Research Article

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Carryover effect of direct-fed microbial supplementation and early weaning on the growth performance and carcass characteristics of growing Najdi lambs

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Abstract: Growing Najdi lambs were randomly selected from lambs weaned at 30, 45, and 60 days old which were treated with 3 doses of direct-fed microbial (DFM) at 5, 10, and 15 days old to investigate the carryover effect of early weaning and DFM supplementation on their growth performance and carcass characteristics. Ten lambs from each group were transferred to individual pens for a feeding trial using the total mixed ration (Wafi). Lambs treated with DFM and weaned at 60 days old showed numerically higher body weight and average daily gains compared to other groups. Concerning the carcass and meat quality, there were significant differences between all groups in all carcass and tissue measurements, except for the back-fat and body wall thickness. In conclusion, treatment did not have any significant negative effect on body weight, feed intake, and conversion ratio compared with the control, but positively affected Zn and Cu absorption. DFM also played an important role in fat metabolism, which affects fat deposits in carcasses. The most important finding was that early weaning can be performed using DFM supplementation without any negative effect on the lambs' performance during growth.

Keywords: direct-fed microbial, Najdi lambs, growing period, performance, meat quality

1 Introduction

Increasing the survival rate of newborn lambs from birth up to weaning and the growth rate during growth periods is the most significant factor that affects the profit of sheep farms. The main cause of high mortality rates of newborn lambs is the management system followed by farmers [1]. Proper vaccination, feeding, and other management practices using feed additives also play an important role in newborn health and productivity [2]. Many feed additives have therefore been introduced in the market, such as direct-fed microbial (DFM) supplementation, to improve newborn lambs' health and performance by accelerating rumen and reticulum development, improving the rumen environment, and altering the metabolic activities of rumen microorganisms [3,4].

DFM differs from probiotics in that DFM involves the utilization of naturally available living microbes [5,6]. DFM is a combination of single or mixed cultures of living microbes, primarily lactic acid-producing bacteria, that produce beneficial effects on health by enhancing the gastrointestinal complex microbial balance and keeping a healthy system inside the intestinal tract, making it unsuitable for most of the harmful bacteria [7–10]. The DFM improves animal performance by promoting digestion, increasing feed intake, reducing ruminal problems, and improving animal performance [9]. In addition to boosting ruminant performance, it detoxifies unwanted chemical compounds, strengthens the immune system, preserves stomach peristalsis, and promotes the integrity of intestinal mucous [6,11–14].

Several studies have reported the effect of feeding microbial supplementation, especially yeast, on adult ruminants, and their effects on productivity and general performance [15–18]. However, limited studies with inconsistent findings applied DFM in preruminant

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animals [11,19,20]. According to our pilot study, feeding DFM to newborn lambs increased their growth rate, health status, and general performance, due to the acceleration of forestomach growth and development. Thus, lambs can be weaned early at 30 and 45 days old using DFM supplementation without major negative effects on their performance compared with weaning at 60 days old [21], which eventually will improve the profitability of sheep owners by shortening the lambing interval. As a continuation of the aforementioned work, this study aimed to investigate the carryover effect of early weaning with or without DFM on growth performance, blood metabolites, and carcass characteristics of Najdi lambs during their growth period.

2 Materials and methods

2.1 Animals and management

This experiment was carried out at the Department of Animal Production Farm Al-Ammari, King Saud University, Riyadh region, Saudi Arabia (24°47'57.3"N and 46°33'50.4"E). Ten lambs from each group were randomly selected from lambs weaned at different ages and treated with DFM as follows: control (C) (weaning at 60 days old); T1, DFM (weaning at 30 days old); T2, DFM (weaning at 45 days old); and T3, DFM (weaning at 60 days old). Through the oral route, 3 doses of DFM (5 mL each) were administered to each lamb from the treated groups at 5, 10, and 15 days old. The lambs were then transferred to individual separate pens for the feeding trial, which began in July 2013. A 1-month adaptation period to the new dietary and management system was also performed. Then, Wafi total mixed ration was formulated in the trial phase to feed the growing lambs (Table 1). Lambs were then individually fed the recommended amount of Wafi diet and alfalfa hay as a pellet to cover their nutrient requirements for 12 weeks. Feed intake and refusal were recorded daily and samples from the total mixed ration diet and the refusals were then taken for laboratory analysis [22].

2.2 Sample collection

Body weight (BW) and blood samples of all experimental lambs were taken at the beginning of the study and every 4 weeks until the end of the feeding trial. The blood serum samples were analyzed for glucose, total protein, cholesterol, creatinine, urea-N, and triglycerides as well as concentrations of calcium, phosphorus, zinc, and copper

Table 1: Ingredients and chemical composition of the feeding trial diet (Wafi)

Ingredients	%
Barley	17.0
Feed wheat	29.92
Wheat bran	5
Alfalfa hay	10.05
Palm kernel meal	20
Soya hulls	11.03
Salt	0.47
Limestone	2.58
Molasses	3
Acid buffer ¹	0.80
Commercial premix ²	0.15
Composition	
DM	91.40
CP	13.79
CF	11.98
EE	2.61
Ash	9.09
ME, Mcal/kg ³	2.79
Macro-minerals, (%)	
Calcium	1.7
Phosphorous	0.42
Magnesium	0.28
Potassium	0.75
Micro-minerals, (mg/kg)	
Iron	336
Copper	12.4
Zinc	39.3
Manganese	99.2
Selenium	0.25

¹Neutral buffer derived from seaweed (Celtic Sea Company, Ireland). ²Contained per kg, 10,000 IU vitamin A, 1,000 IU vitamin D, 20 IU vitamin E, 300 mg Mg, 24 mg Cu, 0.6 mg Co, 1.2 mg I, 60 mg Mn, 0.3 mg Se, and 60 mg Zn. ³Based on tabulated values.

using UDICHEM-310 spectrophotometer (semi-automated, United Diagnostic Industry, Dammam, KSA) and commercial kits (Randox Laboratories Ltd, BT, United Kingdom).

At the end of the feeding trial, six lambs from each group were randomly selected and BWs were recorded before slaughtering by severing the jugular vein and carotid artery after a 16 h fast. Empty body and hot and cold carcasses were weighed as well as samples from the head, skin, four feet, lungs, trachea, heart, liver, spleen, kidneys, genital organs, and tail were taken. The *Longissimus dorsi* (LD) muscle was similarly collected from each slaughtered lamb for quality control measurements. The thickness of the back and kidney fat was also measured. In addition, the fat distribution between 9 and 13 ribs was evaluated.

For the chemical analyses, muscle from LD was ground 3 times through a 3 mm screen and mixed after

each grinding. The samples were stored frozen for subsequent chemical analyses according to the Association of Official Analytical Chemists [22].

2.3 Color

The colored values of the CIELAB Color System (1976), L^* (lightness), a^* (redness), and b^* (yellowness), were determined on the LD muscle 20 min after slaughter using a chromameter (Konica Minolta, CR-400-Japan).

2.4 Temperature and pH

The pH and temperature were also measured directly in the muscle immediately after slaughter using a microprocessor pH meter (model pH 211, Hanna Instruments). Two readings were taken and the mean value was calculated for each carcass.

2.5 Cooking loss

The frozen LD muscles were thawed overnight at 4°C. Then, they were placed on a commercial indoor countertop grill and cooked to an internal temperature of 70°C. The temperature was monitored by inserting a thermocouple thermometer probe (Ecoscan Temp JKT, Eutech Instruments) into the geometric center of the muscle. The muscles were then weighed before and after cooking to determine cooking loss as the difference between the initial and final weights.

2.6 Shear force

The cooked samples used to determine cooking loss were used to evaluate the shear force according to the protocol of Wheeler et al. [23]. They were cooled to room temperature (21°C), and then 5 round cores each with diameter of 1.27 cm were removed from each muscle sample parallel to the longitudinal orientation of the muscle fibers. The cores were then obtained using a handheld coring device. Shear force was determined as the maximum force (kg) perpendicular to the fibers using a Texture Analyzer (TA-HD-Stable MicroSystems, England) equipped with a Warner-Bratzler attachment. The crosshead speed was set at 200 mm/min.

2.7 Fat thickness

Fat thickness (rib fat or back fat) was measured (in mm) at a point 75% of the length of the LD muscle from the split chine bone using a digital Vernier caliper.

