Turkish Journal of Agriculture - Food Science and Technology, 10(6): 1142-1148, 2022
DOI: https://doi.org/10.24925/turjaf.v10i6.1142-1148.4965

Turkish Journal of Agriculture - Food Science and Technology

Available online, ISSN: 2148-127X | www.agrifoodscience.com | Turkish Science and Technology Publishing (TURSTEP)

Administration of Branched-Chain Amino Acids in the Pre- or Post-Hatch Period Improves the Fiber Characteristics of *Pectoralis major* Muscle in Turkey Poults Subjected to Early or Delayed Feeding

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A R T I C L E I N F O

Research Article

Received : 09/12/2021
Accepted : 29/06/2022

Keywords:
Amino acid
Poult breast
Muscle fiber type
In ovo injection
Post-hatch feeding

A B S T R A C T

Poultry meat quality is controlled by muscle fiber properties associated with body weight gain in the immediate pre- or post-hatch period. This study aimed to determine the effect of administration of branched-chain amino acids blend (BCAAb, 3 L-leucine:1 L-isoleucine:2 L-valine) in the pre- or post-hatch period on the growth performance and fiber characteristics of the *Pectoralis major* (PM) muscle in turkey poults subjected to early or delayed feeding. Newly hatched poults from eggs injected *in ovo* with BCAAb or received no injection were used in a 21-d study. Poults (n=192) produced without injection of BCAAb received a starter diet supplemented (BCAA) or not (C) with the BCAAb (2 g/kg) immediately or with a delay of 48 h (48BCAA and 48C) after hatching. Poults (n=36) produced with (IOBCAA) *in ovo* injection of BCAAb (2 mg /ml) received the diet and water immediately. The BCAA and IOBCAA poults had higher body weight than those of the C, 48C, and 48BCAA birds. The body weight of the C poults was higher than those of the 48C and 48BCAA birds. The BCAA poults had higher PM muscle weight than those of the C and 48C poults, while that of the IOBCAA poults was higher than that of the 48C poults. The IOBCAA treatment increased the protein content of the PM muscle meat, compared with the C and 48C treatments. The 48C treatment increased the type IIB fiber area and the type I and type IIA fiber percentages but decreased the type IIB fiber, compared with other treatments. The numbers of type IIB and total fibers in the IOBCAA poults were higher than those of the 48C birds. In conclusion, administering BCAAb in ovo or in a starter diet enhanced early growth performance and improved the fiber characteristics of the PM muscle in turkey poults.

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Introduction

In poultry production systems, the embryonic and immediate post-hatch period (the first week before or after hatch) is a critical phase not only for achieving a high poultry performance, but also for overall growth and morphologicy of the *Pectoralis major* (PM) muscle (Harthan et al., 2014; Reed et al., 2017; Ghanaatparast-Rashti et al., 2018; Velleman et al., 2019a; Payne et al., 2020). Indeed, delayed feeding and watering (up to 72 h) due to transportation from the hatchery to the rearing farms result in a decrease in body and organ weights at placement, as well as a reduction in post-hatch growth (Willemsen et al., 2010; Proszkowiec-Weglarzet al., 2019; Kop-Bozbay and Ocak, 2020); this subsequently, leads to a decline in skeletal muscle development and biochemical, biophysical, histochemical and histological muscle fiber characteristics (Chodová and Tůmová, 2017; Payne et al., 2020). Therefore, there is an increasing demand for new feeding strategies for commercial meat-type poultry, characterized by increased breast muscle, resulting from enhancing muscle characteristics. Early nutrition strategies, such as *in ovo* feeding (IOF) and post-hatch feeding of certain specific supplements (egg protein, amino acids blend, and β-hydroxy-β-methyl butyrate) are among the most considerable research on body weight (BW) gain, feed-nutrient utilization and muscle fibre characteristics (Ghanaatparast-Rashti et al., 2018; Kop-Bozbay and Ocak, 2019; Kop-Bozbay and Ocak, 2020; Ospina-Rojas et al., 2019; Lee et al., 2020; Ma et al., 2020). In these studies, the supplements have been promoted early growth performance and, also, improved fiber characteristics of the PM muscle but with contradictory results.

