Table 3. Results of two-way Welch's ANOVA evaluating the effects of sex, feed, and their interaction on the weight (g) of chickens in three different feeding systems, with Games–Howell post hoc test significant effect and interaction estimates

Age (days)	Sex	Feed	Interaction	Significant feed effect estimates in the post hoc test
79	Q = 229.67, *P* = 0.001	Q = 14.67, *P* = 0.002	Q = 2.65, *P* = 0.281	VP_pasture < FerVP_pasture (Ψ' = -292.43, *P* = 0.030) VP_pasture < VP (Ψ' = -444.9, *P* = 0.001)
86	Q = 338.08, *P* = 0.001	Q = 40.23, *P* = 0.001	Q = 2.71, *P* = 0.273	VP_pasture < FerVP_pasture (Ψ' = -700.99, *P* = 0.001) VP_pasture < VP (Ψ' = -567.36, *P* = 0.001)
93	Q = 306.93, *P* = 0.001	Q = 46.13, *P* = 0.001	Q = 2.63, *P* = 0.285	VP_pasture < FerVP_pasture (Ψ' = -839.05, *P* = 0.001) VP_pasture < VP (Ψ' = -515.49, *P* = 0.003)
100	Q = 230.49, *P* = 0.001	Q = 25.34, *P* = 0.001	Q = 5.67, *P* = 0.070	VP_pasture < FerVP_pasture (Ψ' = -681.14, *P* = 0.001) VP_pasture < VP (Ψ' = -812.33, *P* = 0.001)
107	Q = 258.18, *P* = 0.001	Q = 49.36, *P* = 0.001	Q = 1.19, *P* = 0.561	VP_pasture < FerVP_pasture (Ψ' = -884.20, *P* = 0.001) VP_pasture < VP (Ψ' = -111.13, *P* = 0.001)
114	Q = 176.54, *P* = 0.001	Q = 59.58, *P* = 0.001	Q = 12.07, *P* = 0.005	VP_pasture < FerVP_pasture (Ψ' = -1171.33, *P* = 0.001) VP_pasture < VP (Ψ' = -1430.87, *P* = 0.001) VP_pasture × Sex(f-m) (Ψ' = -544.04, *P* = 0.022) VP × Sex(f-m) (Ψ' = -579.73, *P* = 0.002)
				f:VP_pasture < f:FerVP_pasture (Ψ' = -295.80, *P* = 0.024) f:VP_pasture < f:VP (Ψ' = -443.42, *P* = 0.024)
				m:VP_pasture < m:FerVP_pasture (Ψ' = -875.53, *P* < 0.001) m:VP_pasture < m:VP (Ψ' = -987.46, *P* < 0.001)
121	Q = 184.46, *P* = 0.001	Q = 53.74, *P* = 0.001	Q = 18.16, *P* = 0.001	VP_pasture < FerVP_pasture (Ψ' = -1052.11, *P* = 0.001) VP_pasture < VP (Ψ' = -1251.12, *P* = 0.001) VP_pasture × Sex(f-m) (Ψ' = -695.26, *P* = 0.001) VP × Sex(f-m) (Ψ' = -5379.12, *P* = 0.016)
				m:VP_pasture < m:FerVP_pasture (Ψ' = -873.64, *P* < 0.001) m:VP_pasture < m:VP (Ψ' = -994.12, *P* < 0.001)

Q: Welch's two-way ANOVA test statistic; Ψ': Huber's Psi, robust post hoc test estimate of difference between the 5% trimmed means in Welch's ANOVA; VP_pasture: dry feed with pasture access; FerVP_pasture: fermented feed with pasture access; VP: dry feed (indoor system); m: males; f: females.

In the final measurement, on day 121, females did not show any significant differences in weight over groups, while males continued to show the lowest weight averages in the VP_pasture treatment. Post hoc analysis confirmed that chickens in the VP system achieved significantly higher body weights compared to the VP_pasture treatment. While the FerVP_pasture treatment showed lower group averages, they did not differ significantly from the VP treatment.