2.8 Body wall thickness

The body wall thickness measurement (in mm) was taken from the outside of the rib to the outside fat at 4–5 in. below the rib-eye using a digital Vernier caliper.

2.9 Carcass composition

The rack cut (four ribs) was taken and physically dissected into four components: lean, bone, fat, and trimmings (blood vessels, nerves, and connective tissue). These components were estimated as a percentage based on the cut weight.

2.10 Rib-eye area (REA)

The REA was measured in the LD muscle between the 12th and 13th ribs. An acetate sheet was then rubbed over the rib-eye and then peeled off. The rib-eye muscle wetted a certain area of the sheet that was traced with a pen. Then, the area was colored black and scanned (150 dpi Res.), after which it was saved as a monochrome bitmap image for later analysis using the analyzing digital imaging software (AreaScan 2 MFC Application 2000).

2.11 Water-holding capacity (WHC)

The WHC was determined based on the technique described by Wilhelm et al. [24]. Two replicates of around 2 g were collected from the LD muscle of each sample and cut into cubes. Then, the sample was placed between 2 filter papers and two plexiglasses and left under a 10 kg weight for 5 min. Subsequently, the sample was weighed and the WHC was determined as the difference between the initial and final weights.

2.12 Carcass liner measurements

The linear measurements of the carcass were taken after being chilled at 4°C for 24 h before cutting. Measurements were made with a measuring plastic tape (to the nearest

0.5 cm) as follows: carcass length (CL), the distance between the front edge of the pubic bone and the front edge of the first rib at its midpoint; carcass width (CW), the distance between the sternum and the withers; rump width (RW), the maximum width between the trochanters of the femurs; and hind leg length (HLL), the measurement from the inner edge of the proximal end of the medial malleolus of the tibia to the anterior tip of the symphysis pubis.

2.13 Drip loss (DL)

The DL was determined in the LD muscles by weighing the samples 20 min after slaughtering and then kept in plastic bag and left in a cooler (4°C) for 24 and 48 h. Thereafter, the samples were re-weighed to determine the DL values as a percentage based on the initial sample weight.

2.14 Carcass compactness (CC)

CC was determined as a ratio of cold carcass weight to CL as described by Bonvillani *et al.* [25].

2.15 Statistical analysis

Data were subjected to analysis of variance using the General Linear Model procedure of Statistical Analysis System Institute, Inc. (SAS, [26]) for a completely randomized design, according to the following model:

$$Y_{ij} = \mu + C_i + e_{ij},$$

where Y_{ij} = measurement of the variables, μ = overall mean, C_i = effect of the i th treatment (C and 3 treatments), and e_{ij} = residual error.

BW, bodyweight gain (BWG), daily gain (DG), daily feed intake (DFI), feed conversion, biochemical parameters, and mineral concentrations of blood serum, carcass characteristics, and stomach development were dependent variables. The effects of blood sampling time, treatment, and interaction were considered for the biochemical parameters and mineral concentrations of blood serum. Differences among treatment means were detected using the protected least significant difference procedure (PLSD), with $P < 0.05$ considered statistically significant unless otherwise noted.

3 Results

3.1 Growth performance and feed efficiency

Table 2 shows the growth performance and feed efficiency of lambs from the same treated groups during the growing trial to detect the carryover effect of treatment and early weaning. There was no significant effect of DFM treatment of growing lambs on BW, BWG, FI, and feed conversion ratio (FCR) compared to the control. However, lambs from T3 reported a numerically higher final BW, body weight gain, and average daily gain than the other groups.

3.2 Blood serum metabolites and mineral levels

The effect of treatment on the levels of different metabolites in blood serum at different periods of growing lambs is reported in Table 3. There was no significant effect of treatment at different sampling times on glucose, total protein, and creatinine levels at different measured periods, but a significant effect was reported on cholesterol (56 days), urea-N (56 and 84 days), and triglycerides (84 days). Treatment caused a significant increase in cholesterol and triglyceride levels compared to the control group. The same trend was also reported for the urea-N levels.

At the end of the feeding trial, treatment, time, and interaction, only triglyceride levels in the blood serum were affected as a general trend (Table 4). Significantly lower glucose and urea-N levels were detected in the blood serum of lambs from T2 and higher triglyceride levels of lambs from T3 than the other groups (Table 4).

Furthermore, the phosphorus concentration in the blood serum of the control group lambs was significantly lower than those of the lambs in the treated groups (T1, T2, and T3; Table 5) during the growth trial. The same trend was also reported for Zn and Cu. Zinc was significantly lower ($P < 0.0004$) for lambs in the control group than for those in T1 and T3, but not different compared with lambs in T2. A significantly ($P < 0.0003$) higher Cu concentration was also reported in lambs from the treated groups compared to the control lambs. Moreover, the effect of treatment and time of sampling is reported in Table 6. A significant effect of treatment, time, and interaction was observed on Cu and Zn levels in blood serum, which was high compared to lambs from the treated groups (T1, T2, and T3; Table 6).

Table 2: Growth and feed efficiency of growing Najdi lambs during the feeding trial (mean value $\pm$ SE)

Item	Treatment				P-value
	C	T1	T2	T3	
BW (kg)					
BW 1	22.78 $\pm$ 1.59	21.86 $\pm$ 1.56	24.09 $\pm$ 0.99	21.78 $\pm$ 1.15	0.52
BW 28	29.00 $\pm$ 2.04	28.78 $\pm$ 2.41	30.32 $\pm$ 1.49	29.80 $\pm$ 1.39	0.92
BW 56	37.96 $\pm$ 2.46	37.02 $\pm$ 3.13	39.30 $\pm$ 1.96	39.09 $\pm$ 1.72	0.89
BW 84	45.11 $\pm$ 2.35	45.49 $\pm$ 2.94	45.86 $\pm$ 2.24	46.67 $\pm$ 2.08	0.97
BWG (kg)					
1–28	6.21 $\pm$ 0.86	6.69 $\pm$ 1.11	6.84 $\pm$ 0.48	8.02 $\pm$ 0.48	0.34
28–56	8.96 $\pm$ 0.64	9.55 $\pm$ 0.85	9.39 $\pm$ 0.38	9.30 $\pm$ 0.79	0.94
56–84	7.15 $\pm$ 0.50	7.11 $\pm$ 0.31	6.61 $\pm$ 0.64	7.58 $\pm$ 0.88	0.75
1–84	22.33 $\pm$ 1.26	23.35 $\pm$ 1.85	22.84 $\pm$ 1.09	24.89 $\pm$ 1.52	0.59
FI (kg)					
1–28	31.75 $\pm$ 2.16	31.25 $\pm$ 3.19	34.44 $\pm$ 0.85	32.91 $\pm$ 1.26	0.59
28–56	43.96 $\pm$ 2.05	44.95 $\pm$ 2.68	47.77 $\pm$ 0.36	45.14 $\pm$ 1.77	0.38
56–84	50.91 $\pm$ 1.22	51.02 $\pm$ 1.75	52.83 $\pm$ 0.33	51.59 $\pm$ 0.91	0.49
1–84	126.63 $\pm$ 5.24	127.22 $\pm$ 7.57	135.04 $\pm$ 1.25	129.64 $\pm$ 3.82	0.47
DFI (kg)					
1–28	1.13 $\pm$ 0.08	1.12 $\pm$ 0.11	1.23 $\pm$ 0.03	1.18 $\pm$ 0.04	0.58
28–56	1.57 $\pm$ 0.07	1.61 $\pm$ 0.10	1.71 $\pm$ 0.01	1.61 $\pm$ 0.06	0.38
56–84	1.82 $\pm$ 0.04	1.82 $\pm$ 0.06	1.89 $\pm$ 0.01	1.84 $\pm$ 0.03	0.48
ADFI (1–84)	1.51 $\pm$ 0.06	1.52 $\pm$ 0.09	1.61 $\pm$ 0.01	1.54 $\pm$ 0.05	0.46
Daily gain (kg)					
1–28	0.22 $\pm$ 0.03	0.24 $\pm$ 0.04	0.25 $\pm$ 0.02	0.29 $\pm$ 0.02	0.31
28–56	0.32 $\pm$ 0.02	0.34 $\pm$ 0.03	0.34 $\pm$ 0.01	0.33 $\pm$ 0.03	0.91
56–84	0.26 $\pm$ 0.02	0.25 $\pm$ 0.01	0.24 $\pm$ 0.02	0.27 $\pm$ 0.03	0.79
ADG (1–84)	0.27 $\pm$ 0.01	0.28 $\pm$ 0.02	0.27 $\pm$ 0.01	0.30 $\pm$ 0.02	0.51
FCR (intake: gain ratio)					
1–28	6.01 $\pm$ 1.03	5.14 $\pm$ 0.56	5.20 $\pm$ 0.27	4.17 $\pm$ 0.22	0.21
28–56	5.01 $\pm$ 0.29	4.79 $\pm$ 0.30	5.16 $\pm$ 0.20	5.00 $\pm$ 0.26	0.81
56–84	7.42 $\pm$ 0.64	7.25 $\pm$ 0.43	9.17 $\pm$ 1.45	7.62 $\pm$ 1.05	0.56
1–84	5.72 $\pm$ 0.18	5.51 $\pm$ 0.25	6.02 $\pm$ 0.25	5.29 $\pm$ 0.20	0.13