Skeletal muscle, the major food product, comprises the largest proportion of animal mass. Muscle fibers, which are highly specialized cells acting as the structural units of skeletal muscle tissue, are the major components of muscles (Tůmová and Teimouri, 2009; Choi et al., 2013; Chodová and Tůmová, 2017). The muscle weight or mass is a function of the total number and cross-sectional area of muscle fibers that have a critical role in meat quality (Tůmová and Teimouri, 2009; Choi et al., 2013). Poult growth immediately after the hatch induces greater muscle weight simultaneously by increasing the fiber size, density, and number (Willemsen et al., 2010). In addition, compared with older satellite cells, satellite cell proliferation or mitotic activity is more responsive to nutrition at the first week after the hatch (Harthan et al., 2014; Velleman et al., 2019a; Ma et al., 2020). Therefore, adequate early nutrition programming is undoubtedly key for skeletal muscle growth and fiber features.

Previously, it has been determined that a blend composed of leucine, isoleucine and valine at a 3:1:2 ratio (hereafter BCAAb) promoted the embryonic and early neonatal development of turkey poults by enhancing skeletal muscle development (Kop-Bozbay and Ocak, 2020). These observations indicate that there is insufficiency or imbalance in terms of these essential amino acids in the eggs of breeding turkeys (Kop-Bozbay and Ocak, 2020; Ma et al., 2020). The processes of muscle fiber hypertrophy and hyperplasia (increase in size and number, respectively) that impacted the development of skeletal muscle occur in two different phases throughout embryonic and post-hatch periods (Velleman et al., 2018). Some feeding applications during the embryonic and immediate post-hatch period may program these processes (Harthan et al., 2014; Velleman et al., 2019a). Delayed feeding for 48 h usually caused significant decreases in BW or muscle weight in the starter period despite a compensatory growth in later phases (Velleman et al., 2018; Kop-Bozbay and Ocak, 2020). In poultry, the appearance and eating qualities of meat are controlled by the histological (total number of fibers, cross-sectional area of fibers, and fiber type composition) and biochemical (or nutrient composition) muscle fiber characteristics associated with BW gain in the immediate pre- or post-hatch periods (Ismail and Joo, 2017). However, it is unclear how to administe a blend of branched-chain amino acids in these periods impacts the early growth performance and subsequent muscle fibre characteristics of the PM muscle in turkey poults. Accordingly, this research aimed to determine the effect of feeding the BCAAb in the immediate pre- (IOF) or post-hatch period on the growth performance and fiber characteristics of the PM muscle in turkey poults subjected to early or delayed feeding.

Materials and Methods

Incubation, Preparation of BCAAb, the IOF procedure and Diet

The preparation of the solution of BCAAb and the procedures of IOF and incubation were described in detail by Kop-Bozbay and Ocak (2019). In brief, on day 22 of the incubation, a 1.5 ml sterile solution with 0.9% (saline of 0.0135 mg) salt (positive control) or 0.2% (BCAAb of 0.003 mg) BCAAb (*in ovo* BCAA) were injected into the amniotic fluids of the embryonated eggs or were not injected (negative control). The 2 g BCAAb contained 1.00 g L-leucine (Cas no: 61-90-5), 0.33 g L-isoleucine (Cas no: 72-18-4), and 0.67 g L-valine (Cas no: 73-32-5) per L of IOF solution (Kop-Bozbay and Ocak, 2020) and kg of the starter diet (Kop-Bozbay and Ocak, 2019). In preparation for the diet supplemented with the BCAAb, the BCAAb of 2 g was initially sprinkled onto a small amount (approximately 23 g) of feed. Then, this premix was mixed with a feed of 250 g. The last mixture obtained was homogeneously mixed with a feed of 750 g to achieve the desired final concentration.