At day 121, the distribution of final body weight clearly differed among treatments (Figure 1), with lower values observed in the VP_pasture group compared to FerVP_pasture and VP groups.



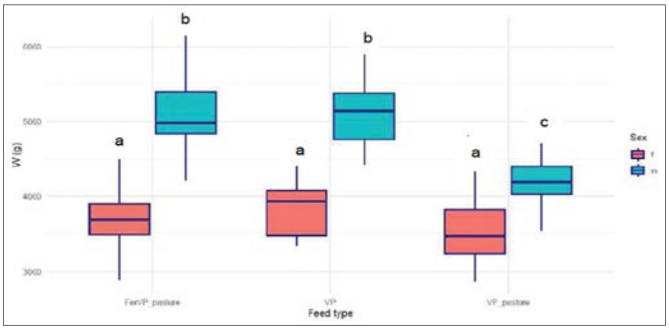
Distribution of final body weight (g) of broiler chickens according to feeding treatment. FerVP_pasture: fermented feed with pasture access; VP: dry feed (indoor system); VP_pasture: dry feed with pasture access. Boxes represent the interquartile range (25th–75th percentile), horizontal lines indicate medians, and whiskers extend to 1.5 × IQR. Differences between treatments were assessed using robust one-way ANOVA based on 5% trimmed means. A significant effect of feeding treatment was detected (P < 0.001). Post hoc comparisons were performed using a one-step M-estimator (Huber's Psi). Different letters (a, b) indicate statistically significant differences between groups (P < 0.05).

Figure 1. Distribution of body weight (g) of R8 broiler chickens by feeding treatment at 121 days of age

The results indicate a temporal shift in the dominant factors influencing broiler growth. During the early growth phase (days 37–72), the effect of the feeding regime on body weight was inconsistent. This variability is also reflected in Table 2, where the absence of consistent statistical differences suggests heterogeneous growth responses during the early stage. Such patterns are commonly reported in alternative production systems, where adaptation to diet and environmental conditions may delay the manifestation of treatment effects (Dal Bosco et al., 2010; Castellini et al., 2002).

From day 72 onward, corresponding to the later growth phase, the effect of the feeding regime became more consistent and statistically significant. A clear underperformance of the pasture system with dry feed is evident, as is the difference between males and females, but no interaction effect is visible (Table 3). These findings are consistent with the increasing influence of sexual dimorphism in broilers with age, driven by physiological differences in growth potential and protein deposition (Gous et al., 1999; Havenstein et al., 2003). As shown in Table 2, mean body weight values increased steadily, while variability (SD) remained relatively high, suggesting persistent individual variation in growth performance.

In the final growth phase (days 114–121), the interaction between feeding regime and sex had a highly significant effect on body weight. Chickens in the VP group and FerVP_pasture treatment did not differ significantly, while VP_pasture birds showed significantly lower values (Table 2; Table 4). The interaction between feeding regime and sex indicates that sex-related differences in growth were partly dependent on feeding strategy (Figure 2).



Distribution of final body weight (g) of broiler chickens by feeding treatment and sex. FerVP_pasture: fermented feed with pasture access; VP: dry feed (indoor system); VP_pasture: dry feed with pasture access. Boxes represent the interquartile range (25th–75th percentiles); horizontal lines indicate medians, and whiskers extend to 1.5 × IQR. Statistical analysis was performed using robust two-way ANOVA based on 5% trimmed means, including the effects of feeding treatment, sex, and their interaction. Feeding treatment, sex and interaction had a significant effect on body weight (P < 0.001). Post hoc comparisons were conducted using a one-step M-estimator (Huber's Psi). Different letters (a,b,c) indicate statistically significant differences between feeding treatments within each sex.

