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Research Article

Dietary Supplementation of Olive Pomace in Lactating Buffaloes: Effects on Milk and Yogurt Composition and Fatty Acid Profile toward Heart Health

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ABSTRACT

Olive Pomace (OP) is a residue of the oil extraction of olive fruit, which can be used in animal feed as one of the cheap and nutritious residues. This study was conducted to investigate the effect of olive pomace in three levels of OP0, OP7.5 and OP15 on milk and yogurt composition and milk fatty acid profile of dairy buffalo. Fifteen lactating buffaloes have been used with an average age of 4 ± 1 years and an average weight of 570 ± 40 kg and two parities (second parturition). Treatments of 0%, 7.5%, and 15% olive pomace substituted with wheat flour, which was fed two times for 45 days in a completely randomized design. At the end of the week, milk from each animal (mix of morning and evening milk) was used to test milk composition and microbial load. At the end of the experiment, the fatty acids profile of milk and yogurt composition of all buffaloes were measured. Milk fat and SNF (Solids Not Fat) increased in olive cake treatments ($p<0.05$) but milk production, protein, and pH were not different ($p>0.05$). The acidity, microbial load, Staphylococci, and SNF of yogurt produced from buffalo's milk did not influence by treatments ($p>0.05$). Yogurt fat decreased ($p<0.05$) in OP7.5 and increased ($p<0.05$) in OP15 buffalos. Protein concentration in yogurt dropped by olive pomace diets, and coliforms were the highest for OP15 olive pomace ($p<0.05$). The concentration of short and medium chains fatty acids (C4:0, C8:0, C10:0, C12:0), linolenic acid, and conjugated fatty acids (CLA isomers) of buffalo's milk in OP15 is more than other treatments ($p<0.05$). The replacement of olive pomace with wheat flour increased milk production of dairy buffaloes. The concentration of short and medium chain fatty acids and unsaturated and conjugated fatty acids increased in OP15 buffalos. Furthermore, a lower saturated/unsaturated ratio and atherogenic index suggest an improvement in nutritional characteristics of milk buffaloes. Therefore, it's recommended in the diet of dairy buffaloes.

Keywords: Olive pomace; Buffalo; Milk; Yogurt; Fatty acid profile

INTRODUCTION

Arid and semi-arid climatic conditions in Iran and a reduction in crop production have caused livestock feeding to allocate a significant portion of animal husbandry costs and affect revenues from the production of livestock products [1]. Given that feeding in animal husbandry is one of the most critical and costly issues, the economic condition focuses on breeding and feeding livestock to use cheap resources based on livestock needs [2,3]. Therefore, diets should be prepared to be cost-effective and provide adequate nutrients for livestock. For this purpose, using agricultural waste is recommended [4,5]. It is obtained from olive oil production and represents an interesting feed supplement [6,7]. Indeed, olive cultivation is widespread in many countries (United States, Argentina, South Africa, India, China, Australia, New Zealand, as well as in the Mediterranean area) [8], and this make particularly accessible its byproducts. Olive pomace includes the pasty part, kernel, skin, and sewage and after drying, olive pomace can be used as a by-product in animal feed [9,10]. Different ways to include olive byproduct in animal diets have been described, varying from feeding it fresh, ensiled, dried or as a component of concentrate pellets and multi-nutrient feed blocks and dried olive pomace represents one of the most suitable for its stability. The reported low