Bird Housing, Treatments, and Experimental Design

Newly hatched poults from eggs injected in ovo with BCAAb (n=36 hatchlings) or received no injection (n=192 hatchlings) were used in a 21-d study. Since there is no statistical difference between the positive and negative control treatments regarding the properties examined, their poults were combined. Thus, no control group was composed of drilled and not inoculated eggs in the study. Then, the half of poults produced without injection of BCAAb received a commercial starter diet (based on corn-soybean meal) supplemented (BCAA) or not supplemented (C) with the BCAAb (2 g/kg) and water immediately after hatching. The other half were subjected to a 48 h delayed access to the BCAA (48BCAA) or C (48C) diets to mimic commercial hatchery settings and operations after hatching. Poults produced with (IOBCAA) in ovo injection of BCAAb (2 mg /ml) received the starter diet and water immediately after hatching.

The IOBCAA treatments had three replicates, each including 12 birds due to some limitations, such as a low hatchability rate and multiple uses to determine the embryonic muscle development and post-hatch poult quality of the IOF poults (Kop-Bozbay and Ocak, 2020). The other treatments (C, BCAA, 48C and 48BCAA) had four replicates, each including 12 poults that were selected equally (i.e., weight and number) from the negative and positive control treatments that did not entail injection of the eggs.

All poults were raised in pens (1.5 × 1.75 m) furnished with wood shavings of 5 cm as litter in an experimental facility. Each pen was equipped with a bulb of 125-watt, automatic drinker, and red circular poultry feeder. The experimental facility's indoor temperature was maintained at 32±1°C during the first week and gradually decreased to 27±1°C by third week and relative humidity was maintained within a range of 60-70%, using the thermostatic heater and natural ventilation. In the experiment performed at similar management practices, with the exception of feeding time and diet, the turkey starter diet in mush form was formulated, taking into account the management guidelines (https://www.hybridturkeys.com accessed on 5 June 2014) for Hybrid Converter turkeys (Table 1). Although the feed and water were provided *ad libitum*, because the 48C and 48 BCAA poults achieved the diet 48 h later, they consumed the feed for only 19 days.

To determine BW and feed intake (FI) and subsequent feed conversion ratio (FCR, g feed/g BW gain), poults and feed were weighed at placement and 21-d old. On 21-d of age, after feed withdrawal of 10 h, one male and one female poult (six and eight poults per IOBCAA and other treatments, respectively) with the nearest BW to the mean pen mass were slaughtered by neck cut. After exsanguinated, the slaughtered poults were manually plucked and their abdominal cavity was eviscerated carefully (Verdiglione and Cassandro, 2013). To determine the biochemical, biophysical, histochemical, and histological characteristics of the PM muscle, 15 min post-mortem, the breast samples of the air chilled-carcasses were taken (Verdiglione and Cassandro, 2013; Koomkrong et al., 2015). The PM muscle meats excised from the right and left breasts were frozen in liquid nitrogen, and then, stored at –80°C until the histochemical and biochemical analyses of the muscle fibres took place (Verdiglione and Cassandro, 2013).

Table 1. Ingredients and calculated composition of the experimental diet (as-fed basis)

Ingredients	g/kg	Nutrient levels	g/kg
Maize	386.0	Metabolizable energy (MJ/kg)	12.0
Soybean meal (CP 48%)	240.0	Crude protein (CP)	265.0
Sunflower meal (CP 36%)	96.3	Ether extract	34.6
Fish meal (CP 65%)	60.0	Lysine	11.4
Full-fat soybean	100.0	Methionine	2.2
Wheat bran	100.0	Methionine + Cystine	9.2
Vegetable oil	5.0	Threonine	10.1
Limestone	6.1	Tryptophan	2.9
Dicalcium phosphate, 18%	2.6	Arginine	18.1
Sodium chloride	1.5	Histidine	6.5
Vitamin–mineral premix[1]	2.5	Phenylalanine	12.1
		Leucine[2]	21.0 (22.00)
		Isoleucine	11.4 (11.73)
		Valine	12.9 (13.57)
		Calcium	10.6
		Available phosphorus	7.0