Figure 2. Distribution of body weight (g) of broiler chickens by feeding treatment and sex at 121 days of age

However, when body weight was analysed separately within each sex on the final measuring day (Tables 2 and 3, Figure 2), differences between feeding treatments were not significant in females. Males remained consistent in their response to the three treatments, with the VP_pasture group showing the worst performance. When interpreted together with feed intake data (Table 4), the higher body weight observed in the VP group appears to be associated with greater feed consumption, whereas the FerVP_pasture group may indicate improved feed utilization efficiency. This observation is consistent with previous findings that fermented feed enhances nutrient digestibility and gut health, thereby improving nutrient utilization (Missotten et al., 2013; Sugiharto and Ranjiktar, 2019). Overall, the combined analysis of descriptive inferential statistics (Tables 2 and 3) and graphical representations (Figures 1 and 2) provides a comprehensive understanding of growth dynamics.

As presented in Table 4, FCR (feed conversion ratio) was calculated as the ratio of total feed intake to total body weight gain per group. ADG (average daily gain) was calculated as the difference between final and initial body weight over the experimental period (days 29–121). Values are based on group-level data. Growth performance and feed efficiency indicators further highlighted differences between treatments. The highest average daily gain (ADG) was observed in the VP group (38.1 g/day), followed by FerVP_pasture (35.4 g/day), while the VP_pasture group showed the lowest growth rate (30.3 g/day). Despite not achieving the highest final body weight, the FerVP_pasture group exhibited the most favorable feed conversion ratio (FCR = 4.26), indicating more efficient feed utilization compared to the VP (5.40) and VP_pasture (6.03) groups. The relatively high FCR observed in the VP_pasture group suggests that pasture intake did not fully compensate for lower concentrate consumption, resulting in reduced growth efficiency.

Chemical composition of the breast meat

The VP group showed the highest intramuscular fat content (25.1 g/kg), followed by the FerVP_pasture group (15.4 g/kg), while the VP_pasture group had the lowest value (11.4 g/kg). These results, presented in Table 5, confirm that increased physical activity and pasture access reduce lipid deposition, whereas indoor systems promote fat accumulation due to lower energy expenditure (Dal Bosco et al., 2010; Fanatico et al., 2005). The highest TBARS values were observed in the VP group, likely associated with its higher fat content. In contrast, lower TBARS values in pasture-based systems suggest improved oxidative stability, potentially due to the intake of natural antioxidants from forage (Simopoulos, 2002). Regarding meat color, differences were observed for lightness (L*) and redness (a*), whereas yellowness (b*) was not affected. The VP group exhibited lower lightness values compared to FerVP_pasture, while redness was higher in the FerVP_pasture group compared to both dry feed treatments. These differences may be attributed to dietary effects on muscle pigmentation and metabolic activity. Fermented feed has been associated with improved nutrient utilization and modulation of gut microbiota, which may indirectly influence muscle characteristics and color parameters (Canibe and Jensen, 2012).

Table 4. Growth performance and feed efficiency of broiler chickens by treatment (days 29–121)

Group	n	Initial BW (kg)	Final BW (kg)	ADG (g/day)	Total feed (kg)	FCR
FerVP_pasture	49	1.02	4.28	35.4	680	4.26
VP (indoor)	22	1.02	4.53	38.1	417	5.40
VP_pasture	47	1.02	3.80	30.3	789	6.03

FCR (feed conversion ratio) was calculated as the ratio of total feed intake to total body weight gain per group. ADG (average daily gain) was calculated as the difference between final and initial body weight over the experimental period (days 37–121). Values are based on group-level data.

Table 5. Chemical composition and meat quality parameters of broiler chickens under different feeding treatments

Parameter	Unit	FerVP_pasture	VP	VP_pasture
Dry matter	g/kg	275.4	293.6	275.6
Ash	g/kg	20.1	24.8	26.8
Crude protein	g/kg	245.3	240.6	247.3
Fat	g/kg	15.4	25.1	11.4
TBARS	mg MDA/g	53.94	65.34	57.07
Lightness (L*)	–	52.34	50.31	51.56
Redness (a*)	–	1.15	0.58	0.28
Yellowness (b*)	–	15.15	14.07	14.86

Values are presented as mean (n = 30; 10 samples per treatment group). FerVP_pasture: fermented feed with pasture access; VP: dry feed (indoor system); VP_pasture: dry feed with pasture access. Dry matter, ash, crude protein, and fat are expressed in g/kg, while TBARS values are expressed as mg MDA/g. Meat color parameters (L*, a*, b*) are expressed according to the CIE Lab* system.

