



NUTRIENT CONTENT, ANTIOXIDANT CAPACITY, AND FATTY ACIDS PROFILE OF CHERRY LAUREL (*Laurocerasus officinalis* Roemer) UNSHELLED KERNEL TO BE USED IN POULTRY NUTRITION

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Abstract: This study aims to assess the total phenolic and ascorbic acid contents, antioxidant capacity, and fatty acid profile, as well as nutrient content estimation of the cherry laurel (*Laurocerasus officinalis* Roemer) unshelled kernel (CLUK) that is considered to have the potential to improve product quality and general health in poultry nutrition. The CLUK blend obtained from fruit collected to represent cherry laurel produced in Türkiye was dried, unshelled, and ground to pass through a 1-mm sieve. This CLUK blend was analyzed according to the relevant method of each parameter to describe assessment results. The crude protein, ether extract, neutral detergent fiber, and acid detergent fiber contents of the CLUK blend were recorded to be 28.94, 34.55, 26.25, and 36.70%, respectively. The ferric reducing antioxidant power (FRAP), the radical-scavenging potencies such as DPPH (2,2-diphenyl-1-picrylhydrazyl), and ABTS (2,2'-azino-bis-(3-ethylbenzothiazoline-6-sulfonic) acid) were 139.84, 11.79, and 8.00 µg trolox equivalents mg⁻¹, respectively. A total phenolic of 3.31 mg gallic acid equivalent g⁻¹ and ascorbic acid of 1.57% contents was determined for the CLUK blend. The primary fatty acids for the CLUK blend were identified as oleic (66.61%), linoleic (15.61%), and palmitic (11.78%). These results reveal that the studied CLUK blend has the potential for quality, healthy, and eco-friendly poultry production.

Keywords: Hard stone fruit kernel, Radical-scavenging potency, Phenolic content, Fatty acid profile, Antioxidative potential, Feed ingredient and additive

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Received: November 10, 2023

Accepted: December 10, 2023

Published: January 01, 2024

Cite as: Barasoglu E, Kop Bozbay C. 2024. Nutrient content, antioxidant capacity, and fatty acids profile of cherry laurel (*Laurocerasus officinalis* Roemer) unshelled kernel to be used in poultry nutrition. *BSJ Agri*, 7(1): 51-56.

1. Introduction

Products that undergo industrialisation processes (such as sorting/cleaning, processing, cooking, and packaging) have a great biodiversity and produce non-traditional wastes (leaves, peels, pulp, kernels, etc.) that can be used in animal nutrition (Kathirvelan et al., 2015; Gungor and Erener, 2020). Recently, because fruit residues, including shelled or unshelled kernels, are inexpensive, readily available, and include bioactive molecules, several studies have shifted to such residues as sources of antioxidants (Ibrahim et al., 2017; Jideani et al., 2021). Indeed, stone fruit kernels such as candlenut (Rohaida, 2014), grape (Karabacak et al., 2015), apricot (Orhangazi, 2017), date (Tareen et al., 2017), sour cherry (Gungor and Erener, 2020), and mango (Beriso et al., 2022) kernels have attracted great interest due to their potential for quality, healthy, cost-effective, and eco-friendly poultry production. All these studies have suggested that fruit processing by-products are effective and less expensive natural sources of bioactive compounds that exhibit significant antioxidant and synbiotic properties (Ibrahim et al., 2017; Jideani et al.,

2021).

Cherry laurel (*Laurocerasus officinalis* Roemer) belongs to the Rosaceae family and is naturally found in the eastern regions of the Black Sea, Taurus Mountains, Northern and Eastern Marmara (Yildiz et al., 2014). Karabegović et al. (2014) noted that it is native to Asia Minor, Serbia, Bulgaria, Western Europe, Caucasus, Iran, and some Mediterranean countries. It is commonly consumed in fresh form and processed into jam, marmalade, fruit juice, tea, canned, dried, or pickled forms (Islam et al., 2020; Munekata et al., 2022). Kernels released during fresh consumption or food processing of cherry laurel fruits (Ayla et al., 2019) can also be considered among the kernels mentioned above (Barasoglu, 2022). However, these kernels, including the Taflan kernel, are sometimes misjudged as well as judged as effective remedies (Munekata et al., 2022) because of the adverse effects of their anti-nutritional factors on humans (Kovačević et al., 2020) and poultry (Hasted et al., 2021). In several previous investigations, several attempts have been made to evaluate the bioactive compounds, the antioxidant potential and phenolic profile, as well as nutrients of cherry laurel fruit (Kolayli et al., 2003;