[1]Supplied per kilogram of feed: Retinyl acetate, 12 000 IU; cholecalciferol, 0.125 mg; vitamin E (DL-a-tocopheryl acetate), 100 mg; vitamin K (menadione), 4 mg; thiamine, 3 mg; riboflavin, 8 mg, D-Pantotenat, 15 mg; niacin, 50 mg; pyridoxine, 4 mg; folic acid, 2 mg; cobalamin, 0.015 mg; D-biotin, 0.25 mg; choline, 200 mg; Fe, 50 mg; Cu, 15 mg; Mn, 120 mg; Zn 100 mg; Co, 0.2 mg, I, 1.5 mg; Se, 0.3 mg. [2]Values in parentheses are the leucine, isoleucine, and valine contents of the starter diet with BCAAb (2 g/kg).

Histochemical and Biochemical Analysis

Before the analyses, the PM muscle samples were transferred to the cryostat chamber (Cryotome E; Thermo Electron Corporation, Cheshire, UK) at –20°C for a thermal adaptation of 40 min. Four mid-belly sections at a thickness of 10 μm were prepared using the cryostat microtome (Chodová and Tůmová, 2017). The myosin ATPase staining technique was used to determine the muscle fiber types and numbers in these mid-belly sections, as described by Sen et al. (2016). The cross-sectional muscle areas in each mid-belly section were photographed with a microscope with 10x eyepiece (Nikon Eclipse E600, Nikon Corporation, Tokyo, Japan), connected to an image capture system (Clemex, Vision Lite, Longueuil, Canada). The number and areas of muscle fiber types classified as type I (slow-twitch, oxidative, red), type IIA (fast-twitch oxide-glycolytic, intermediate) and type IIB (fast-twitch, glycolytic, white) (Mehmood and Zhang, 2020) were determined using image Processing-Analysis Software (Laica Q Win V3.4). Then, the area of each fiber type (μm^2), fiber density (number/1 mm^2 of the area) and number (%) were computed (Verdiglione and Cassandro, 2013; Koomkrong et al., 2015).

The breast meat samples, excised from the right breast, were used to analyze the biochemical properties. After these samples were mashed, dry matter (DM), ash, crude protein (CP), and ether extract (EE) contents of the samples were analysed using the chemical methods (method mumbers: 930.15, 942.15, 990.03, and 920.39, respectively) approved by the AOAC International (AOAC, 2007). Chemical composition results performed in triplicate were expressed on a wet basis.

Statistical Analysis

The analysis of unbalanced data within a randomized design was performed for the mixed model using residual (or restricted) maximum likelihood (REML) analysis of the Statistical Package for the Social Sciences (IBM SPSS Statistics, version 21.0). For growth performance and the characteristics of the muscle fibres, the experimental unit was the pen mean and the breast sample, respectively. Tukey's multiple comparison test was used to separate statistically different means and any $P \leq 0.05$ was deemed significant in all cases. At the time of hatching, the poult weights were used as the covariates because the initial BW was biased in favor of the IOBCAA treatment. The distribution type (normal, β, or Poisson) of the response variable was included in the REML model statement to compensate for the skewed distribution of each fiber and total fiber numbers in the muscle area. The muscle fiber percentage were submitted to arcsine square root transformation prior to analysis $[(\% \text{ data}/100)+0.05]^{0.5}$.

Results

No mortality in any of the treatment groups throughout the experiment was observed. Table 2 shows the BW, FI, and FCR results. The BCAA and IOBCAA poults had higher BW than the C, 48C, and 48BCAA birds ($P<0.05$). The BW of the C poults was higher than those in the 48C and 48BCAA groups ($P<0.05$). The 48C and 48BCAA poults consumed less feed than the poults undergoing other treatments ($P<0.05$). The FCR in all treatments was similar.