Overall, the results demonstrate that feeding regimes and housing systems probably influence fat deposition, oxidative stability, and certain color attributes of broiler meat, while protein content remains unaffected. The combination of fermented feed and pasture access appears to improve meat quality by reducing fat content and enhancing oxidative stability, supporting its suitability for alternative poultry production systems.

Fatty-acid content

The dietary treatments significantly affected the polyunsaturated fatty acid (PUFA) profile of the meat ($P < 0.05$), as presented in Table 6. The VP_pasture group exhibited the highest concentration of n-6 PUFA (23.28%), which was significantly higher compared to the FerVP_pasture group (F (2, 27) = 4.46; $P = 0.021$), while the VP group showed intermediate values. This increase in n-6 PUFA levels in the VP_pasture group may be associated with the high inclusion of corn- and sunflower-based components in the basal diet, combined with altered lipid metabolism in slow-growing hybrids reared under pasture conditions (Wood et al., 2008). Regarding n-3 PUFA, the indoor VP group achieved the highest deposition (9.88%), significantly exceeding both FerVP_pasture and VP_pasture groups (F (2, 27) = 5.43; $P = 0.01$), which did not differ significantly from each other. This can be attributed to the consistent intake of the basal diet containing unrefined rapeseed oil, a known source of alpha-linolenic acid (ALA). In monogastric animals, the fatty acid composition of tissues closely reflects the dietary lipid profile (Daley et al., 2010); therefore, partial replacement of concentrate feed with pasture intake could alter the balance of dietary lipid sources and reduce the relative contribution of concentrate-derived n-3 PUFA in pasture-based groups. The nutritional quality of the meat, presented in Table, expressed as the n-6/n-3 PUFA ratio, was significantly affected by dietary treatment (F (2, 27) = 7.83; $P = 0.002$). The VP_pasture group exhibited the highest (less favorable) ratio (2.71), which was significantly higher than both VP and FerVP_pasture groups ($P < 0.05$), while no significant difference was observed between the latter two groups (Figure 3). The relatively lower ratio observed in the FerVP_pasture group suggests a beneficial effect of fermented feed on lipid metabolism and nutrient utilization (Canibe and Jensen, 2012). Importantly, all observed n-6/n-3 PUFA ratios remained well within the recommended range for human health, staying below the upper limit of 4.0 (Simopoulos, 2002). Although Ponte et al. (2008) suggested that pasture has no great influence on the content of fatty acid in red broiler breast meat, this is yet to be further investigated in field conditions.

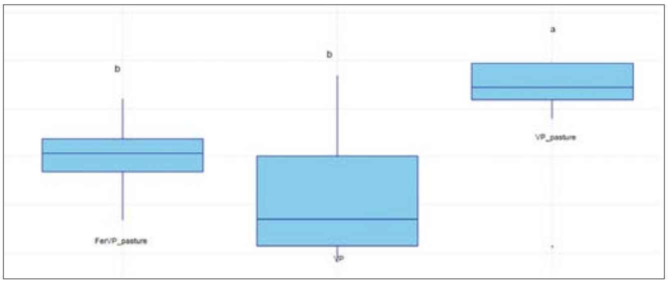
Table 6. Effect of dietary treatments on n-6 and n-3 polyunsaturated fatty acids (PUFA) and the n-6/n-3 ratio in the breast meat of broiler chickens (n = 30)