C – Control (weaning at 60 days old). T1 – Weaning at 30 days old with 3 doses of DFM (5, 10, and 15 days old). T2 – Weaning at 45 days old with 3 doses of DFM (5, 10, and 15 days old). T3 – Weaning at 60 days old with 3 doses of DFM (5, 10, and 15 days old). BW, body weight; BWG, body weight gain; ADG, average daily gain; ADFI, average daily feed intake; FI, feed intake; FCR, feed conversion ratio; SE, standard error.

3.3 Carcass characteristics and stomach development

Slaughter components are presented in Table 7. All parameters were not significantly ($P > 0.05$) different between the treatments. However, T2 achieved the highest weight (kg) values for slaughter (53.07), empty body (47.97), hot carcass (26.48), and cold (26.10) carcass. This result was also true regarding the weight (kg) values of the head (2.66), four feet (1.45), lungs and trachea (0.47), liver (0.81), kidneys (0.13), and genital organs (0.65). The control group (C) followed the T2 group in achieving the second-best weight (kg) values for slaughter (52.62) and empty body (47.37) measurements.

The results of the size of the digestive system and the final pH of the rumen fluid are presented in Table 8. All treatment groups did not show significant differences for the parameters tested. The weight of empty stomach and full stomach (full stomach – empty stomach) increased in treated groups compared to the control group. This trend was also noticed in rumen fluid pH, where the C group showed the lowest rumen fluid value (5.69) followed by T3 (5.77), then T2 (5.79), and finally T1 with the highest value (5.85). The results of the intestine measurement showed inconsistencies in their values.

The results of temperature, pH, and color of the LD muscle of growing Najdi lambs are shown in Table 9. Treatment did not show significant differences

Table 3: Blood serum metabolites of growing Najdi lambs during the feeding trial (mean value $\pm$ SE)

Parameters	Treatment				P-value
	C	T1	T2	T3	
Glucose (mg/dL)					
1	61.13 $\pm$ 4.53	73.68 $\pm$ 5.79	70.73 $\pm$ 4.73	64.35 $\pm$ 6.33	0.36
56	64.98 $\pm$ 4.50	66.70 $\pm$ 2.22	74.92 $\pm$ 6.58	67.46 $\pm$ 7.28	0.60
84	86.56 ^a $\pm$ 2.66	88.38 ^a $\pm$ 4.01	73.00 ^b $\pm$ 4.99	83.70 ^{ab} $\pm$ 3.49	0.05
Total protein (g/dL)					
1	6.21 $\pm$ 0.79	6.46 $\pm$ 1.00	7.03 $\pm$ 0.60	6.50 $\pm$ 0.88	0.91
56	5.07 $\pm$ 0.53	6.75 $\pm$ 0.75	5.20 $\pm$ 0.51	6.53 $\pm$ 0.55	0.12
84	5.72 $\pm$ 0.36	5.33 $\pm$ 0.26	5.65 $\pm$ 0.47	4.87 $\pm$ 0.35	0.36
Cholesterol (mg/dL)					
1	36.30 $\pm$ 4.06	49.54 $\pm$ 3.34	48.54 $\pm$ 5.07	43.92 $\pm$ 3.65	0.12
56	37.97 ^b $\pm$ 2.31	53.86 ^a $\pm$ 2.46	62.72 ^a $\pm$ 5.01	51.72 ^a $\pm$ 6.05	0.01
84	51.83 $\pm$ 5.08	57.17 $\pm$ 6.42	54.64 $\pm$ 2.71	55.32 $\pm$ 3.00	0.87
Creatinine (mg/dL)					
1	0.90 $\pm$ 0.10	0.80 $\pm$ 0.08	0.79 $\pm$ 0.05	0.64 $\pm$ 0.07	0.18
56	0.84 $\pm$ 0.05	0.77 $\pm$ 0.04	0.79 $\pm$ 0.02	0.75 $\pm$ 0.04	0.51
84	0.83 $\pm$ 0.03	0.69 $\pm$ 0.07	0.82 $\pm$ 0.04	0.85 $\pm$ 0.04	0.12
Urea nitrogen (mg/dL)					
1	28.18 $\pm$ 3.54	35.62 $\pm$ 3.50	27.02 $\pm$ 3.59	26.82 $\pm$ 5.56	0.40
56	33.68 ^a $\pm$ 3.40	25.62 ^b $\pm$ 2.30	39.80 ^a $\pm$ 3.08	35.10 ^a $\pm$ 1.57	0.01
84	38.08 ^b $\pm$ 2.68	44.70 ^a $\pm$ 3.59	39.30 ^c $\pm$ 0.84	48.13 ^a $\pm$ 2.07	0.02
Triglycerides (mg/dL)					
1	45.53 $\pm$ 2.40	40.72 $\pm$ 3.33	40.13 $\pm$ 2.02	39.53 $\pm$ 2.70	0.38
56	36.63 $\pm$ 2.07	36.30 $\pm$ 1.35	37.80 $\pm$ 1.86	42.85 $\pm$ 3.78	0.23
84	36.94 ^b $\pm$ 3.16	41.28 ^b $\pm$ 3.22	38.22 ^b $\pm$ 2.20	58.58 ^a $\pm$ 3.93	0.0003

C – Control (weaning at 60 days old). T1 – Weaning at 30 days old with 3 doses of DFM (5, 10, and 15 days old). T2 – Weaning at 45 days old with 3 doses of DFM (5, 10, and 15 days old). T3 – Weaning at 60 days old with 3 doses of DFM (5, 10, and 15 days old). SE, standard error. Mean values followed by a common letter are not significantly different by the PLSD test at 95% level of significance.

Table 4: Effect of DFM supplementation on Najdi newborn lambs, sampling time, and interaction with blood serum during the feeding trial

Parameters	Treatment				SEM	Trt	Time	Trt * time
	C	T1	T2	T3				
Glucose (mg/dL)	70.89	76.25	72.88	71.84	2.88	0.58	0.0001	0.14
Total protein (g/dL)	5.67	6.18	5.96	5.97	0.36	0.80	0.04	0.35
Cholesterol (mg/dL)	42.03	53.52	55.30	50.32	2.49	0.002	0.005	0.35
Creatinine (mg/dL)	0.86	0.75	0.80	0.75	0.03	0.08	0.92	0.22
Urea nitrogen (mg/dL)	33.31	35.31	34.71	36.68	1.84	0.63	0.0001	0.004
Triglycerides (mg/dL)	39.70	39.43	38.72	46.99	1.60	0.001	0.03	0.001

C – Control (weaning at 60 days old). T1 – Weaning at 30 days old with 3 doses of DFM (5, 10, and 15 days old). T2 – Weaning at 45 days old with 3 doses of DFM (5, 10, and 15 days old). T3 – Weaning at 60 days old with 3 doses of DFM (5, 10, and 15 days old). SEM, standard error of mean values. Trt, treatment.

($P > 0.05$) between the tested groups with respect to the mentioned parameters. The initial (pH_i) and ultimate (pH_u) pH values were within the normal range. Moreover, the three-color components were increased in the final time (24 h postmortem). T3 attained the highest lightness (L^*) value (39.83) at the initial time (1 h postmortem) followed

by the control group (39.82). In contrast to the results mentioned above, the control group reached the highest value for the L^* component (42.64) at the final time, while T3 reached the lowest value (40.86) for the same period.

The carcass measurement data are displayed in Table 10. These include REA, CC, and carcass linear measurements (CLM).