The biophysical, histological, and chemical composition of the PM muscle fibers in turkey poults, fed with the BCAAb in the immediate pre-and post-hatch periods, are presented in Table 3. The PM muscles had a unique distribution of the fiber types (i.e., type I, IIA, and IIB). The BCAA poults had a higher PM muscle weight compared to the C and 48C poults (P<0.05). The IOBCAA poults had higher PM muscle weight than that of the 48C poults (P<0.05). The IOBCAA treatment increased the protein content of PM muscle compared whit the C and 48C treatments (P<0.05). By comparison with other treatments, type IIB fiber area and the percentages of type I and IIA fibers were higher in the 48C poults, but the corresponding values of type IIB fiber were lower (P< 0.05). In terms of type IIB and total fiber numbers, the IOBCAA poults showed higher values than the C48 birds (P<0.05).

Table 2. The body weight (BW), feed intake (FI), and feed conversion ratio (FCR) of turkey poults fed on the starter diet with the leucine, isoleucine, and valine blend (BBCAb) at a 3:1:2 ratio in the immediate pre-and post-hatch periods

Parameters[1]	C[2]	BCAA	48C	48BCA	IOBCAA	SEM	P values
BW (g/bird)							
At hatch	53.9[b]	53.6[b]	53.7[b]	53.6[b]	61.4[a]	1.03	0.030
At d 21	502.6[b]	546.8[a]	479.6[c]	478.5[c]	536.0[a]	10.72	0.051
FI (g/poult)	778.2[a]	782.4[a]	728.2[b]	709.8[b]	791.8[a]	13.04	0.015
FCR (g feed:g gain)	1.72	1.74	1.71	1.66	1.67	0.024	0.648

[a,b,c]Within a row, means with different superscripts differ (P<0.05); [1]Means represent three and four pens of 12 poults per IOBCAA treatment and other treatments, respectively; [2]Treatment: C, received the starter diet without BCAAb and water immediately after hatching; BCAA, received the starter diet supplemented with the BCAAb (2 g/kg) and water immediately after hatching; 48C, subjected to 48 h delayed access to the C diet; 48BCAA, subjected to 48 h delayed access to BCAA diet; IOBCAA, received the C diet and water immediately after hatched from eggs injected with BCAAb (2 g/L); SEM, Standard error of the mean;

Table 3. The relative weight of the *Pectoralis major* (PM) mascle, the meat chemical composition and the muscle histological characteristics of the right and left PM, respectively in turkey poults fed on the starter diet with the leucine, isoleucine, and valine blend (BBCAb) at a 3:1:2 ratio in the immediate pre-and post-hatch periods

Parameters[1]	C[2]	BCAA	48C	48BCAA	IOBCAA	SEM	P values
PM weight (%)	40.5[b]	48.5[a]	37.3[c]	42.0b[c]	45.2[ab]	0.13	0.048
Chemical composition (%)							
Dry matter	24.84[b]	26.38[ab]	25.51[ab]	26.06[ab]	27.26[a]	0.231	0.014
Protein	22.97[b]	24.40[ab]	23.16[b]	24.02[ab]	25.30[a]	0.239	0.013
Ether extract	0.17	0.18	0.15	0.21	0.11	0.019	0.213
Ash	1.38	1.41	1.41	1.31	1.47	0.020	0.546
Histological characteristics Fibre cross-sectional area (μm^2)							
Type I	167.7	128.9	173.3	97.0	173.2	10.27	0.053
Type IIA	43.6	39.7	46.4	35.5	44.3	3.24	0.861
Type IIB	7.8[b]	8.3[b]	14.0[a]	9.3[b]	6.3[b]	0.65	<0.001
Fiber density (number/mm^2)							
Type I	70.5	81.9	64.8	123.6	61.9	7.97	0.078
Type IIA	265.4	271.5	342.9	372.6	232.5	30.01	0.581
Type IIB	1301.6[ab]	1255.6[ab]	755.8[b]	1290.6[ab]	1688.8[a]	87.67	0.011
Total	1637.5[ab]	1609.0[ab]	1163.5[b]	1786.8[ab]	1983.2[a]	88.91	0.040
Muscle fiber number percentage (%)							
Type I	2.9[b]	3.4[b]	5.0[a]	3.0[b]	3.1[b]	0.18	<0.001
Type IIA	9.8[b]	11.9[b]	16.8[a]	12.7[ab]	11.8[b]	0.66	0.006
Type IIB	87.3[a]	84.7[a]	78.2[b]	84.3[a]	85.1[a]	0.78	0.001