Yaylaci-Karahalil and Sahin, 2011; Beyhan et al., 2018; Islam et al., 2020). Such as, there has been increasing awareness about how much the unshelled kernel from Cherry laurel fruit (CLUK) contains the total phenolic and ascorbic acid contents, antioxidant capacity, and fatty acid profile, as well as its nutrients (Barasoglu, 2022). In this context, the study reported herein aims to assess the total phenolic compounds as gallic acid equivalent (GAE) and ascorbic acid contents, antioxidant capacity [ferric reducing antioxidant power, FRAP and radical-scavenging potencies such as DPPH (2,2-diphenyl-1-picrylhydrazyl) and ABTS (2,2'-azino-bis-(3-ethylbenzothiazoline-6-sulfonic) acid)] as trolox equivalent (TE), and fatty acid profile, as well as nutrient content estimation of the CLUK, a plant-based agriculture waste, and, thus, it is functional feed ingredient/additive capacity for poultry sectors.

2. Materials and Methods

Cherry laurel fruits were harvested in August 2021 from the districts of Taflan in Samsun province, Devrek in Zonguldak province, and Adapazarı in Sakarya province of Türkiye. Subsequently, the fruits were packed in an insulated package and transported under a proper cold chain to the Department of Animal Science Laboratory at the Faculty of Agriculture, Eskişehir Osmangazi University. After removing the seeds from the fruits, they were dried in an oven at 50 °C for four days. Then, the peel was removed and ground to a particle size of 1 mm (Figure 1). Thus, the CLUK samples prepared for analysis were a blend that represented cherry laurel produced in Türkiye.

The dry matter (DM, method 930.15), ash (method 942.05), and crude protein (CP, method 976.05) analyses of the CLUK blend were analysed using approved methods (AOAC, 2006). The ether extract (EE) content was determined using an automatic fat extraction

(ANKOMXT15 Extractor) system (Seenger et al., 2008). Fibre content, including acid detergent fiber (ADF) and neutral detergent fiber (NDF), was determined following the literature (Van Soest et al. 1991) using the ANKOM A200/220 Fiber Analyzer (ANKOM Technology Corp., Fairport, NY, USA).

To assess total phenolic compounds, an ethanol and distilled water mixture (80:20 v/v) containing 0.1% HCl was acidified and utilized to extract phenolic compounds, followed by determination of total phenolic content using the Folin-Ciocalteu test (Singleton et al., 1999). To accomplish this, 1000 µl of sample extract was mixed with 500 µl of Folin-Ciocalteu and 250 µl of 20% sodium carbonate, and the final volume was adjusted to 10 ml with distilled water. The mixtures obtained were kept in a dark environment at room temperature for 30 minutes, and their absorbances were measured at 760 nm. Using gallic acid, a standard curve was obtained, and the results were computed as mg GAE g⁻¹ dry weight.

Antioxidant activity was assessed using three different *in vitro* antioxidant assays. The results for all three tests were expressed in µg TE mg⁻¹ dry weight. DPPH free radical scavenging activity was conducted following the method outlined by Brand Williams et al. (1995), with some minor adaptations. Specifically, 100 µl of the sample extract was transferred to a test tube and treated with 2900 µl of a 0.1 mM DPPH solution. The mixtures were agitated and incubated at 30 °C for 30 minutes. Subsequently, the absorbance measurements were taken against the control at 517 nm. The free radical scavenging activity of ABTS was carried out according to the protocol of Re et al. (1999). Initially, a solution of ABTS stock (7 mM) containing potassium persulfate (2.45 mM) was left in the dark for 12 h to facilitate radical cation formation and produce the ABTS test solution. The ABTS test solution was diluted with ethanol until the absorbance reached 0.700±0.02 at 734 nm.