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nutritive value of olive pomace is mainly due to the high content of lignin; however, the stoning process reduces the lignin content and might improve digestibility that, in addition to a good energy value for the high residual lipid content, makes the pomace a potential good energy supplement. Olive pomace (dried) contains high lignin (28% to 32% DM), cellulose (9% to 13% DM), and hemicellulose (13% to 17% DM), and its low nutritional value is mainly due to high lignin content, although the extraction processes reduce lignin content with high residual lipid content that makes olive pomace a potential good energy supplement [11]. The latest technologies are able to preserve olive phenol content and integrity by controlling polyphenol oxidase and peroxidase [12]. These molecules can be found in milk from buffaloes and plays an important role in prevention of human pathologies as cancer, chronic heart diseases and atherosclerosis [13,14], being particularly significant for the production of healthier foods. Conversely, one of the problems of using olive pomace is the effect of these substances as anti-nutritional components, playing both potential health benefits and adverse effects on digestion in the livestock [15]. Raw olive pomace contains high amounts of 16 and 18 carbon fatty acids, and about 96% of olive pomace fatty acids include linoleic acid and oleic acid [16,17]. Feeding ruminants with olive pomace containing tocopherol, retinol, and phenols can positively change milk quality and fat and its oxidative stability [18,19]. Using of 0, 10, and 20% olive pomace instead of barley in Lori lambs' diet increased purine derivatives and microbial protein production in the rumen compared to the control diet. In another study, levels of 0%, 33%, 66%, and 100% olive pomace in the Dalagh sheep (Iranian breed) showed that using olive pomace increased feeding efficiency, increased carcass weight and reduced production costs up to 66%. Also, replacing different levels of olive pomace in the diet of Arab sheep improved the digestibility of organic matter, protein, and neutral detergent fibers. Using dried olive kernels in dairy buffaloes increased milk production [20]. Inclusion of unsaturated fatty acids often reduces the saturated fatty acids in the milk as cis-9 18: 1, 18: 2n-6, and 18: 3n-3 relatively similar changes. Fat supplement 18: 2n-6 increases concentrations of CLA in cow's milk.

Most of traditional and industrial buffalo breeders in southwestern of Iran use wheat flour as the only grain source in buffalo feed. Using this grain source leads to rapid changes in ruminal pH and can disrupt the rumen microbial population. Using wheat flour, an essential part of human foods, increases diet costs. Therefore, replacing olive pomace with wheat flour and supplying diet energy can also have better efficiency for beneficial and long chain fatty acids. However, no research has been reported on olive pomace replaced with wheat flour in buffalo nutrition. Also, buffalo milk is high in protein and calcium and it's a suitable milk for making yogurt. Therefore, this experiment was conducted to investigate the effect of olive pomace on the production performance, milk and yogurt quality, and fatty acid profile of milk in lactating buffalo.

MATERIALS AND METHODS

Treatments and animal management

This experiment was conducted in a buffalo farm in southwestern of Iran which has a humid and tropical climate.

Fifteen lactating buffaloes have been used with average age; 4 ± 1 years and average weight; 570 ± 40 kg and two parities (second parturition). Treatments were included 0%, 7.5%, and 15% olive cake substituted with wheat flour, which fed two times daily for 45 days (15 days adaptation period and 30 days experiment) in a completely randomized design (Table 1). Diets were formulated according to the NRC (2001).

The olive pomace used in this study was prepared in an oil factory and directly dried by air drying method and sunlight. The pomaces were stirred and mixed several times daily. The amount of protein, metabolizable energy, neutral detergent fiber, acid detergent fibers, fat, and ash of olive pomace was analyzed and was 6.5%, 2.65 Mk/kg DM, 56, 45, 9, and 3.2%, respectively.

Table 1: Ingredients and chemical composition of experimental diets.

Items	Treatments		
	OP0 (%)	OP7.5 (%)	OP15 (%)
Corn silage	25	25	25
Sugarcane scum (bagasse)	10	10	10
Wheat straw	15	15	15
Wheat bran	13	13	13
Rice bran	8	8	8
Olive cake	0	7.5	15
Wheat flour	20	12.5	5

Barley	5	5	5
Soybean meal	1.7	1.7	1.7
Urea	0.2	0.2	0.2
Calcium carbonate	0.6	0.6	0.6
Salt	0.5	0.5	0.5
Vitamin and mineral supplement	1	1	1
Chemical composition (%)			
Dry matter (g/kg)	580	580	580
NEL (Mcal/kg)	1.42	1.4	1.38
Protein	10.15	9.77	9.4
Ether extract	2.51	2.9	3.28
NDF	43.55	43.55	43.55
ADF	26.3	26.3	26.3

Note: Olive Pomace (OP) substituted with wheat flour at levels 0% (OP0), 7.5% (OP7.5), and 15% (OP15).