[a,b,c]Within a row, means with different superscripts differ (P<0.05); [1]Means represent six and egith poults per the IOBCAA treatment and other treatments, respectively; [2]Treatment: C, received the starter diet without BCAAb and water immediately after hatching; BCAA, received the starter diet supplemented with the BCAAb (2 g/kg) and water immediately after hatching; 48C, subjected to 48 h delayed access to the C diet; 48BCAA, subjected to 48 h delayed access to BCAA diet; IOBCAA, received the C diet and water immediately after hatched from eggs injected with BCAAb (2 g/L); SEM, Standard error of the mean.

Discussion

The present study results indicated that delayed feeding for 48 h without the BCAAb supplementation in the starter diet resulted in suppressed BW and FI, as well as delayed the development and myofiber of PM muscle in the turkey poults, compared with the BCAAb administration *in ovo* or in the starter diet. The delaying fed poults weighed significantly less in body and muscle than early fed poults, without affecting the FCR, as also revealed previously (Powell et al., 2013; Velleman et al., 2018; Payne et al., 2019). In our previous (Kop-Bozbay and Ocak, 2020) and the current study, for the delays in the BW and muscle weight gains caused by the feed withdrawal period, no compensatory growth was observed during the starter phase. This may be probably because i) the ability to synthesize protein within the PM muscle decreased (Powell et al., 2016; Payne et al., 2019), ii) the PM muscle is utilized as amino acid and energy source (Kornasio et al., 2019; Tonniges and Velleman, 2019), and iii) ontogenic gene expression in the PM muscle is negatively affected (Payne et al., 2020) in the unfed poults within 48 h after hatching. Indeed, a feed withdrawal of 48 h during the post-hatch period has been impaired growth rate and muscle development by suppressing the ontogenic expression of transcripts that arrange growth, metabolism, and protein synthesis (Payne et al., 2019, 2020). Because there must be an increase in protein synthesis for post-hatch muscle growth to occur, an imbalance between protein anabolism and catabolism in the muscle has led to a diminution of muscle tissue (Velleman et al., 2019b; Payne et al., 2020). In addition, the delayed feeding has been modified satellite cell differentiation and proliferation (Powell et al., 2013; Payne et al., 2019), which is critical during the first week of life (Harthan et al., 2014; Velleman et al., 2019a). The results of our study supported this notion due to the delayed feeding decreased the protein content of the PM muscle and negatively impacted its fiber characteristics. Therefore, our results confirmed that the first week after hatching is a crucial time frame for the growth of poults or chicks and the development of the PM muscle (Harthan et al., 2014; Velleman et al., 2018, Velleman et al., 2019a; Ma et al., 2020).