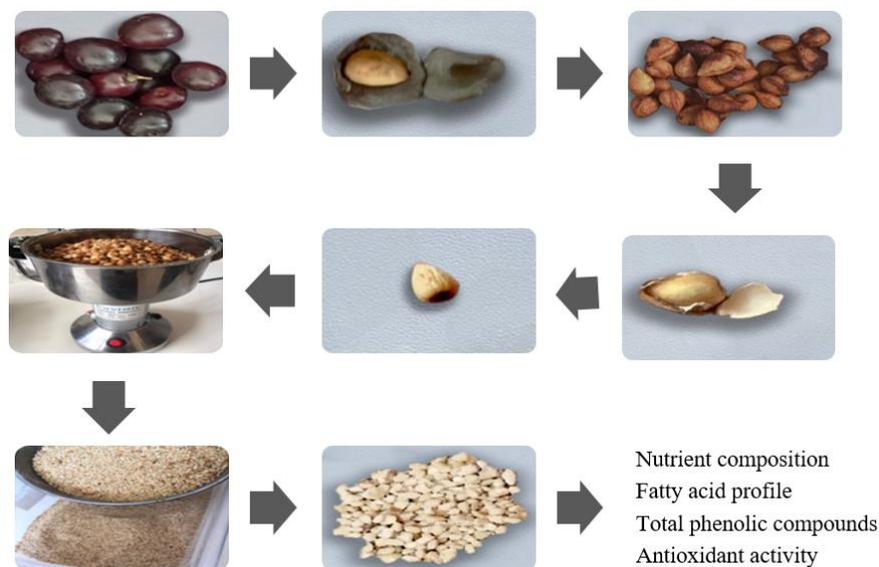


Figure 1. Flowchart summarising the steps involved in the preparation of cherry laurel (*Laurocerasus officinalis* Roemer) unshelled kernel for analyses.

The sample extract (50 µl) and the ABTS test solution (2950 µl) were diluted accordingly and mixed in a test tube. Following a 6-minute incubation period, absorbance readings were carried out at 734 nm. FRAP analysis was conducted using the method outlined by Benzie and Strain (1996) with some adjustments. The FRAP reagent was prepared by combining solutions of 2,4,6-Tris(2-pyridyl)-s-triazine (10 mmol/L), FeCl₃.6H₂O (20 mmol/L), and acetate buffer (0.3 mol/L, pH 3.6) in the appropriate ratios (1:1:10). The absorbance of a mixture containing 100 µl of sample extract, 900 µl of distilled water, and 2000 µl of FRAP reagent was measured using a spectrophotometer at a wavelength of 593 nm. The measurement was taken after a 4-minute incubation period at 37 °C.

To determine the ascorbic acid content, the spectrophotometric method, based on decolorizing the 2,6 dichlorophenol indophenol through ascorbic acid. The excess dye extracted with xylene was measured using a spectrophotometer set at a wavelength of 500 nm. The ascorbic acid content was then computed utilizing a calibration curve that was established with ascorbic acid solutions of varying concentrations (0-25 mg L⁻¹), according to Cemeroglu (2010).

The fatty acid profile was analyzed, as reported by Folch et al. (1957), utilizing the Supelco™-2380 capillary column with helium gas as carrier gas with a flow rate of 1ml/min. Chromatograms of all substances leaving the gas chromatograph column were obtained using the GC device. These were then compared with the chromatograms acquired from fatty acid standards to make qualitative and quantitative determinations (by TS EN 14214 or ISO 5508).

The measurements were undertaken in duplicates and outlined as the average value of the individual measurements.

3. Results

The crude protein, ether extract, neutral detergent fiber, and acid detergent fiber contents of the CLUK blend were recorded to be 28.94, 34.55, 26.25, and 36.70%, respectively (Table 1). The FRAP, DPPH, and ABTS were 139.84, 11.79, and 8.00 µg trolox equivalents mg⁻¹, respectively (Table 2). A total phenolic of 3.31 mg gallic acid equivalent g⁻¹ and ascorbic acid of 1.57% contents was determined for the CLUK blend. As shown in Table 3, the primary fatty acids for the CLUK blend were identified as oleic (66.61%), linoleic (15.61%), and palmitic (11.78%).

Table 1. Nutrient content of cherry laurel (*Laurocerasus officinalis* Roemer) unshelled kernel

Nutrient	%
Dry matter	95.22
Ash	3.60
Crude protein	28.94
Ether extract	34.55
Acid detergent fibre	26.25
Neutral detergent fibre	36.70

Table 2. Total phenolic content, antioxidant activity, and ascorbic acid of cherry laurel (*Laurocerasus officinalis* Roemer) unshelled kernel

Bioactive component	Quantity
Total phenolic content, mg GAE/g ⁻¹	3.31
Antioxidant activity, µg TE/mg ⁻¹	
DPPH radical scavenging	11.79
ABTS radical scavenging	8.00
Ferric reducing antioxidant power (FRAP)	139.84
Ascorbic acid, %	1.57

DPPH= 2,2-diphenyl-1-picrylhydrazyl; ABTS= 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid); FRAP= fluoride-resistant acid phosphatase.

