

Effect of emulsifier supplementation on UCP3 gene expression in different tissues of broiler chickens

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Abstract

Objective

Emulsifiers can change the access of cells to fatty acids by increasing fat digestion and absorption and affect the pathways regulating energy metabolism. The Uncoupling Protein 3 (UCP3) gene is known as one of the genes related to mitochondrial function, fatty acid oxidation, and

maintaining cellular energy balance. The aim of this study was to investigate the effect of dietary emulsifier supplementation on the relative expression of the UCP3 gene in different tissues of broiler chickens.

Materials and methods

In this experiment, 320 Ross broiler chickens were studied in a completely randomized design. The experimental treatments included four types of diets (basal diet, diet with a 5% reduction in protein, diet with a 100 kcal reduction in metabolic energy, and diet with a simultaneous reduction of 5% protein and 100 kcal of energy) at two levels of no consumption and consumption of emulsifier supplement. Each treatment included four replicates and each replicate included 10 chickens. After the end of the 42-day rearing period, femur muscle, breast muscle, liver and spleen tissue samples were collected and frozen in liquid nitrogen. Total RNA was extracted, cDNA was synthesized and UCP3 gene expression was assessed using Real-Time PCR. GAPDH gene was used as a reference gene and changes in gene expression were calculated using the $2^{-\Delta\Delta Ct}$ method.

Results

The results showed that emulsifier supplement caused a relative decrease in UCP3 gene expression in all tissues studied. In femur muscle, a decrease in UCP3 expression was observed compared to the control group, but this change was not statistically significant ($P > 0.05$). In breast muscle, emulsifier consumption caused a significant decrease in UCP3 gene expression ($P < 0.01$). Also, the greatest decrease in expression was observed in liver tissue, which was statistically highly significant ($P < 0.001$). In contrast, changes in UCP3 expression in spleen tissue were not significant ($P > 0.05$).

Conclusion

Based on the results of this study, emulsifier supplementation, in addition to possibly improving dietary fat utilization, can modulate the expression of gene related to energy metabolism in different tissues of broiler chickens. The decrease in UCP3 expression in breast muscle and liver probably indicates an improvement in cellular energy status, a decrease in mitochondrial metabolic stress, and a decrease in the need to activate adaptive pathways related to fatty acid oxidation. These findings highlight the importance of investigating molecular responses to nutritional additives alongside performance indicators and could help develop more precise nutritional strategies in the poultry industry.