Table 5: Mineral concentration in blood serum of lambs during growth at different sampling times (mean value $\pm$ SE)

Parameters	Treatment				P-value
	C	T1	T2	T3	
Calcium (mg/dL)					
1	7.89 $\pm$ 0.79	9.01 $\pm$ 0.95	8.98 $\pm$ 0.73	8.02 $\pm$ 0.65	0.63
56	8.57 $\pm$ 0.66	6.64 $\pm$ 0.38	8.29 $\pm$ 0.81	8.14 $\pm$ 1.07	0.32
84	7.24 $\pm$ 0.55	7.89 $\pm$ 0.56	7.07 $\pm$ 0.17	8.53 $\pm$ 0.25	0.09
Phosphorus (mg/dL)					
1	5.75 $\pm$ 0.32	6.28 $\pm$ 0.38	6.46 $\pm$ 0.88	5.69 $\pm$ 0.57	0.73
56	5.28 $\pm$ 0.24	5.69 $\pm$ 0.33	5.91 $\pm$ 0.64	5.28 $\pm$ 0.42	0.67
84	4.18 ^b $\pm$ 0.59	5.44 ^a $\pm$ 0.50	6.11 ^a $\pm$ 0.38	6.71 ^a $\pm$ 0.30	0.01
Zinc (μ g/dL)					
1	69.78 ^b $\pm$ 4.82	147.68 ^a $\pm$ 13.29	120.86 ^a $\pm$ 7.79	133.79 ^a $\pm$ 15.31	0.001
56	140.08 $\pm$ 10.79	146.27 $\pm$ 6.61	157.78 $\pm$ 11.18	130.35 $\pm$ 18.77	0.49
84	110.83 ^b $\pm$ 9.33	154.14 ^a $\pm$ 10.82	124.67 ^b $\pm$ 3.06	167.58 ^a $\pm$ 8.16	0.0004
Copper (μ g/dL)					
1	161.36 $\pm$ 9.85	156.25 $\pm$ 4.72	166.96 $\pm$ 7.64	162.3 $\pm$ 2.85	0.74
56	156.42 ^b $\pm$ 4.11	142.07 ^c $\pm$ 5.26	172.15 ^a $\pm$ 4.35	150.16 ^{bc} $\pm$ 5.47	0.002
84	155.90 ^c $\pm$ 5.99	198.25 ^a $\pm$ 6.68	173.39 ^b $\pm$ 4.19	179.20 ^b $\pm$ 4.74	0.0003

C – Control (weaning at 60 days old). T1 – Weaning at 30 days old with 3 doses of DFM (5, 10, and 15 days old). T2 – Weaning at 45 days old with 3 doses of DFM (5, 10, and 15 days old). T3 – Weaning at 60 days old with 3 doses of DFM (5, 10, and 15 days old). SE, standard error. Mean values followed by a common letter are not significantly different by the Duncan-test at 95% level of significance.

Table 6: Effect of DFM supplementation to Najdi newborn lambs, time of sampling, and interaction with blood serum minerals during the feeding trial

Parameters	Treatment				SEM	Trt	Time	Trt*time
	C	T1	T2	T3				
Calcium (mg/dL)	7.90	7.85	8.11	8.23	0.39	0.89	0.25	0.17
Phosphorus (mg/dL)	5.07	5.80	6.16	5.89	0.29	0.06	0.30	0.17
Zinc (μ g/dL)	106.90	149.36	134.44	143.91	6.26	0.0001	0.003	0.003
Copper (μ g/dL)	157.90	165.52	170.83	163.89	3.33	0.05	0.0001	0.0001

C – Control (weaning at 60 days old). T1 – Weaning at 30 days old with 3 doses of DFM (5, 10, and 15 days old). T2 – Weaning at 45 days old with 3 doses of DFM (5, 10, and 15 days old). T3 – Weaning at 60 days old with 3 doses of DFM (5, 10, and 15 days old). SEM, standard error of mean values. Trt, treatment.

REA and CW were significantly ($P < 0.05$) different between treatments. Alternatively, CC and the rest of the linear measurements and CL, RW, and HLL were not significantly affected. T3 achieved the highest values of 17.12 and 0.39 cm² for REA and CC, respectively. It was noticeable that T2 had gained the best CLM than the other treatment groups. The obtained values for CL, CW, RW, and HLL in this group were 69.33, 32.33, 45.17, and 45.50 cm, respectively.

The results of shear force, WHC, cooking loss, and DL are shown in Table 11. There were no significant differences between the treatments in these parameters. However, the WHC of the meat was improved compared to the control group (C) as weaning days increased. The best WHC was achieved by group T3 (38.72%), followed by

T2 > T1 < C. The tenderness was enhanced in T1 and T2 compared to the C group and T3. Contrary to the expected result based on the WHC results, the cooking loss of T3 was the highest (33.94%), followed by T2 > T1 > C. The DL of T2 was the lowest in all treatment groups for both the 24 h (7.78) and 48 h (9.35) periods. Alternatively, the highest values in 24 h were attained by T1 (8.63) and in 48 h by the C group (10.15).

The carcass composition (meat, bone, fat, and trimmings) of the growing Najdi lambs is presented in Table 12. The rack cut and carcass components were not significantly ($P > 0.05$) different between the treated groups. The T3 group had the highest meat content (43.67%) compared to the other groups, while the T1 group had

Table 7: Slaughter components of DFM-offered growing Najdi lambs weaned at different ages (mean value $\pm$ SE)

Parameter (kg)	Treatment				P-value
	C	T1	T2	T3	
Slaughter wt	52.62 $\pm$ 1.26	49.18 $\pm$ 1.62	53.07 $\pm$ 2.38	48.13 $\pm$ 1.90	0.21
Empty body wt*	47.37 $\pm$ 1.57	44.57 $\pm$ 1.33	47.97 $\pm$ 2.05	43.68 $\pm$ 1.75	0.25
Hot carcass wt	24.05 $\pm$ 1.45	25.22 $\pm$ 0.88	26.48 $\pm$ 1.16	25.70 $\pm$ 0.89	0.75
Cold carcass wt	23.68 $\pm$ 1.44	24.83 $\pm$ 0.87	26.10 $\pm$ 1.16	25.30 $\pm$ 0.89	0.50
Head wt	2.50 $\pm$ 0.12	2.51 $\pm$ 0.08	2.66 $\pm$ 0.09	2.52 $\pm$ 0.11	0.64
Skin wt	4.95 $\pm$ 0.58	4.82 $\pm$ 0.27	4.98 $\pm$ 0.33	5.15 $\pm$ 0.33	0.95
Four feet wt	1.29 $\pm$ 0.08	1.28 $\pm$ 0.04	1.45 $\pm$ 0.10	1.43 $\pm$ 0.07	0.28
Lung and trachea wt	0.43 $\pm$ 0.02	0.45 $\pm$ 0.03	0.47 $\pm$ 0.02	0.45 $\pm$ 0.03	0.76
Heart wt	0.20 $\pm$ 0.01	0.18 $\pm$ 0.01	0.20 $\pm$ 0.01	0.18 $\pm$ 0.01	0.49
Liver wt	0.72 $\pm$ 0.04	0.76 $\pm$ 0.04	0.81 $\pm$ 0.06	0.70 $\pm$ 0.03	0.36
Spleen wt	0.07 $\pm$ 0.01	0.06 $\pm$ 0.002	0.07 $\pm$ 0.01	0.07 $\pm$ 0.004	0.60
Kidneys wt	0.12 $\pm$ 0.01	0.12 $\pm$ 0.01	0.13 $\pm$ 0.01	0.12 $\pm$ 0.01	0.51
Genital organs wt	0.52 $\pm$ 0.06	0.53 $\pm$ 0.04	0.65 $\pm$ 0.08	0.48 $\pm$ 0.05	0.24
Tail wt	1.85 $\pm$ 0.19	2.27 $\pm$ 0.16	2.27 $\pm$ 0.25	2.88 $\pm$ 0.37	0.18

*Empty body wt = (full stomach – empty stomach) + (full intestine – empty intestine) – slaughter/1,000. C – Control (weaning at 60 days old). T1 – Weaning at 30 days old with 3 doses of DFM (5, 10, and 15 days old). T2 – Weaning at 45 days old with 3 doses of DFM (5, 10, and 15 days old). T3 – Weaning at 60 days old with 3 doses of DFM (5, 10, and 15 days old). SE, standard error.