As is well known, the IOF is a technique of administering exogenous nutrients into the avian embryo and thereby improves the growth performance of poultry by enhancing egg nutrition during the post-hatch period (Jha et al., 2019; Kop-Bozbay and Ocak, 2019). Indeed, we determined that the IOF of the BCAAb decreased hatchability (76.7% versus 84.6% and 84.3%) but increased the hatching weight (61.8 g versus 54.3 g and 53.4 g) by improving the development of skeletal muscle (1.38 g/100 g BW versus 1.01 and 0.99 g/100 g BW) compared with the positive and negative control groups (Kop-Bozbay and Ocak, 2019). Although a low hatchability means loss, this result indicated that the IOF of the BCAAb has the potential to meet the needs of poultry producers for consistently high-quality poults. The BCAAb administration *in ovo* or in the feed had a similar effect on the studied parameters. The results of the present study indicated that the administering BCAAb *in ovo* or in the starter diet enhanced the early growth performance, promoted the development of skeletal muscles in turkey poults until 21-d old and subsequently, improved the studied fiber characteristics of the PM muscle. Leucine, isoleucine and valine (individually or as a combination) are involved in protein synthesis, energy homeostasis, and lipid metabolism in the PM muscle (Zhang et al., 2017), intestinal histomorphology, and required for optimized growth (Corzo et al., 2010; Ospina-Rojas et al., 2017; Ospina-Rojas et al., 2020). Furthermore, leucine is another essential amino acid that initiated translation for protein synthesis in skeletal muscle and acted as a substrate for producing other amino acids (Payne et al., 2019). The results of our previous study have indicated that the BCAAb promoted post-hatch development in the unfed turkey poults within 48 h or 72 h after hatching (Kop-Bozbay and Ocak, 2020). The improved growth in response to BCAAb can be related to enhanced muscle growth because an enlargement in the histological characteristics of muscle fibers improved the muscle mass (Ma et al., 2020). These notions can be explained why BCAA and IOBCAA poults that had the higher muscle protein content had a greater PM weight, as also reported previously (Corzo et al., 2010; Ospina-Rojas et al., 2020; Payne et al., 2020). However, if there was also a group composed of the *in ovo* BCAA poults subjected to a 48 h delayed access to the stater diet (i.e., 48IOBCAA), the effects of the BCAAb on muscle development of turkey could be better explained or more convincing. Unfortunately, it was impossible to establish such a group in this study due to due to the limitations mentioned above. The BCAAb used in the present study might help develop new diets, which could minimize retardation of the BW gain and, especially, the muscle development in the poults subjected to delayed feedings. Therefore, the BCAA, 48BCAA, and IOBCAA treatments may be one of innovative approaches in the early feeding practices for producing high-quality poult, and consequently profitable meat-type poultry production (Kornasio et al., 2019). Also, our results confirmed that the nutritional status of birds in the immediate post-hatch period is an important trimmer of muscle growth and metabolism (Tonniges and Velleman, 2019; Payne et al., 2020).

A notable suggestion of our study is that IOBCAA and BCAA improved histochemical muscle fibre characteristics during the early post-hatch period. Theoretically, this may lead to an increase in meat yield until marketing (Ma et al., 2020). However, the present study was not conducted over a sufficiently long period to confirm this recommendation. The improvement in the histochemical fiber characteristics of the PM muscle indicated that BCAAb promotes post-hatch muscle hypertrophy in turkeys. The branched-chain amino acids, especially leucine and β-hydroxy-β-methyl butyrate, a metabolite of leucine, promoted muscle cell growth, by stimulating muscle protein (Ospina-Rojas et al., 2020), as reported herein. The administering of BCAAb in the starter diet was very important because they are positioned among limiting amino acids (Corzo et al., 2010; Kidd et al., 2021; Maynard et al., 2021). However, there are conflicting effects of branched-chain amino acids on breast meat yield (Mejia et al., 2011; Ospina-Rojas et al., 2017; Kop-Bozbay and Ocak, 2020). These variable results may be attributed to differences (such as strain, age, and growth rate) in poultry, the leucine, isoleucine, and valine levels, experimental diets, access time to feed, the feeding period, and the management process (Ospina-Rojas et al., 2019; Kop-Bozbay and Ocak, 2020; Lee et al., 2020).

The number, cross-sectional area, and density of muscle fibers are greatly variable in both poultry species and poultry breeds (Choi et al., 2013; Sen et al., 2016; Chodová and Tůmová, 2017). However, PM muscle fibers in most commercial poultry typically are composed of type IIB i.e., fast-twitch, glycolytic (white) fibers (Tůmová and Teimouri, 2009; Chodová and Tůmová, 2017; Velleman et al., 2018). The present study attained similar findings. The composition of the muscle fiber types is very important for determining fresh turkey meat quality traits (Velleman et al., 2019b). The selection to enhance the growth rate and breast meat yield has led to a shift toward type IIB rather than type I muscle fibers in poultry (Tůmová and Teimouri, 2009). Although the PM muscle of the young poultry comprises neonatal, transforming, and adult muscle fiber profiles, the percentage of type I (i.e., dark) fibers in the neonatal muscle is greater than those of transforming and adult muscle profiles (Rosser et al., 2000; Choi et al., 2013). As such, at 21-d old, the poult PM muscles probably exhibited large lighter (intermediate dark and white) fiber profiles, as well as a greater dark (red) fiber profile, i.e., type I fiber. The findings concerning the histochemical fiber characteristics are in accordance with the results relating to many skeletal muscles in the poultry (Choi and Kim, 2009; Chodová and Tůmová, 2017; Zhang et al., 2017) and supported the idea that the different PM muscle weights exhibited different histochemical fiber characteristics (Choi et al., 2013).