Keywords: Emulsifier, Energy Metabolism, Gene Expression, Real-Time PCR, UCP3

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Introduction

The poultry industry is one of the most important food production sectors in the world due to its high growth rate, favorable feed-to-meat conversion efficiency, and important role in providing animal protein (Mohammadabadi et al., 2025). With the increase in feed costs, optimizing the use of nutrients, especially energy sources, has become one of the main goals of nutritional research in poultry. Fats, as dense energy sources in the diet of broilers, in addition to providing the energy required for growth, improve feed consumption and increase production efficiency (Nejad et al., 2024). However, limitations in fat digestion and absorption, especially in the early stages of growth, can prevent the full use of energy in the diet (Khezri et al., 2025). The use of emulsifiers is one of the nutritional strategies to increase the efficiency of fats in poultry diets. These compounds improve lipid digestion and absorption by reducing the size of fat droplets, increasing their contact surface with digestive enzymes, and facilitating micelle formation. Compounds such as lecithin, lysolecithin, and other commercial emulsifiers have received attention as functional additives in poultry nutrition in recent years (Guerreiro Neto et al., 2011). Various studies have shown that emulsifier supplementation can increase fat digestibility, energy retention, and growth performance of broiler chickens. Park et al. (2018) reported that the use of lysolecithin in broiler diets improved feed conversion ratio and increased energy and fat retention of the diet. Also, a recent meta-analysis showed that the use of lecithin and lysolecithin can improve growth performance and nutrient digestibility in poultry (Priyatno et al., 2025). Although most studies on emulsifiers have focused on performance indicators, nutrient digestibility, carcass characteristics, and blood parameters, emerging evidence suggests that the effects of these compounds may extend beyond the gastrointestinal tract and be exerted through the regulation of molecular pathways related to energy metabolism (Gholami et al., 2024). Increased fatty acid uptake and greater cellular access to energy sources can affect mitochondrial activity, fatty acid oxidation, and the expression of genes related to energy balance. Among the genes related to energy metabolism, the Uncoupling Protein 3 (UCP3) gene is of particular importance due to its role in mitochondrial function, fatty acid oxidation, and the control of reactive oxygen species production. Mitochondrial uncoupling proteins (UCPs) control the coupling between the electron transport chain and ATP production by regulating the proton gradient across the inner mitochondrial membrane and can release part of the energy from nutrient oxidation as heat (Ricquier, 2011; Mohammadabadi et al., 2024). UCP3, which was first identified in 1997, is mainly expressed in tissues with high metabolic activity, especially skeletal muscle, and plays a role in the transport of fatty acids into mitochondria, increasing lipid oxidation and reducing oxidative damage caused by fatty acid accumulation (Vidal-Puig et al., 2000). In birds, UCP3 is considered one of the important genes involved in the regulation of energy metabolism and its expression is influenced by factors such as age, environmental temperature, dietary energy level, fatty acid composition and oxidative stress conditions. One of the important pathways regulating UCP3 expression is the peroxisome proliferator-activated receptor (PPAR) pathway. PPAR α and PPAR δ act as important transcription factors in regulating the transport, storage, and oxidation of fatty acids, and increased entry of free fatty acids into the cell can activate this pathway and increase the expression of metabolic genes, including UCP3 (Kindall et

al., 2025). In addition, the AMPK pathway and the transcription factor PGC-1 α are important regulators of energy homeostasis, mitochondrial biogenesis, and increased oxidative capacity of the cell, and are closely related to the function of UCP3 (Lin et al., 2002). Given that emulsifiers increase the bioavailability of fats and change the entry of fatty acids into tissues, it is likely that their effects are not limited to improving digestion and absorption, but are also exerted through the regulation of molecular pathways related to energy metabolism. However, studies conducted in broilers have focused more on the functional effects of emulsifiers, and there is limited information on the effect of these compounds on the expression of genes related to mitochondrial function (Dridi et al., 2022). The metabolic response of the bird body to nutritional factors depends on the specific characteristics of the tissues. In the present study, breast muscle, femur muscle, liver and spleen were investigated due to their different roles in meat production, energy consumption, fat metabolism and regulation of physiological responses. Skeletal muscles, especially femur muscle, play an important role in energy consumption due to their higher capacity for fatty acid oxidation, while breast muscle shows a distinct metabolic response with a different muscle fiber composition. The liver is also the main center of fatty acid synthesis and metabolism in birds, and changes in fat absorption can affect its metabolic activities. Also, although the spleen is not a major organ of energy metabolism, the close relationship between metabolism and immune function suggests that nutritional status can influence cellular processes in this tissue. Despite numerous studies on the functional effects of emulsifiers, there is still limited information on their direct effect on mitochondrial gene regulation in different tissues of broiler chickens. Therefore, investigating UCP3 expression in tissues with different functions may provide new information on the molecular mechanisms of broiler chickens' response to emulsifier supplementation.

Materials and methods

Animals, experimental design, and experimental diets: This experiment was conducted using 320 one-day-old male broiler chicks of Ross strain (Ross 308) at the Animal Science Training and Research Station of Shahid Bahonar University of Kerman. The experimental design was designed as a completely randomized 4 $\times$ 2 factorial arrangement including four dietary levels and two levels of emulsifier supplementation. The experimental treatments included: 1) basal diet, 2) basal diet with a 5% reduction in crude protein, 3) basal diet with a 100 kcal reduction in metabolizable energy, and 4) basal diet with a simultaneous 5% reduction in crude protein and 100 kcal reduction in energy. Each of these four types of diets was studied in two conditions with and without emulsifier supplementation. In total, eight experimental treatments were considered with four replications for each treatment and 10 chicks in each replication. The chicks had free access to feed and water during the 42-day rearing period and were reared under the same management conditions including standard lighting, temperature and ventilation schedule. The emulsifier used in this study was Artifier (Artiot, USA). This supplement consisted of 25% lysophospholipids including lysophosphatidylcholine, lysophosphatidic acid, lysophosphatidyl inositol and lysophosphatidylethanolamine, 40% lecithin, 6% polyethylene glycol ricinoleate (PEGR) and 29% carrier.