Table 8: Digestive system and final rumen fluid pH of DFM-offered growing Najdi lambs weaned at different ages

Parameter	Treatment				SEM	P-value
	C	T1	T2	T3		
Full stomach (g)	4185.33	4531.33	5154.17	4393.83	404.47	0.39
Empty stomach (g)	1223.67	1265.67	1463.17	1340.83	93.77	0.32
Full intestine (g)	2500.50	2338.67	2686.67	2436.17	149.90	0.31
Empty intestine (g)	1157.17	1099.50	1277.00	1178.70	77.69	0.46
Rumen fluid pH	5.69	5.85	5.79	5.77	0.09	0.66

C – Control (weaning at 60 days old). T1 – Weaning at 30 days old with 3 doses of DFM (5, 10, and 15 days old). T2 – Weaning at 45 days old with 3 doses of DFM (5, 10, and 15 days old). T3 – Weaning at 60 days old with 3 doses of DFM (5, 10, and 15 days old). SEM, standard error of mean values.

the lowest value (39.17%). T1 also achieved the highest fat content (23.17%), followed by T3 (22.17%), C (22.0%), and T2 with the lowest fat content (18.33%).

Table 13 shows the carcass fat deposits of the treatment groups. The treatment groups revealed significant ($P < 0.05$) differences in back-fat and body wall contents. Other fat deposits such as omental, mesentery, pericardial, and kidney knob channel fats did not differ significantly ($P > 0.05$) between the groups. The T2 attained the highest back-fat thickness of 8.72 mm, while the T1 attained the lowest amount of 4.73 mm. The C and T3 groups had got 5.05 and 4.91 mm, respectively. The body wall fat deposit (mm) was as follows: C (5.70) $>$ T2 (5.49) $>$ T1 (4.50) $>$ T3 (2.65).

Alternatively, the chemical composition of the LD muscles is reported in Table 14, which shows only a

significantly higher intramuscular fat content of lambs in the T3 group than in the other groups.

4 Discussion

Microbial feed additives have several objectives, which differ from those reported for non-ruminants. During the preruminant stage of newborn life, commercial benefits of feeding DFM feeds can be achieved by enhancing the rate of beneficial microbial growth in the rumen and reticulum. This feeding method accelerates the onset of weaning without any negative effect on the lamb growth, performance, and health [19]. The main finding of the pilot part of this study was that feeding DFM to newborn

Table 9: Temperature, pH, and color values of LD muscle from DFM-offered growing Najdi lambs weaned at different ages (mean value $\pm$ SE)

Parameter	Treatment				P-value
	C	T1	T2	T3	
Temperature ($^{\circ}\text{C}$)	25.08 $\pm$ 0.24	25.70 $\pm$ 0.37	25.33 $\pm$ 0.14	25.46 $\pm$ 0.31	0.48
pH _i ¹	6.62 $\pm$ 0.09	6.51 $\pm$ 0.09	6.73 $\pm$ 0.11	6.36 $\pm$ 0.11	0.12
pH _u ²	5.79 $\pm$ 0.04	5.85 $\pm$ 0.05	5.73 $\pm$ 0.02	5.82 $\pm$ 0.06	0.27
Color at 1 h postmortem					
L*	39.82 $\pm$ 0.98	38.80 $\pm$ 1.10	39.54 $\pm$ 1.17	39.83 $\pm$ 0.81	0.88
a**	17.02 $\pm$ 0.53	16.95 $\pm$ 1.28	17.37 $\pm$ 0.68	17.23 $\pm$ 0.71	0.98
b***	4.86 $\pm$ 0.29	5.08 $\pm$ 0.50	5.23 $\pm$ 0.17	5.27 $\pm$ 0.30	0.82
Color at 24 h postmortem					
L*	42.64 $\pm$ 0.81	41.74 $\pm$ 1.42	41.39 $\pm$ 0.95	40.86 $\pm$ 0.44	0.62
a**	20.56 $\pm$ 0.60	19.79 $\pm$ 0.72	21.06 $\pm$ 0.28	20.97 $\pm$ 0.92	0.53
b***	10.49 $\pm$ 0.63	10.80 $\pm$ 0.54	12.39 $\pm$ 0.68	11.04 $\pm$ 0.77	0.22

C – Control (weaning at 60 days old). T1 – Weaning at 30 days old with 3 doses of DFM (5, 10, and 15 days old). T2 – Weaning at 45 days old with 3 doses of DFM (5, 10, and 15 days old). T3 – Weaning at 60 days old with 3 doses of DFM (5, 10, and 15 days old). SE, standard error. pH_i¹ – Initial pH and pH_u² – Ultimate pH. L* for lightness, a** for redness, and b*** for yellowness.

lambs improves their growth rate, health status, and general performance, due to faster growth and development of the forestomach [21]. Therefore, lambs can be weaned early at 30 and 45 days using DFM supplementation without a significant negative effect on their performance compared to weaning at 60 days old. This supplementation will increase the profitability of sheep owners by shortening the ewes' lambing interval due to early weaning, and more lambing, without any negative effect on the health and mortality rate of their newborn lambs. Within this part, results of the carryover effect of early weaning and DFM supplementation during newborn early life on their performance during growth in terms of growth performance and carcass quality were discussed.

Several studies have discussed the effect of DFM supplementation on ruminant animal performance. There was

no improvement in daily gain due to feeding *Lactobacilli* and other bacteria as a DFM to animals [27–31]. This lack of effect was also observed when growing goat kids were supplemented with probiotics [20,32,33]. These findings completely agree with our results reported in this study. On the contrary, an improvement in the rate of gain (17%) was obtained when $2.5 \times 1,011$ cfu/day of *L. acidophilus* species was added to milk or as a milk replacement [34]. Similarly, an increase in body weight and an improvement in FCR of growing lambs were detected when probiotics were fed [35]. Moreover, these findings agreed with Theodorou et al. [31] who reported that feeding microbial supplementation based on anaerobic fungi to animals increased feed intake and live-weight gain in calves following weaning. Furthermore, the introduction of yeast culture through stomach tubes to newborn lambs increased

Table 10: REA, CC, and CLM of DFM-offered growing Najdi lambs weaned at different ages (mean value $\pm$ SE)

Parameter	Treatment				P-value
	C	T1	T2	T3	
REA (cm ²)	15.39 $\pm$ 0.93	16.45 $\pm$ 0.42	14.93 $\pm$ 0.75	17.12 $\pm$ 0.42	0.12
CC	0.35 $\pm$ 0.02	0.37 $\pm$ 0.01	0.38 $\pm$ 0.01	0.39 $\pm$ 0.02	0.33
CLM (cm)					
CL	67.50 $\pm$ 1.38	68.17 $\pm$ 1.78	69.33 $\pm$ 1.17	66.00 $\pm$ 1.24	0.43
CW	30.67 $\pm$ 0.67 ^b	29.33 $\pm$ 0.33 ^b	32.33 $\pm$ 0.56 ^a	30.33 $\pm$ 0.33 ^b	0.003
RW	43.00 $\pm$ 0.68	42.67 $\pm$ 1.90	45.17 $\pm$ 0.91	43.33 $\pm$ 0.95	0.48
HLL	43.00 $\pm$ 0.63	43.50 $\pm$ 0.76	45.17 $\pm$ 0.87	43.33 $\pm$ 0.92	0.26

C – Control (weaning at 60 days old). T1 – Weaning at 30 days old with 3 doses of DFM (5, 10, and 15 days old). T2 – Weaning at 45 days old with 3 doses of DFM (5, 10, and 15 days old). T3 – Weaning at 60 days old with 3 doses of DFM (5, 10, and 15 days old). SE, standard error. Mean values followed by a common letter are not significantly different by the PLSD test at 95% level of significance.

Table 11: Shear force, cooking loss, WHC, and DL of LD muscle of DFM-offered growing Najdi lambs weaned at different ages (mean value $\pm$ SE)

Parameter	Treatment				P-value
	C	T1	T2	T3	
Shear force (kg)	4.45 $\pm$ 0.75	4.26 $\pm$ 0.49	3.84 $\pm$ 0.25	4.54 $\pm$ 0.49	0.79
WHC (%)	37.57 $\pm$ 0.51	37.42 $\pm$ 1.74	38.37 $\pm$ 1.67	38.72 $\pm$ 1.15	0.89
Cooking loss (%)	30.28 $\pm$ 1.16	30.46 $\pm$ 1.49	30.64 $\pm$ 1.16	33.94 $\pm$ 0.61	0.13
DL in 24 h	8.48 $\pm$ 2.93	8.63 $\pm$ 1.42	7.78 $\pm$ 1.57	8.15 $\pm$ 2.87	0.94
DL in 48 h	10.15 $\pm$ 2.43	9.72 $\pm$ 2.02	9.35 $\pm$ 1.41	9.75 $\pm$ 2.03	0.94

C – Control (weaning at 60 days old). T1 – Weaning at 30 days old with 3 doses of DFM (5, 10, and 15 days old). T2 – Weaning at 45 days old with 3 doses of DFM (5, 10, and 15 days old). T3 – Weaning at 60 days old with 3 doses of DFM (5, 10, and 15 days old). SE, standard error.