Table 3. Fatty acids profile of cherry laurel (*Laurocerasus officinalis* Roemer) unshelled kernel

Fatty acid (FA)	g/100 g FA
Palmitic acid (C16:0)	11.78
Palmitoleic acid (C16:1)	2.60
Heptadecenoic acid (C17:1)	0.09
Stearic acid (C18:0)	2.54
Oleic acid (C18:1)	66.61
Linoleic acid (C18:2)	15.61
Alpha-linolenic acid (C18:3, omega-3)	nd
Arachidic acid (C20:0)	0.48
Eicosenoic acid (C20:1)	0.19
Eicosatrienoic acid (C20:3, omega-3)	nd
Linolenic acid (C20:3, omega-6)	0.06
Lignoceric acid (C24:0)	0.05
Saturated fatty acids	14.85
Monounsaturated fatty acids	69.49
Polyunsaturated fatty acids	15.67

nd= not detected.

4. Discussion

One of the feeding strategies for sustainable animal production is the utilisation of alternative resources and wastes. For this purpose, scientific research focuses on the waste products generated in crop production and discusses the usability of these natural products in animal nutrition. The nutrient and phytochemical composition of fruit and agricultural waste is crucial for animal health and overall performance. Bioactive and therapeutic compounds have properties that significantly enhance the health of livestock and poultry. Phytochemicals' antimicrobial and/or antioxidant

properties are vital in combatting pathogenic microorganisms, fungal growth, inflammation prevention, and health-threatening oxidative free radicals. Consequently, they contribute to the rapid growth and development of the livestock and poultry industry (Patel et al., 2017). While the CLUK blend is not a commonly used plant source as a feed additive (Barasoglu, 2022), our current results suggest that the CLUK blend can positively impact poultry's general health status, growth, and laying performance (Dhama et al., 2014). The metabolites within the CLUK blend can inhibit harmful agents in farm and poultry animals, thus halting their growth and development. Additionally, the nutrients contained within the CLUK blend can hold importance in metabolic reactions and physiological transformations within the animal body.

Choudhary et al. (2023) stated that mango kernel contains 53.34-76.81% carbohydrate, 5.20-10.48% protein, 9.84-18.0% fat, and 0.26-10.60% crude fiber. The researchers concluded that mango kernel, a significant source of phytochemicals and nutrients, may be used as a maize substitute in rations. Beyene et al. (2019) reported that mango kernels can replace maize entirely in laying hen diets, resulting in improved economic returns without impacting egg yield or quality parameters. Moreover, Beriso et al. (2022) reported that incorporating up to 15% boiled mango kernel in Hubbard broiler diets can improve production performance and feed conversion parameters without adverse effects. Our results on nutrient content indicate the potential of the CLUK blend as a feedstuff is controversial for poultry nutrition due to the high ADF and NDF percentages in the CLUK blend. Fiber components naturally found in poultry diets directly affect intestinal morphology, organ growth, nutrient utilisation, and microflora modulation to varying degrees (Tejeda and Kim, 2021). The effects of each fiber component need to be determined chemically and physiologically. A high fiber content and/or the presence of antinutritional substances in the kernel can reduce the digestibility of feed. Furthermore, it should be noted that the fiber values identified in our study were obtained from the kernel after removing the shell of the cherry laurel kernel. Fruit kernels may contain antinutritional elements, which could harm poultry (Rad et al., 2015). Therefore, it is necessary to establish whether the CLUK blend has toxic or antinutritional properties. It has been reported that date kernel has a negative impact on the performance parameters of broiler chickens, resulting in a decrease in live weight when included at levels of 10% and 20% (Masoudi et al., 2011; Kheiri and Nasr, 2013). Arbouche et al. (2012) showed that adding apricot kernel to the diet above the level of 6% adversely affected the growth performance of broiler chickens due to the antinutritional factors present in its kernels. However, there are measures to reduce these antinutritional factors, effectively eliminating the negative effects (Jazi et al., 2017). Hence, *in vivo* studies are necessary to determine and use antinutritional factors of CLUK blend

at varying doses. In addition, the production and consumption amounts of the cherry laurel fruit in our country still need to be clarified. As such, regrettably, its use as a raw material cannot be recommended. After clarifying the abovementioned issues, it is possible to reduce the cost by using it as feed raw material at low doses in small enterprises regionally.