Tissue sampling: At the end of the rearing period (day 42), the chicks were killed and tissue sampling of liver, spleen, breast muscle and femur muscle was performed. Immediately after slaughter, small pieces of each tissue were separated using sterile instruments and, to prevent RNA degradation, were placed in aluminum foil and quickly frozen in liquid nitrogen. After complete freezing, the samples were transferred to a -80°C freezer and stored until RNA extraction. Three independent samples were prepared from each tissue for molecular experiments.

Chemicals and kits used: The materials used in this study included frozen tissue samples, Trizol (Zavar Zist Azma, Iran), chloroform (Merck, Germany), isopropanol (Merck, Germany), 70% ethanol (Nasr, Iran), DEPC water (Cynagen, Iran), cDNA synthesis kit (Pars Tous, Iran), Mastermix PCR Red (Ampliqon, Denmark), Mastermix Real-Time PCR (Pars Tous, Iran), agarose, DNA Ladder and Safestain (Cynagen, Iran), and specific primers (Gene Fan Avaran, Iran).

RNA extraction and RNA quality assessment: Total RNA was extracted from tissue samples using the TRIZol method. After extraction, RNA quantity was determined using a Nanodrop device and optical absorption ratio at wavelengths of 260/280 and 260/230 nm. The quality of extracted RNA was also assessed using agarose gel electrophoresis. RNA samples of appropriate quality were used for cDNA synthesis.

cDNA synthesis: To convert extracted RNA into complementary DNA (cDNA), a cDNA synthesis kit from Pars Tous (Iran) was used according to the manufacturer's instructions. The cDNA synthesis reaction was performed in a final volume of 20 µl. The reaction temperature program included an initial incubation step at 25°C for 10 min, cDNA synthesis at 47°C for 60 min, and a reaction stop step at 85°C for 5 min. After synthesis, cDNA samples were stored at -20°C until Real-Time PCR was performed.

Real-Time PCR gene expression assay: In order to examine the relative expression of UCP3 gene, Real-Time PCR method was used using cDNA as a template. GAPDH gene was used as an internal reference gene to normalize gene expression data. Target gene and reference gene specific primers are presented in Table 1.

Table 1- Sequence of primers used in Real-Time PCR

Gene	primer	Sequence (5'→3')	Fragment length (bp)	Annealing temperature (°C)
UCP3	Forward	AAGGATGGAGGTGTTTCACAG	161	57
	Reverse	GTGAGGAATACCCGGACTCA		
GAPDH	Forward	ATGGCATCCAAGGAGTGAGC	66	58
	Reverse	AACAAAGGGTCCTGCTTCCC		

Conventional PCR reaction and optimization of amplification conditions: In order to determine the optimal amplification conditions and to investigate the specificity of the primers for the UCP3 gene and the reference gene GAPDH, a conventional PCR reaction was performed using cDNA as a template. The reaction in a final volume of 20 µL consisted of 10 µL of Red

PCR Master-Mix, one μL of each of the Forward and Reverse primers (10 pmol/ μL), one μL of cDNA, and deionized water to a final volume of 20 μL . The PCR reaction was performed for 35 cycles including initial denaturation at 95°C for 5 min, denaturation at 95°C for 1 min, primer annealing at 57°C for UCP3 and 58°C for GAPDH for 1 min, extension at 72°C for 1 min, and final extension at 72°C for 5 min. PCR products were examined by agarose gel electrophoresis and the accuracy of the expected fragments was confirmed. The accuracy of PCR products was examined using agarose gel electrophoresis. Also, in the Real-Time PCR reaction, the amplification curve and the melt curve were evaluated to ensure the specificity of the products.