In turkeys, an increase in fiber cross-sectional area versus fiber density indicates the increase of fiber size, which has deleterious effects on meat quality (Velleman et al., 2018; Mehmood and Zhang, 2020). This situation shows that the 48C treatment might be reduced meat quality due to its impacts on the cross-sectional area and density of the muscle fibers. Furthermore, Ismail and Joo (2017) stated that the increased BW and muscle weight diminished type I and IIA fibers, whereas augmented type IIB fiber, as also revealed herein. Also, skeletal muscle mass is influenced by muscle fiber size and number (Ismail and Joo, 2017; Payne et al., 2020). As such, our results agree with previous studies that examined the effect of nutrient restriction on the growth and the muscle development of poults in the immediate post-hatch period (Choi et al., 2013; Chodová and Tůmová, 2017; Tonniges and Velleman, 2019; Kop-Bozbay and Ocak, 2020). Based on these findings, it can be said that the total fibre number and the type IIB fibre percentage of the PM muscles from BCAAb-fed poults might positively affect their breast meat yield and quality (Choi et al., 2013; Velleman et al., 2018; Mehmood and Zhang, 2020). The results regarding the histological characteristics of the PM muscle indicated that the BCAAb had a more potent effect on type IIB fiber than on type I and type IIA in the immediate pre-and post-hatch periods. This situation may be related to the fact that type IIB fiber is larger than type IIA in the immediate pre-and post-hatch periods due to the high growth rate of type IIB fibers during these periods (Choi et al., 2013; Zhang et al., 2017). In addition, the changes in the muscle fiber size and numbers depend on the BW gain of young poultry (Choi et al., 2017). These may explain why the BCAAb-fed poults had the increased BW and muscle weight, and in consequence, showed the improved type IIB to type IIA ratio and the enhanced density of type IIB fiber (Choi et al., 2013; Chodová and Tůmová, 2017).

Conclusions

The initiation of BW gain and muscle development in neonatal poults is directly linked to food availability in the immediate pre-and post-hatch periods. The early growth performance of turkey poults and muscle fiber characteristics of the PM muscle meat is positively affected, as a result of the poults being, fed with the BCAAb in the immediate pre-and post-hatch periods. Based on these results, the IOF of the BCAAb or the use of the starter diet with the BCAAb has the potential to enhance the early growth performance of turkey poults and improve the fiber characteristics of the PM muscle. However, to determine the significance of muscle fibers in terms of high performance and meat quality at market age, responses of turkey poults to the diet supplemented with BCAAb should be further studied. In conclution, the BCAAb administration in the starter diet is a better application for turkey poults subjected to early or delayed feeding due to the similar effects of BCAA and IOBCAA treatments on the studied parameters.

Acknowledgements

Research, a part of the PhD Dissertation of the first author, was funded through the Scientific Research Fund of Ondokuz Mayıs University (Grant/Award Number: PYO.ZRT.1904.13.011) and Berk Yem Sanayi and Hasinoglu Tavuk Isletmesi. The authors are grateful for the support of the staff and facilities of the Animal Science Department, Faculty of Agriculture, Ondokuz Mayıs University.

Ethical Committee Report

The Local Ethical Committee of Ondokuz Mayıs University for Experimental Animals ascertained that this study is not an unnecessary repetition of previous experiments and approved the study protocol (Protocol no = 2012/56).

Conflict of Interest

Authors declare that they have no conflict of interests.

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