Table 12: Carcass composition (%) of DFM-offered growing Najdi lambs weaned at different ages (mean value $\pm$ SE)

Parameter (%)	Treatment				P-value
	C	T1	T2	T3	
Cut	3.83 $\pm$ 0.17	3.50 $\pm$ 0.22	3.50 $\pm$ 0.22	3.50 $\pm$ 0.34	0.47
Meat	40.17 $\pm$ 1.94	39.17 $\pm$ 2.80	41.67 $\pm$ 1.52	43.67 $\pm$ 1.56	0.61
Bone	29.17 $\pm$ 2.17	26.33 $\pm$ 2.06	29.17 $\pm$ 1.56	25.17 $\pm$ 2.41	0.35
Fat	22.00 $\pm$ 2.59	23.17 $\pm$ 2.73	18.33 $\pm$ 2.04	22.17 $\pm$ 2.47	0.57
Trimming	7.67 $\pm$ 0.88	8.83 $\pm$ 0.60	9.83 $\pm$ 0.75	8.17 $\pm$ 0.70	0.17

C – Control (weaning at 60 days old). T1 – Weaning at 30 days old with 3 doses of DFM (5, 10, and 15 days old). T2 – Weaning at 45 days old with 3 doses of DFM (5, 10, and 15 days old). T3 – Weaning at 60 days old with 3 doses of DFM (5, 10, and 15 days old). SE, standard error.

the number of cellulolytic bacteria and protozoa [36]. This observation was due to an earlier creation of suitable conditions for microbial growth, thus allowing early weaning of lambs, but no measurements were collected regarding the performance of these lambs. A study by Abdelrahman [19] using the same DFM and different doses targeting the same objectives concluded that the use of DFM accelerated the early weaning of newborn lambs (30 and 45 days old) by improving the development of rumen microorganisms,

especially at the 45-day weaning age without any negative effect on their growth and performance compared to weaning at 60 days old. The explanation of our results regarding growth and feed efficiency is that Najdi lambs' DFM supplementation was not performed during the feeding trial, but during early life up to weaning. The most important finding in this study was that early weaning can be performed using DFM supplementation without any negative effect on the lambs' performance during growth.

Table 13: Carcass fat deposits of DFM-offered growing Najdi lambs weaned at different ages (mean value $\pm$ SE)

Parameter	Treatment				P-value
	C	T1	T2	T3	
Back-fat thickness (mm)	5.05 $\pm$ 0.94 ^b	4.73 $\pm$ 0.51 ^b	8.72 $\pm$ 0.56 ^a	4.91 $\pm$ 0.80 ^b	0.002
Body wall (mm)	5.70 $\pm$ 1.03 ^a	4.50 $\pm$ 0.36 ^a	5.49 $\pm$ 0.54 ^a	2.65 $\pm$ 0.13 ^b	0.004
Omental fat (g)	1,121.17 $\pm$ 173.77	964.00 $\pm$ 100.68	1,102.33 $\pm$ 159.61	880.00 $\pm$ 118.29	0.61
Mesentery fat (g)	551.00 $\pm$ 72.16	507.17 $\pm$ 59.74	467.67 $\pm$ 66.38	536.00 $\pm$ 92.33	0.86
Pericardial fat (g)	105.00 $\pm$ 6.45	86.83 $\pm$ 16.39	107.33 $\pm$ 14.05	116.00 $\pm$ 9.40	0.41
KKCF* (g)	805.17 $\pm$ 112.63	754.17 $\pm$ 118.57	743.67 $\pm$ 121.85	714.50 $\pm$ 77.07	0.95

C – Control (weaning at 60 days old). T1 – Weaning at 30 days old with 3 doses of DFM (5, 10, and 15 days old). T2 – Weaning at 45 days old with 3 doses of DFM (5, 10, and 15 days old). T3 – Weaning at 60 days old with 3 doses of DFM (5, 10, and 15 days old). *Kidney knob and channel fat. SE, standard error. Mean values followed by a common letter are not significantly different by the PLSD test at 95% level of significance.

Table 14: Effect of treatment on the meat chemical composition of growing Najdi lambs

Measurements (%)	Treatment				SEM	P-value
	C	T1	T2	T3		
Moisture	66.13	63.01	60.64	63.06	1.30	0.583
Crude protein	20.91	20.07	20.98	21.83	0.31	0.669
Ash	0.977	1.047	1.247	1.32	0.06	0.073
Ether extract	11.79 ^c	17.33 ^b	13.10 ^c	19.79 ^a	1.00	0.0001

C – Control (weaning at 60 days old). T1 – Weaning at 30 days old with 3 doses of DFM (5, 10, and 15 days old). T2 – Weaning at 45 days old with 3 doses of DFM (5, 10, and 15 days old). T3 – Weaning at 60 days old with 3 doses of DFM (5, 10, and 15 days old). SEM, standard error of mean values. Mean values followed by a common letter are not significantly different by the PLSD test at 95% level of significance.

The change in metabolite levels in blood serum reflects the effect of DFM supplementation on nutrient digestibility and absorption. There was no significant effect of treatment at different sampling times on glucose, total protein, and creatinine levels, but a significant effect was reported on cholesterol (56 days), urea-N (56 and 84 days), and triglycerides (84 days). The treatment caused a significant increase in cholesterol and triglyceride levels compared to the control group. This finding disagreed with that of Lubbaddeh et al. [37] who reported a reduction in cholesterol levels of blood serum and meat of growing lambs as a result of *Lactobacillus acidophilus* supplementation. An increase in urea-N was also noticed due to DFM supplementation. This result disagrees with several reports, which suggested an improvement in rumen nitrogen use by supplemented probiotics [38–40].

At the end of the feeding trial, treatment, time, and interaction, only triglyceride levels in the blood serum were affected as a general trend (Table 4). Significantly lower glucose and urea-N levels were also detected in the blood serum of lambs from T2 and higher triglyceride levels of lambs from T3 than others (Table 4). As a general trend, metabolite levels also fell within the recommended levels [41].

Furthermore, the phosphorus concentration in the blood serum of lambs from the control group was significantly lower than lambs from all treated groups (T1, T2, and T3; Table 5) during the growing trial. The same trend was also reported for Zn and Cu levels. Zinc was significantly lower ($P < 0.0004$) for the lambs in the control group than for those in T1 and T3, but not different for the lambs in the T2 group. Alternatively, significantly ($P < 0.0003$) higher Cu concentrations were also reported in lambs from the treated groups compared to control lambs. Moreover, the effect of treatment and time of sampling is reported in Table 6. A significant effect of treatment, time, and interaction on Cu and Zn levels in blood serum was also found to be high for lambs from the

treated groups (T1, T2, and T3; Table 6). A similar observation was reported on the growth of lambs under DFM supplementation and their mineral absorption status, especially Zn [19] as well as with *Lactobacillus* bacterial supplementation [42]. The efficiency of absorption and bioavailability of many minerals affected by many factors is mainly related to diet, rumen, and reticulum fermentation process. The ruminant diet is usually high in fiber, and considerable fiber digestion occurs through the microbial fermentation in the rumen. However, the association of minerals with fiber in feedstuffs [43] or binding of minerals to undigested fractions in the gastrointestinal tract negatively affects the bioavailability of minerals [44]. Furthermore, rumen pH affects the solubility of some mineral complexes formed in the rumen [45]. Due to that, the mineral concentration and other metabolites in the blood are crucial to be determined after using the DFM supplementation because of their effect on nutrient digestibility, availability, and absorbability via the digestive tract [46,47]. These results will help avoid nutrient deficiencies or toxicity. As a general trend, all mineral levels in the blood serum of this study dropped within the normal range [48].