Calculation of relative gene expression: The relative expression level of the UCP3 gene was calculated using the $2^{-\Delta\Delta\text{Ct}}$ method. First, the ΔCt value was calculated for each sample from the following equation:

$$\Delta\text{Ct} = \text{Ct (target gene)} - \text{Ct (reference gene)}$$

Then the $\Delta\Delta\text{Ct}$ value was determined using the following equation: $\Delta\Delta\text{Ct} = \Delta\text{Ct (treatment sample)} - \Delta\text{Ct (control sample)}$ Finally, the relative changes in gene expression were calculated from the following equation:

$$2^{-\Delta\Delta\text{Ct}} = \text{relative change in gene expression}$$

Statistical analysis: Data obtained from Real-Time PCR were extracted and processed using Rotor-Gene Q Series Software. Statistical analysis of data was performed using GraphPad Prism 8 and Excel software. One-way ANOVA was used to compare the effects of experimental treatments on UCP3 gene expression, and comparisons of means were performed at a significance level of 0.05. Results were reported as mean $\pm$ standard error of the mean (Mean $\pm$ SEM).

Results

Quality and quantity of extracted RNA: The quality of RNA extracted from femur muscle, breast muscle, liver and spleen tissues was examined using 2% agarose gel electrophoresis. As can be seen in Figure 1, the 28S and 18S ribosomal bands were clearly visible without degradation, indicating the appropriate quality of the extracted RNA. Also, the examination of the quantity and purity of RNA using the Nanodrop device showed that the absorption ratio of 260/280 and 260/230 was within the standard range and the RNA samples were free of significant protein contamination and organic compounds. Therefore, the extracted RNA was of sufficient quality and purity for cDNA synthesis and Real-Time PCR reaction.

Verification of cDNA synthesis and primer specificity: In order to verify the accuracy of cDNA synthesis, a PCR reaction was performed using GAPDH reference gene primers. The observation of a specific band with the expected size (66 bp) indicated the success of cDNA synthesis. Also, to determine the appropriate annealing temperature of the UCP3 gene primers, a gradient PCR reaction was performed. The electrophoresis results showed that at the optimal annealing temperature, only one specific band with a size of 161 bp was produced, indicating the appropriate specificity of the primers and the absence of amplification of non-specific products (Figure 2).

Evaluation of Real-Time PCR Reaction: The amplification curves obtained from Real-Time PCR showed that the samples entered the exponential phase of amplification within the

range of 15 to 25 cycles and then entered the linear phase. Also, the examination of the melting curve for the UCP3 gene showed only one specific peak, indicating the specific amplification of the target gene and the absence of primer dimer formation or non-specific products. In addition, electrophoresis of the Real-Time PCR products also confirmed the presence of a specific band with a size of 161 bp, which was consistent with the melting curve results.

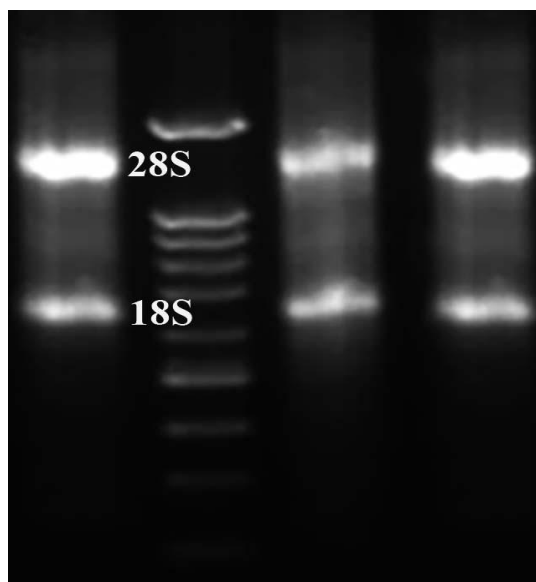


Figure 1. Electrophoresis of total RNA extracted from different tissues of broilers on 2% agarose gel. Clear bands of 18S and 28S ribosomal RNA indicate the appropriate quality of the extracted RNA