Each kilogram of vitamin and mineral supplements contains 600,000 IU vitamin A, 200,000 IU vitamin D_3, 200 mg vitamin E, 2500 mg antioxidant, 195 g calcium, 80 g phosphorus, 21,000 mg magnesium, 22 mg of manganese, 3000 mg of iron, 300 mg of copper, 300 mg zinc, 100 mg of cobalt, 120 mg of iodine and 1.1 mg of selenium.

Samples collections and analysis

Milking was done twice a day in the morning and evening, daily and recorded. At the end of every week of experiment, 200 mL milk was collected from each animal twice, daily then mixed in equal proportion and was sent to the laboratory to test milk composition and microbial load. Samples were collected for four consecutive weeks. At the end of the experiment after 45 days, morning and evening milk of each animal were mixed to determine the fatty acid pattern (15 samples). About 5 L milk samples taken from all animals were boiled separately (15 samples). After reaching the proper temperature of 43°C, yogurt rennet was added, and yogurt's microbial composition and microbial load were measured. Milk scanner S50, mid-infrared scanner was used to measure milk and yogurt protein, fat, and lactose, and a gyroscope device and 2.5 ccs of milk were used for 2 minutes to measure the freezing point. To determine the pH of milk and yogurt, suitable electrical meters were used. Also, to measure milk solids and Solids Not Fat (SNF), a lactometer was used.

For determining the microbial load of milk and yogurt for each animal (15 samples), 7 test tubes were needed. Nine ml physiological serum was poured into 7 test tubes and autoclaved for each sample. After cooling, 1 ml of milk sample was poured into the first tube, then 1 ml was taken from the first tube and ran into the second tube, and the same was continued to the last pipe until the serial dilutions were created. To determine the microbial load of milk or total count, 1 ml of each dilution was taken separately and cultured in a plate (1 ml with a sterile pipette poured into a sterile container of the culture medium of Kant agar plate, which is close to coagulation at a temperature of about 40°C-50°C) and incubated at 31°C for three days. The number of colonies was counted, and the counted average was calculated in different dilutions and was considered the microbial load or total count of milk. The number of molds were measured through the surface culture in PDA medium at 25°C for 48 h, and results reported base on the number of colonies obtained from each mL of milk (colony/mL). To determine the number of *Staphylococcus* and *Coliforms* bacteria, colony counting with serial dilution and specific culture media (blood agar and Rugosa agar) were used.

X=Number of colonies × Reverse the amount of injection × Reverse dilution

At the end of the experiment, to determine the fatty acids, 150 mL morning and evening milk samples of each treatment were mixed, and 20 cc of each treatment was taken and quickly sent to the standard research institute for fatty acid profiles with protocol numbers 13126-2 and 13126-4 according to the method of national standards of Iran. First, 2 cc of milk was taken from each sample and mixed with 2 cc of hexane. Milk fat entered hexane, and then 1 cc of hexane was taken, and 200 µmol of 2 M methanolic potash was added and vortexed. After 5 minutes, the two phases were separated from each other (but, if the two phases are not separated, we centrifuge to separate the two

phases). Then, the upper phase was injected into the GCFID device (Gas Chromatography (GC) with Flame-Ionization Detection (FID). Thus, the amount and type of fatty acids were reported separately.

Atherogenic (AI) and Thrombogenic (TI) indices were calculated according to the following formula:

AI=(C12:0+4 × C14:0+C16:0)/(MUFA+∑n6+∑n3);

TI=(C14:0+C16:0+C18:0)/(0.5 × MUFA+0.5 × ∑n6+3×∑n3+∑n3/∑n6).