One of the important hypotheses of using DFM is to improve the rate of rumen microbial growth and forestomach development, thus accelerating the onset of weaning, without any negative impact on performance and health. A numerically general improvement in the forestomach capacity was also observed, especially in lambs fed with DFM and weaned at 45 days old when slaughtered after the feeding trial. Results are shown in Table 8. Improvements in the digestive system capacity and development of lambs fed with DFM explain the better performance of animals during these feeding trials as a carryover effect.

There were no significant differences between the groups in all measurements of carcass components, including those of the hot and cold carcass weights, as shown in Table 7.

Furthermore, only a numerically higher weight of the liver, kidney, and genital organs was found in slaughtered lambs from T2 compared with lambs from the other groups. The liver and kidney also play an important role in animal metabolism. The tail weights in lambs from T3 were numerically higher compared to control, T1, and T2, which reflect a higher accumulation of fat in this group. Similarly, no significant effect was detected by taking probiotics in the lungs, heart, and kidney on the weight of growing lambs [49]. In contrast, several studies noticed a significantly higher dressing percentage of growing lambs and kids when fed with probiotics [50,51].

Concerning meat quality, shear force, cooking loss, WHC, and DL of LD muscles did not indicate any significant differences between all groups in the above measurements of meat quality. A numerically higher value for some LD measurements such as cooking loss for lambs from T3 ($33.94 \pm 0.61\%$), DL in 24 h for lambs' meat from T1 ($8.63 \pm 1.42\%$), and DL in 48 h for lambs' meat from C ($10.15 \pm 2.43\%$; Table 11) was observed. Alternatively, the chemical compositions of the LD muscles were significantly higher for the intramuscular fat contents of lambs from the T3 group compared to the other groups. On the contrary, no change in LD muscle fat content was observed as a result of the DFM supplement in growing lambs [19].

The component of rack meat cut (meat, bone, fat, and trimmings) did not show significant differences between all groups, except for a numerically higher percentage of rack cut meat for lambs from the T3 group (43.67%) compared with the control, T1, and T2 (40.17, 39.17, and 41.67%, respectively; Table 12).

The REA (cm^2) for T3 lambs showed numerically higher values than the other groups. No significant differences were reported between the groups in terms of CC, CL, width, and HLL as well (Table 10). Alternatively, the CW (cm) was significantly ($P < 0.003$) wider in lambs from T2 compared with the other groups (Table 10). Unfortunately, no solid research regarding these results was cited to compare with or to justify some of these findings in this experiment, especially the change in fat deposits and intramuscular measurements. Therefore, more studies should be conducted to identify the mechanism of DFM in fermentation and the negative or positive effect on the quality of meat of newborn lambs and the general performance considering many factors that affect the results.

For carcass fat deposits, back-fat thickness (mm) and body wall fat (mm) were the only deposits that significantly differed between the experimental groups (Table 13). The thickness of the back-fat was also significantly ($P < 0.002$) higher in T2 lambs. Significantly lower body

wall fat (mm) for lambs in the T3 group was also observed when compared to others. However, the intramuscular fat content in the meat of lambs from T3 was significantly higher ($P < 0.0001$; 19.79%), followed by the meat of T1 lambs (17.33%) compared to the lambs of control and T2 (11.79 and 13.10%, respectively) as shown in Table 14. Generally, many studies have reported a relationship between probiotic supplementation in ruminants and fat metabolism. An increase in the back-fat thickness in response to probiotic supplementation compared to that of the control groups was noticed [33] and an increase in total body fat percentage due to probiotic supplementation to animals [52] was noticed. One of the most reasonable explanations for this finding is a shift in the rumen fermentation pattern, which changed the concentration and ratios of volatile fatty acids and consequently increased the lipogenesis and distribution of fat within animal tissues [53].

5 Conclusion

At the end of the trial, feeding DFM to the three groups did not have any negative effect on the final BW, BWG, FI, and FCRs compared to the control, only triglyceride levels in the blood serum were affected as a general trend. All mineral levels in the blood serum of this study dropped within the normal range improving the digestive system capacity and development of lambs. Concerning the carcass and meat quality, DFM played an important role in fat metabolism, which affected fat deposits in carcasses. Since a considerable variation in the magnitude of different animal performance responses to commercial feed (DFM) was observed, additional knowledge is required regarding the mechanism of action through which these performance improvements are mediated. Furthermore, identifying and characterizing existing commercial probiotic strains, optimal doses, and needed combinations for a certain animal physiological status are vital to assess and justify their stability throughout feed processing and digestion processes.

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References

- [1] Flinn T, Kleemann DO, Swinbourne AM, Kelly JM, Weaver AC, Walker SK, et al. Neonatal lamb mortality: major risk factors and the potential ameliorative role of melatonin. *J Anim Sci Biotechnol.* 2020;11(1):107.
- [2] Markowiak P, Slizewska K. The role of probiotics, prebiotics and synbiotics in animal nutrition. *Gut Pathog.* 2018;10(1):21.
- [3] Abe F, Ishibashi N, Shimamura S. Effect of administration of bifidobacteria and lactic acid bacteria to newborn calves and piglets. *J Dairy Sci.* 1995;78(12):2838–46.
- [4] Azzaz H, Aziz HA, Farahat ES, Murad H. Impact of microbial feed supplements on the productive performance of lactating nubian goats. *Glob Vet.* 2015;14(4):567–75.
- [5] Kenney N, Vanzant E, Harmon D, McLeod K. Direct-fed microbials containing lactate-producing bacteria influence ruminal fermentation but not lactate utilization in steers fed a high-concentrate diet. *J Anim Sci.* 2015;93(5):2336–48.
- [6] Krehbiel C, Rust S, Zhang G, Gilliland S. Bacterial direct-fed microbials in ruminant diets: performance response and mode of action. *J Anim Sci.* 2003;81(14_suppl_2):E120–32.
- [7] Dawson KA. Manipulating ruminal fermentations: are there natural alternatives to ionophores for beef production. *Biotechnology in the Feed Industry Proceedings of Alltech's 4th Annual Symposium Alltech Technical Publications, Nicholasville, KY, USA; 1988. p. 101–12.*
- [8] Gilliland SE, Nelson CR, Maxwell C. Assimilation of cholesterol by *Lactobacillus acidophilus*. *Appl Env Microbiol.* 1985;49(2):377–81.
- [9] Elghandour MM, Salem AZ, Castañeda JSM, Camacho LM, Kholif AE, Chagoyán JCV. Direct-fed microbes: a tool for improving the utilization of low quality roughages in ruminants. *J Integr Agriculture.* 2015;14(3):526–33.
- [10] Puniya AK, Salem AZ, Kumar S, Dagar SS, Griffith GW, Puniya M, et al. Role of live microbial feed supplements with reference to anaerobic fungi in ruminant productivity: a review. *J Integr Agriculture.* 2015;14(3):550–60.
- [11] McAllister T, Beauchemin K, Alazzez A, Baah J, Teather R, Stanford K. The use of direct fed microbials to mitigate pathogens and enhance production in cattle. *Can J Anim Sci.* 2011;91(2):193–211.
- [12] Jeyanathan J, Martin C, Morgavi D. The use of direct-fed microbials for mitigation of ruminant methane emissions: a review. *Animal.* 2014;8(2):250–61.
- [13] Khan RU, Shabana N, Kuldeep D, Karthik K, Ruchi T, Abdelrahman MM, et al. Direct-fed microbial: beneficial applications, modes of action and prospects as a safe tool for enhancing ruminant production and safeguarding health. *Int J Pharmacol.* 2016;12(3):220–31.
- [14] Seo J-K, Kim S-W, Kim M-H, Upadhaya SD, Kam D-K, Ha J-K. Direct-fed microbials for ruminant animals. *Asian-Australasian J Anim Sci.* 2010;23(12):1657–67.
- [15] Retta KS. Role of probiotics in rumen fermentation and animal performance: a review. *Int J Livest Prod.* 2016;7(5):24–32.
- [16] Ezema C. Probiotics in animal production: a review. *J Veterinary Med Anim Health.* 2013;5(11):308–16.
- [17] Ghazanfar S, Khalid N, Ahmed I, Imran M. Probiotic yeast: mode of action and its effects on ruminant nutrition. *Yeast-Industrial Applications.* Rijeka, Croatia: IntechOpen; 2017; p. 179–202.
- [18] Chaucheyras-Durand F, Walker N, Bach A. Effects of active dry yeasts on the rumen microbial ecosystem: Past, present and future. *Anim Feed Sci Technol.* 2008;145(1–4):5–26.
- [19] Abdelrahman MM. Effect of direct-fed microbial (DFM)® supplements on general performance of newborn awassi lambs. *Egypt J Sheep Goats Sci.* 2010;5(1):1–18.
- [20] Kumar M, Dutta T, Singh G, Chaturvedi I. Effect of *Lactobacilli* culture on the performance of preweaned Barbari kids. *Indian Res J Gen Biotech.* 2013;5(4):278–86.
- [21] Alhidary IA, Aljumaah RS, Amran RA, Abdelrahman MM, Alowaimier AN, Elsabagh M. Effects of direct-fed microbial supplementation on growth performance and metabolic profile of newborn lambs weaned at different ages. *Indian J Anim Res.* 2019;B-1052:1–5.
- [22] Helrich K. Official methods of analysis of the association of official analytical chemists. Arlington, Texas, United States: Association of Official Analytical Chemists; 1990.
- [23] Wheeler T, Shackelford S, Koohmaraie M. Shear force procedures for meat tenderness measurement. Clay Center, NE: Roman L Hruska US Marc USDA; 2001.
- [24] Wilhelm AE, Maganhini MB, Hernández-Blázquez FJ, Ida EI, Shimokomaki M. Protease activity and the ultrastructure of broiler chicken PSE (pale, soft, exudative) meat. *Food Chem.* 2010;119(3):1201–4.
- [25] Bonvillani A, Peña F, De Gea G, Gómez G, Petryna A, Perea J. Carcass characteristics of Criollo Cordobés kid goats under an extensive management system: effects of gender and live weight at slaughter. *Meat Sci.* 2010;86(3):651–9.