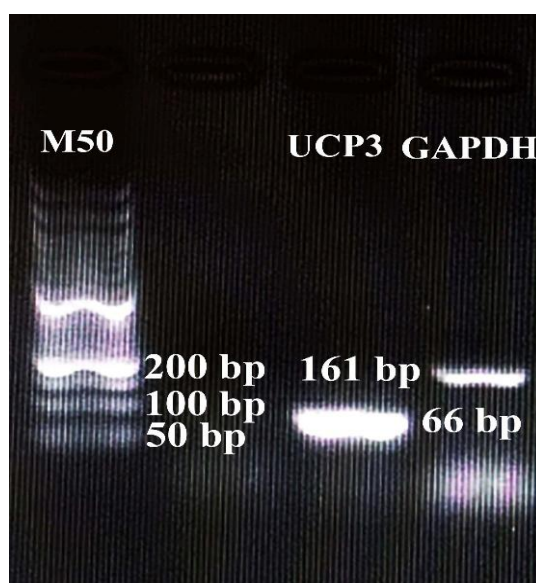


Figure 2. Verification of cDNA synthesis and amplification specificity of the UCP3 and GAPDH genes using conventional PCR reaction and electrophoresis of PCR products on agarose gel. The amplified fragments include UCP3 with a length of 161 bp and GAPDH with a length of 66 bp

Relative expression of the UCP3 gene in different tissues

Femur muscle: The effect of emulsifier supplementation on the expression of the UCP3 gene in the femur muscle is presented in Figure 3. Consumption of the diet containing emulsifier caused a decrease in the relative expression of this gene compared to the control group, but this decrease was not statistically significant ($P>0.05$).

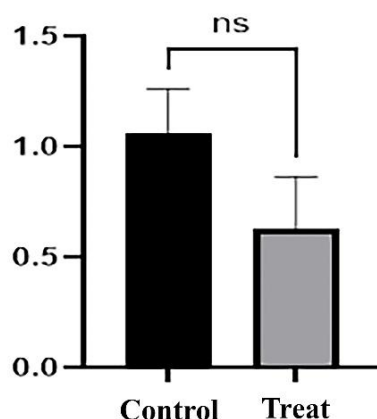


Figure 3. Effect of dietary emulsifier supplementation on the relative expression of the UCP3 gene in femur muscle tissue of broiler chickens. Values are shown as mean $\pm$ standard error of the mean (Mean $\pm$ SEM)

Breast muscle: As can be seen in Figure 4, dietary supplementation with emulsifier significantly reduced UCP3 gene expression in the breast muscle of broiler chickens compared to the control group ($P<0.01$).

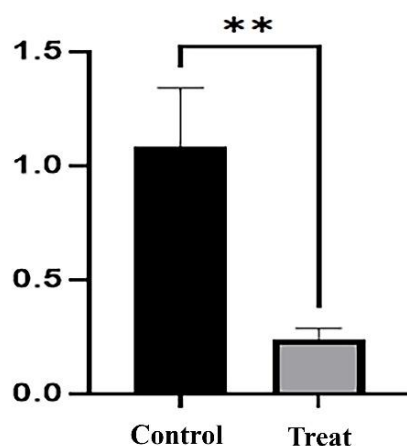


Figure 4. Effect of dietary emulsifier supplementation on the relative expression of the UCP3 gene in the breast muscle tissue of broiler chickens. Significant differences between the emulsifier-treated group and the control group are indicated by ** ($P<0.01$)

Liver: The results related to liver tissue (Figure 5) showed that the consumption of emulsifier significantly reduced UCP3 gene expression compared to the control group. This decrease was statistically significant ($P < 0.001$).