Statistical analysis

The results (milk fatty acid profile and milk and yogurt microbial load) were analyzed as a completely randomized design (t-test) using the General Linear Model (GLM) procedure of the SAS. A comparison of the mean was made with Duncan. The model of this design is a completely randomized design with three treatments, and each treatments includes five replications based on the statistical model: Yij=μ+Ti+ eij.

Where;

Yij=Observation

μ=General mean

Ti=Effect of level of olive cake

eij=SE

Also, repeated data per time (daily milk and milk and yogurt composition) were analyzed based on the mixed statistical model.

Yci=μ+Tc+Ɛic+(T*P)ik

Where;

Yci=Observation

μ=Mean of observations

Tc=Effect of level of olive cake

Ɛic=Effect of experimental error

(t * P)=Period of the experiment, which is analyzed at a significant level of 5%.

RESULTS AND DISCUSSION

Milk fat and Solids Not Fat (SNF) increased in olive cake treatments ($p<0.05$), but there were no differences among experimental diets in dry matter intake, daily milk production, milk protein and pH (Table 2).

Table 2: Dry matter intake, milk production and composition of buffaloes fed with experimental diets.

Variables	Treatments[1]			SEM	P-value
	OP0	OP7.5	OP15		
Dry matter intake (kg/day)	16.86	17.1	17.47	0.39	0.339
Milk production (kg/day)	9.8	10.09	10.31	0.27	0.174
Fat (%)	6.27[a]	6.17[b]	5.49[c]	0.02	0.007
Protein (%)	3.33	3.32	3.29	0.11	0.638
Lactose (%)	4.64	4.62	4.58	0.14	0.697
Acidity (%)	12.6	12.6	11.89	0.51	0.2146
Density (g/ml)	1.02	1.03	1.08	0.12	0.137
Freezing point	545.4	548	543.2	2.28	0.346
Solids Not fat	8.68[a]	8.60[b]	8.47[c]	0.01	0.0001
pH	6.8	6.85	6.82	0.11	0.145

Note: [1]Olive Pomace (OP) substituted with wheat flour at levels 0% (OP0), 7.5% (OP7.5), and 15% (OP15).
[ab] in each row showed a significant difference in the error level of $p<0.05$.

It is noteworthy that buffalo milk is richer than cow milk in all major constituents, with higher energy and nutritional values, higher levels of lactose, protein and as. As just reported by other authors, also in our study the olive pomace inclusion in the diet did not affect milk yield.

It is important to consider that the crude protein associated with the high phenolic content of olive pomace have been related to low digestibility in ruminants. Probably, as speculated by other researchers, apparent intestinal digestibility of rumen undegraded protein may have improved due to the drying processes. Similar results were reported buffaloes as well as for other ruminant species as cows and ewes. It must be said that use of live pomace in buffaloes has been poor studied until now and that often small numbers of animals have been used. So, given the different capabilities of feed exploitation between buffalo and cattle, we are often forced to compare results with other species. The replacement of barley with olive pomace in the Dalaq fattening lambs significantly affected dry matter intake. Although no differences in protein content were reported, fat concentration decreased with the higher inclusion of olive pomace. Including olive pomace in cows, other researchers reported a positive effect on milk protein content and a negative effect on milk fat content. Although diets were isoenergetic and isoproteic, they ascribed this result to the difference that this inclusion gives to the forage/concentration ratio of the diet. However, we can consider also the impact that olive pomace might have had on rumen microbiota and so VFA production, because olive pomace integration may modify rumen microbial activity and population and consequently protein and fat milk content. The inclusion of unsaturated fat supplements such as sunflower seeds, flaxseed, and canola reduced dry matter intake in the dairy cow's diet. Increasing fat milk percentage is probably due to the higher percentage of fat in olive pomace compared to flour. Adding fat supplements to the dairy ewes diet reduces heat stress and increases the fat density in the diet and milk as well as SNF.