- [26] SAS S, Guide SU: Software Version 9.1. 2 Cary, NC: SAS Institute Inc., 2004. In.: USA.
- [27] Abu-Tarboush HM, Al-Saiady MY, El-Din AHK. Evaluation of diet containing lactobacilli on performance, fecal coliform, and lactobacilli of young dairy calves. *Anim Feed Sci Technol.* 1996;57(1–2):39–49.
- [28] Agazzi A, Tirloni E, Stella S, Marocco S, Ripamonti B, Bersani C, et al. ni Savoini G: effects of species-specific probiotic addition to milk replacer on calf health and performance during the first month of life. *Ann Anim Sci.* 2014;14(1):101–15.
- [29] Ellinger DK, Muller LD, Glantz PJ. Influence of feeding fermented colostrum and *Lactobacillus acidophilus* on fecal flora of dairy calves. *J Dairy Sci.* 1980;63(3):478–82.
- [30] Morrill J, Dayton A, Mickelsen R. Cultured milk and antibiotics for young calves. *J Dairy Sci.* 1977;60(7):1105–9.
- [31] Theodorou M, Beever D, Haines M, Brooks A. The effect of a fungal probiotic on intake and performance of early weaned calves. *Anim Prod.* 1990;50:577.
- [32] Titi H, Dmour R, Abdullah A. Growth performance and carcass characteristics of Awassi lambs and Shami goat kids fed yeast culture in their finishing diet. *Anim Feed Sci Technol.* 2008;142(1–2):33–43.
- [33] Whitley NC, Cazac D, Rude B, Jackson-O'Brien D, Parveen S. Use of a commercial probiotic supplement in meat goats. *J Anim Sci.* 2009;87(2):723–8.
- [34] Bechman T, Chambers J, Cunningham M. Influence of *Lactobacillus acidophilus* on performance of young dairy calves. *Journal of Dairy Science:* 1977. 309 W Clark St, Champaign, IL 61820: Amer Dairy Science Association; p. 74–5.
- [35] Jang Y-D, Oh H-K, Piao L-G, Choi H-B, Yun J-H, Kim Y-Y. Evaluation of probiotics as an alternative to antibiotic on growth performance, nutrient digestibility, occurrence of diarrhea and immune response in weaning pigs. *J Anim Sci Technol.* 2009;51(1):25–32.
- [36] Chaucheyras-Durand F, Fonty G. Establishment of cellulolytic bacteria and development of fermentative activities in the rumen of gnotobiotically-reared lambs receiving the microbial additive *Saccharomyces cerevisiae* CNCM I-1077. *Reprod Nutr Dev.* 2001;41(1):57–68.
- [37] Lubbadah W, Haddadin MS, Al-Tamimi MA, Robinson RK. Effect on the cholesterol content of fresh lamb of supplementing the feed of Awassi ewes and lambs with *Lactobacillus acidophilus*. *Meat Sci.* 1999;52(4):381–5.
- [38] Antunović Z, Šperanda M, Amidžić D, Šerić V, Stainer Z, Domačinović M, et al. Probiotic application in lambs nutrition. *Krmiva: Časopis o Hranidbi Životinja, Proizvodnji i Tehnologiji Krme.* 2006;48(4):175–80.
- [39] Antunović Z, Šperanda M, Liker B, Šerić V, Senčić Đ, Domačinović M, et al. Influence of feeding the probiotic Pioneer PDFM® to growing lambs on performances and blood composition. *Acta Veterinaria.* 2005;55(4):287–300.
- [40] Bruno RG, Rutigliano HM, Cerri R, Robinson PH, Santos JE. Effect of feeding *Saccharomyces cerevisiae* on performance of dairy cows during summer heat stress. *Anim Feed Sci Technol.* 2009;150(3–4):175–86.
- [41] Kaneko JJ, Harvey JW, Bruss ML. Clinical biochemistry of domestic animals. San Diego, California, United States: Academic Press; 2008.
- [42] Kalavathy R, Abdullah N, Jalaludin S, Wong C, Ho Y. Effect of *Lactobacillus* cultures and oxytetracycline on the growth performance and serum lipids of chickens. *Int J Poult Sci.* 2008;7(4):385–9.
- [43] Whitehead DC, Goulden KM, Hartley RD. The distribution of nutrient elements in cell wall and other fractions of the herbage of some grasses and legumes. *J Sci Food Agriculture.* 1985;36(5):311–8.
- [44] Kabaija E, Smith O. Trace element kinetics in the digestive tract of sheep fed diets with graded levels of dietary fibre. *J Anim Physiol Anim Nutr.* 1988;59(1–5):218–24.
- [45] Waghorn G, Shelton I, Sinclair B. Distribution of elements between solid and supernatant fractions of digesta in sheep given six diets. *N Zealand J Agric Res.* 1990;33(2):259–69.
- [46] El-Shaer E. Nutritional studies in ruminants. “Effect of yeast culture supplementation and concentrate: roughage ratio on performance of growing lambs.” Ph.D. Thesis. Mansoura, Dakahlia, Egypt: Faculty of Agriculture, Mansoura University; 2003.
- [47] Smirnov A, Perez R, Amit-Romach E, Sklan D, Uni Z. Mucin dynamics and microbial populations in chicken small intestine are changed by dietary probiotic and antibiotic growth promoter supplementation. *J Nutr.* 2005;135(2):187–92.
- [48] Puls R. Mineral levels in animal health: Diagnostic Data. Madison, Wisconsin, United States: Sherpa International; 1988.
- [49] Belewu M, Jimoh N. Blood, carcass and organ measurements as influenced by *Aspergillus Niger* treated cassava waste in the diets of West African dwarf Goat (WAD). *Glob J Agric Sci.* 2005;4(2):125–8.
- [50] Abdelrahman MM, Hunaiti DA. The effect of dietary yeast and protected methionine on performance and trace minerals status of growing Awassi lambs. *Livest Sci.* 2008;115(2–3):235–41.
- [51] Belewu M, Belewu K. Cultivation of mushroom (*Volvariella volvacea*) on banana leaves. *Afr J Biotechnol.* 2005;4(12):1401–3.
- [52] Pelicano ERL, Souza P, Souza HBAD, Oba A, Boiago M, Zeola NMBL, et al. Carcass and cut yields and meat qualitative traits of broilers fed diets containing probiotics and prebiotics. *Braz J Poult Sci.* 2005;7:169–75.
- [53] Elam NA, Gleghorn JF, Rivera JD, Galyean ML, Defoor PJ, Brashears MM, et al. Effects of live cultures of *Lactobacillus acidophilus* (strains NP45 and NP51) and *Propionibacterium freudenreichii* on performance, carcass, and intestinal characteristics, and *Escherichia coli* strain O157 shedding of finishing beef steers. *J Anim Sci.* 2003;81(11):2686–98.