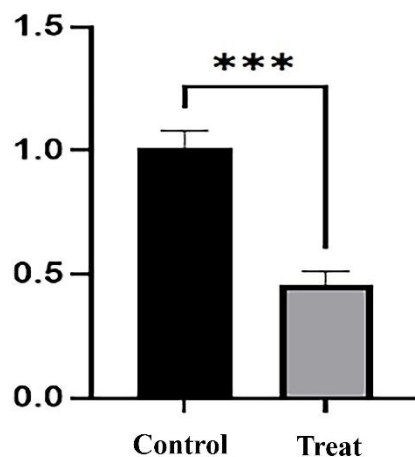


Figure 5. Effect of dietary emulsifier supplementation on the relative expression of the UCP3 gene in the liver tissue of broiler chickens. A significant decrease in gene expression was observed in the emulsifier-fed group compared to the control group ($P < 0.001$)

Spleen: As shown in Figure 6, although the relative expression of the UCP3 gene in the spleen of chickens fed with emulsifier showed some changes compared to the control group, this difference was not statistically significant ($P > 0.05$).

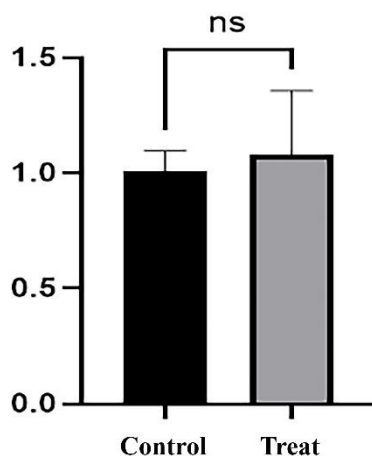


Figure 6. Effect of dietary emulsifier supplementation on the relative expression of the UCP3 gene in the spleen tissue of broiler chickens. Values are presented as mean ± standard error of the mean (Mean ± SEM)

In general, dietary supplementation with emulsifiers decreased UCP3 gene expression in liver, breast muscle, and femur muscle tissues, but this decrease was statistically significant only

in breast muscle and liver. The greatest decrease in gene expression was observed in liver tissue ($P < 0.001$), while there was a non-significant increase in spleen, and the decrease in femur muscle expression was not significantly different from the control group.

Discussion

The present study was conducted to investigate the effect of emulsifier supplementation on UCP3 gene expression in femur muscle, breast muscle, liver and spleen tissues of broiler chickens. The results showed that the use of emulsifier reduced UCP3 gene expression in all tissues studied, but this reduction was statistically significant only in breast muscle and liver. This finding indicates that emulsifiers, in addition to improving fat digestion and absorption, are also able to affect molecular pathways related to energy metabolism. By reducing the surface tension between water and fat, emulsifiers facilitate the formation of stable micelles and, as a result, increase lipid digestion and absorption (Wang et al., 2024). This process increases the access of cells to fatty acids and improves the utilization of dietary energy. Several studies have shown that emulsifier supplementation improves fat digestibility, increases metabolizable energy, and improves growth performance in broiler chickens (Guerreiro Neto et al., 2011; Park et al., 2018; Ahmadi-Sefat et al., 2022; Adli et al., 2026). The UCP3 gene is a member of the mitochondrial uncoupling protein family, which is mainly expressed in metabolically active tissues, especially skeletal muscle (Schrauwen, 2002; Ricquier, 2011). It is now accepted that the main role of this protein is not heat production, but rather the regulation of fatty acid transport and oxidation, maintenance of mitochondrial function, and reduction of reactive oxygen species (ROS) production. Increased UCP3 expression is usually observed in conditions of increased free fatty acids, energy deficiency or oxidative stress, whereas its expression can be reduced in conditions of increased energy metabolism efficiency (Vidal-Puig et al., 2000; Schrauwen, 2002). In the present study, the reduction in UCP3 expression probably reflects an improvement in the energy status of the cell due to increased fat digestibility and absorption. When emulsifiers cause more efficient utilization of fatty acids, the need for the cell to activate compensatory pathways for fatty acid transport and oxidation is reduced, and as a result, UCP3 gene expression is also reduced. This interpretation is consistent with the results of recent studies that have shown that emulsifiers, in addition to improving digestion, also modulate molecular pathways related to lipid metabolism and PPAR signaling (Richards, 2003; Adli et al., 2026). One of the most important findings of the present study was a significant decrease in UCP3 expression in liver tissue. In birds, the liver is the most important organ regulating fat metabolism, fatty acid synthesis, and lipid distribution in the body. Therefore, any increase in fat absorption capacity can directly change the metabolic activity of this organ. It is likely that the increased intake of absorbable fatty acids increased the efficiency of oxidation and reduced the need to activate mitochondrial protective mechanisms, which explains the decrease in UCP3 expression. Recent molecular studies have also shown that emulsifiers can modulate the expression of genes related to fatty acid transport and oxidation, such as CPT1 and PPAR α , and drive lipid metabolism towards more efficient fat utilization (Lin et al., 2002; Schrauwen, 2002; Adli et al., 2026). UCP3 gene expression was also significantly reduced in breast muscle. Broiler breast muscle is mainly composed of glycolytic fibers and has