Parameter	FerVP_pasture	VP (Indoor)	VP_pasture	SEM	*P*-value
n-6 PUFA (%)	20.57^{b}	22.05^{ab}	23.28^{a}	0.64	0.021
n-3 PUFA (%)	8.58^{b}	9.88^{a}	8.65^{b}	0.31	0.011
n-6/n-3 ratio	2.40^{b}	2.25^{b}	2.71^{a}	0.08	0.002

Values are presented as mean ± SEM (n = 30; 10 samples per treatment group) and expressed as a percentage of total fatty acids. Differences between treatments were assessed using one-way ANOVA followed by Tukey-Kramer post hoc test ($P < 0.05$). Different superscript letters (a–b) within a row indicate statistically significant differences between treatments. FerVP_pasture: fermented feed with pasture access; VP (indoor system): dry feed without pasture access; VP_pasture: dry feed with pasture access.

For this research, the amount of pasture intake was not calculated. In further study, that calculation should also be taken into consideration.

Although the extended fattening period increases production time compared to intensive systems, it may better align with (Croatian) consumer demand for high-quality and ethically produced meat, similar to findings of Teixeira and Rodrigues (2021). In this research, feed fermentation and pasture access showed the potential for a possible reduction of feed costs through partial substitution of concentrate feed with forage, while the use of fermented feed may enhance feed utilization efficiency, as reflected in the lower feed intake observed in the FerVP_pasture group in the present study (Table 4).



Boxes represent the interquartile range (25th–75th percentile); horizontal lines indicate medians, and whiskers extend to 1.5 × IQR. Individual points outside this range are shown as outliers. Differences between treatments were assessed using one-way ANOVA followed by Tukey-Kramer post hoc test ($P < 0.05$). Different letters (a, b) indicate statistically significant differences between treatments. FerVP_pasture: fermented feed with pasture access; VP (indoor system): dry feed without pasture access; VP_pasture: dry feed with pasture access.

Figure 3. Effect of dietary treatments on the n-6/n-3 PUFA ratio in broiler breast meat (n = 30)

In addition, adaptation to outdoor rearing conditions and increased physical activity may contribute to improved animal resilience under variable environmental conditions. Small-scale production systems may also benefit from lower initial investment and the possibility of premium product pricing, which can partially offset increased labor requirements (Aertsens et al., 2009). Overall, the combined use of pasture-based systems and alternative feeding strategies represents a promising approach to improving both production sustainability and product value. However, further research, including detailed economic analyses, is required to fully evaluate their practical feasibility.

CONCLUSION

The results of this study show that a 121-day extended fattening period, combined with different feeding regimes, significantly affects both growth performance and meat quality in red broiler chickens. Birds reared under indoor conditions (VP) achieved the highest final body weight, confirming that conventional feeding systems maximize growth potential under the given conditions. The difference was not statistically significant from the treatment combining fermented feed and pasture access. Moreover, the combination of fermented feed and pasture access (FerVP_pasture) resulted in lower feed intake while maintaining comparable growth performance, indicating improved feed utilization efficiency. In terms of meat quality, all treatments produced meat with a favorable fatty acid profile, characterized by relatively high n-3 PUFA content and an n-6/n-3 ratio below the recommended threshold of 4.0 for human health (Simopoulos, 2002). The FerVP_pasture group exhibited a more balanced fatty acid profile compared to the VP_pasture group, suggesting a beneficial effect of combining fermentation with pasture-based systems. Overall, although indoor production systems may maximize growth performance, the integration of fermented feed and pasture access represents a promising alternative strategy for small-scale and extensive poultry production. This approach may contribute to improved feed efficiency, enhanced meat quality, and greater production sustainability. Nevertheless, further research, including comprehensive economic evaluations, is required to fully assess the practical feasibility and scalability of these systems.

ACKNOWLEGMENT

This research was made possible by the support and cooperation of the Ecological Association Vojakovac and the Croatian Regenerative Agriculture Centre (CRAC). The authors thank these organizations for providing the necessary resources and expertise in regenerative farming practices that were essential for the implementation of this study.

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