Fruit and vegetable waste contains numerous antioxidative phytochemicals, such as vitamins C and E, carotenoids, polyphenols, and other bioactive compounds (González-Aguilar et al., 2008). Phenolic compounds-comprising a hydroxyl group attached to a benzene ring-are generally responsible for the odor, color, and flavour of plants. Cherry laurel is abundant in antioxidant substances, including phenolics such as chlorogenic acid, phenolic acids, anthocyanins, and vanillic acid, as well as ascorbic acid (Ayaz et al., 1997; Kolayli et al., 2003; Yaylaci-Karahalil and Sahin, 2011). Engin (2007) reported that the leaves, fruits, and seeds of cherry laurel exhibit antioxidant activity, suggesting their potential use as natural antioxidants in the food, cosmetics, and pharmaceutical industries. Islam et al. (2020) found the total phenolic contents of the fruit of 7 different cherry laurel genotypes between 2.76-8.30 mg GAE g⁻¹ and FRAP and DPPH values between 4.96-25.37 mmol TE g⁻¹ and 1.07-12.19 mmol TE g⁻¹, respectively. Based on the total phenolic content, antioxidant activity, and vitamin C results obtained in this study, we can conclude that CLUK blend demonstrates antioxidant activity and thus, can serve as a suitable feed additive for poultry diets. In fact, various studies assess fruit kernels as feed additives due to their high concentration of antioxidative phytochemicals. Karabacak et al. (2015) stated that the co-administration of the grape kernel with ionophoric antibiotics effectively reduced the adverse effects of these antibiotics. These effects of the grape kernel could be ascribed to the ability of substances with high antioxidant capacity to scavenge radicals and their regulatory impact on the balance between oxidants and antioxidants. Gungor and Erener (2020) noted that cherry kernels, which have anticarcinogenic, anti-inflammatory, antidiabetic, antioxidant, and antimicrobial properties, can be a potential feed additive that improves growth performance when used in broiler diets at a rate of 1%.

In the present study, the EE content and fatty acid profile results suggested that while the CLUK blend contributes to meeting the energy needs of laying hens, it can enrich the fatty acid profile of the egg. In laying hens, we have shown that the level of unsaturated fatty acids in eggs can be enriched by manipulating the diet, with the fatty acid profile of the yolk being highly dependent on the fatty acid profile of the diet (Kop-Bozbay et al., 2021). Similarly, in broiler chickens, Rohaida (2014) suggested that candlenut kernels, which are rich in alpha-linolenic and linoleic acids, can be a source of omega-3 fatty acids and that dietary supplementation can enrich the fatty acid content of chicken meat. It is desirable for

consumers and producers to increase the content of unsaturated fatty acids in foods of animal origin. In light of the information described, it is concluded that the CLUK blend can improve meat and egg quality and contribute to consumer health by modifying the fatty acid profile.

5. Conclusion

These results reveal that the studied CLUK blend has the potential for quality, healthy, and eco-friendly poultry production. Therefore, the possibilities of using CLUK blend as an alternative feed additive, as well as a feedstuff in poultry nutrition are worth investigating due to its high fiber content, total phenolic matter, antioxidant capacity, and fatty acid profile. More extensive research is needed to determine the effects of using CLUK blend as a feed additive on the reproductive, health, and yield characteristics of poultry and to demonstrate its benefits.

Author Contributions

The percentage of the author(s) contributions is presented below. All authors reviewed and approved the final version of the manuscript.

	E.B.	C.K.B.
C	50	50
D		100
S		100
DCP	50	50
DAI	100	
L	50	50
W	50	50
CR		100
SR		100
PM		100
FA		100

C=Concept, D= design, S= supervision, DCP= data collection and/or processing, DAI= data analysis and/or interpretation, L= literature search, W= writing, CR= critical review, SR= submission and revision, PM= project management, FA= funding acquisition.

Conflict of Interest

The authors declared that there is no conflict of interest.

Ethical Consideration

Ethics committee approval was not required for this study because of there was no study on animals or humans.

Acknowledgments

This research was part of an MSc project of the first author and was supported by the Scientific Research Fund of Eskisehir Osmangazi University (202123007). The authors are grateful for the support of the Scientific Research Fund of Eskisehir Osmangazi University. The authors would like to thank Emre TURAN for the analysis of the bioactive components.

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