a lower capacity for fatty acid oxidation than oxidative muscles. Therefore, the improvement in energy status resulting from emulsifier consumption probably reduced the need for this tissue to express genes related to the regulation of mitochondrial function. In contrast, although a trend of decreased expression was also observed in femur muscle, this change was not statistically significant. Femur muscle has a higher mitochondrial density and higher oxidative capacity and is likely to have a greater ability to maintain energy homeostasis in the face of nutritional changes; therefore, its molecular response was more limited than breast muscle. In the spleen, although a trend of increased UCP3 expression was observed, there was no significant difference with the control group. Unlike muscles and liver, the spleen is not considered a major organ of energy metabolism and its role is more in regulating immune responses. For this reason, nutritional changes are expected to have a less direct effect on the expression of genes related to fatty acid oxidation in this tissue. Recent studies have shown that the effects of emulsifiers are not limited to the gastrointestinal tract, and that these compounds can also affect the expression of genes related to lipid metabolism, the activity of lipolytic enzymes, and PPAR-dependent signaling pathways. A meta-analysis that reviewed 57 studies showed that emulsifier supplementation improved fat digestion, energy utilization, and the expression of genes related to fatty acid oxidation, although the magnitude of the response depended on the type of emulsifier, dietary energy level, and tissue type (Adli et al., 2026). The results of this study are consistent with recent reports that the effects of emulsifiers are not limited to improving digestion but can also affect metabolic pathways (Gholami et al., 2024). Despite the valuable results of this study, there were some limitations. In this study, only the mRNA expression of UCP3 was measured and changes in protein level or mitochondrial functional activity were not evaluated. Also, simultaneous measurement of other key genes such as PPAR α , PGC-1 α , AMPK and CPT1 can help to clarify the mechanism of the effect of emulsifiers on energy metabolism in future studies.

Conclusion: The results of this study showed that emulsifier supplementation reduced UCP3 gene expression in femur muscle, breast muscle, and liver tissues of broiler chickens, but this reduction was statistically significant only in breast muscle and liver. These findings indicate that emulsifiers, in addition to increasing fat digestion and absorption, also affect the regulation of genes related to energy metabolism, possibly by improving cellular energy status and modulating mitochondrial function. From a practical perspective, these results indicate that emulsifiers can play a role as a tool in precision poultry nutrition, in addition to improving production performance, in regulating molecular responses related to energy efficiency. However, to fully elucidate the mechanisms involved, it is necessary to conduct complementary studies at the protein level, enzyme activity, and other pathways regulating energy metabolism.

Author contributions

Conceptualization and design: M.M. and M.S.; Data acquisition: M.M. and R.A.M.; Data analysis: A.K., O.B., I.T., O.K., S.U., and M. Momen; Data interpretation: M.M., V.S. and M.S.; Manuscript drafting: M.M.; Critical revision of the manuscript: M.M., and O.B.; Final approval of the manuscript: M.M., M.S. and A.K.; Writing-original draft preparation: M.M., R.A.M., O.B., I.T., O.K., S.U., M. Momen, and V.S.; Writing-review and editing, supervision:

M.M. All authors have read and approved the final version of the manuscript.

Data availability statement

The corresponding author will provide the data upon request.

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Ethical considerations

The current investigation was managed at the Shahid Bahonar University of Kerman, Iran and was approved by the Institutional Animal Care and Use Committee Protocol #IR22979001 based on the guidelines of the Iranian Council of Animal Care (1995).

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Conflict of interest

The authors declare that there are no conflicts of interest related to this investigation.

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اثر مکمل امولسیفایر روی بیان ژن UCP3 در بافت‌های مختلف جوجه های گوشتی

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چکیده

هدف: امولسیفایرها با افزایش هضم و جذب چربی می‌توانند دسترسی سلول‌ها به اسیدهای چرب را تغییر داده و بر مسیرهای تنظیم‌کننده متابولیسم انرژی اثرگذار باشند. ژن Uncoupling Protein 3 (UCP3) به‌عنوان یکی از ژن‌های مرتبط با عملکرد

میتوکندری، اکسیداسیون اسیدهای چرب و حفظ تعادل انرژی سلولی شناخته می‌شود. هدف از این پژوهش، بررسی اثر مکمل امولسیفایر جیره بر بیان نسبی ژن UCP3 در بافت‌های مختلف جوجه‌های گوشتی بود.

مواد و روش‌ها: در این آزمایش، تعداد ۳۲۰ قطعه جوجه گوشتی نژاد راس در قالب طرح کاملاً تصادفی مورد بررسی قرار گرفتند. تیمارهای آزمایشی شامل چهار نوع جیره (جیره پایه، جیره با کاهش ۵ درصد پروتئین، جیره با کاهش ۱۰۰ کیلوکالری انرژی متابولیسمی، و جیره با کاهش همزمان ۵ درصد پروتئین و ۱۰۰ کیلوکالری انرژی) در دو سطح عدم مصرف و مصرف مکمل امولسیفایر بود. هر تیمار شامل چهار تکرار و هر تکرار شامل ۱۰ قطعه جوجه بود. پس از پایان دوره پرورش ۴۲ روزه، نمونه‌های بافت عضله ران، عضله سینه، کبد و طحال جمع‌آوری و در نیتروژن مایع منجمد شدند. RNA کل استخراج، cDNA سنتز و بیان ژن UCP3 با استفاده از Real-Time PCR ارزیابی شد. ژن GAPDH به عنوان ژن مرجع مورد استفاده قرار گرفت و تغییرات بیان ژن با روش $2^{-\Delta\Delta Ct}$ محاسبه شد.

نتایج: نتایج نشان داد که مکمل امولسیفایر موجب کاهش بیان نسبی ژن UCP3 در تمامی بافت‌های مورد بررسی گردید. در عضله ران، کاهش بیان UCP3 نسبت به گروه شاهد مشاهده شد، اما این تغییر از نظر آماری معنی‌دار نبود ($P>0.05$). در عضله سینه، مصرف امولسیفایر موجب کاهش معنی‌دار بیان ژن UCP3 شد ($P<0.01$). همچنین، در بافت کبد بیشترین کاهش بیان مشاهده شد که از نظر آماری بسیار معنی‌دار بود ($P<0.001$). در مقابل، تغییرات بیان UCP3 در بافت طحال معنی‌دار نبود ($P>0.05$).

نتیجه‌گیری: بر اساس نتایج این مطالعه، مکمل امولسیفایر علاوه بر بهبود احتمالی استفاده از چربی جیره، می‌تواند موجب تعدیل بیان ژن‌های مرتبط با متابولیسم انرژی در بافت‌های مختلف جوجه‌های گوشتی شود. کاهش بیان UCP3 در عضله سینه و کبد احتمالاً نشان‌دهنده بهبود وضعیت انرژی سلولی، کاهش فشار متابولیکی میتوکندری و کاهش نیاز به فعال شدن مسیرهای سازگاری مرتبط با اکسیداسیون اسیدهای چرب است. این یافته‌ها اهمیت بررسی پاسخ‌های مولکولی ناشی از افزودنی‌های تغذیه‌ای را در کنار شاخص‌های عملکردی نشان می‌دهد و می‌تواند به توسعه راهبردهای دقیق‌تر تغذیه‌ای در صنعت طیور کمک کند.

کلمات کلیدی: امولسیفایر، بیان ژن، متابولیسم انرژی، Real-Time PCR، UCP3

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