Table 3 shows the yogurt composition produced from the buffalo milk fed with olive pomace. The fat, protein, and coliform population of yogurt produced from the buffalo milk fed with olive pomace have been influenced by experimental treatments, which probably protein reduction may be due to the presence of phenol compounds which bind milk protein. Higher microbial load of milk in animal fed fat-rich diets was due to more microbial protein which cause to higher coliform production. It is reported that feeding with olive pomace and tomato fruit reduced milk casein, and increased the unsaturated fatty acids in Awasi sheep. Researchers showed that, by increasing the fat percentage, the acidity decreases significantly, and the tissue properties improve (increase viscosity and decrease hydration). The increase in viscosity with increasing fat may be due to an increase in total solids and thus an increase in product stiffness, which ultimately reduces crop hydration. Moreover, the fat in mozzarella was higher in dairy buffalo fed 17 kg DM/day dried stoned olive pomace which increased the Monounsaturated Fatty Acids (MUFAs) and the saturated/unsaturated ratio while it decreased the atherogenic indices.

Table 3: Composition and quality of yogurt from buffaloes fed diets with experimental diets.

Variables	Treatment[1]			SEM	P-value
	OP0	OP7.5	OP15		
Fat (%)	4.34[a]	4.11[c]	4.28[b]	0.04	0.033
Protein (%)	3.76[c]	3.86[b]	3.95[a]	0.01	0.0004
Acidity	82.33	82.66	85	4.3	0.894
Coliform (colony/mL)	20.00[a]	15.00[b]	10.00[c]	0.37	0.0001
Staphylococcus (colony /mL)	4.33	4.17	4.28	0.16	0.24
Mold (colony/mL)	20	26	21.33	2.75	0.33
pH	4.34	4.17	4.28	0.09	0.24
Solid not fat (%)	9.12	9.21	9.1	0.51	0.447

Note: [1] Olive Pomace (OP) substituted with wheat flour at levels 0% (OP0), 7.5% (OP7.5), and 15% (OP15).
[ab] in each row showed a significant difference in the error level of $p<0.05$.

Inclusion of OP7.5 and OP15 instead of wheat flour significantly changed concentrations of Saturated Fatty Acids (SFA) of butyric, caproic, caprylic, capric, lauric, palmitic, Polyunsaturated Fatty Acids (PUFA) of oleic, linolenic, CLA8cis 10cis, CLA10cis 12cis, CLA10trans 12cis, CLAcis10 trans12, CLAcic12, while the C14 was not influenced by treatments (Table 4). All fatty acids increased except palmitic acid and oleic acid, which decreased. Fatty acids C16: 1 trans and CLA: 8cis 10cis increased by OP15. Increasing de novo synthesis of fatty acids due to tannins can increase the concentration of short chain fatty acids.

The proposed biohydrogenation theory shows that some nutritional conditions can change the biohydrogenation process of unsaturated fatty acids and thus increase their amount in

milk. Milk and dairy products are significant sources of saturated fatty acids and cis-9, trans-11 CLA in the human diet.

Table 4: Milk fatty acids profile of buffaloes fed with experimental diets (%).

Variables	Treatments[1]			SEM	P-value
	OP0	OP7.5	OP15		
C4:0	1.97^{c}	2.68^{a}	2.66^{a}	0.1	0.024
C5:0	0.02	0.04	0.1	0.19	0.11
C6:0	1.18	3.1	4.78	0.55	0.06
C8:0	0.94^{c}	2.08^{b}	2.78^{a}	0.11	0.012
C9:0	0.23	0.07	0.07	0.19	0.46
C10:0	1.90^{c}	2.92^{b}	4.06^{a}	0.04	0.0014
C11:0	0.11	0.11	0.21	0.09	0.6
C12:0	2.06^{c}	2.95^{b}	4.23^{a}	0.21	0.022
C12:1 cis	0.46	0.52	0.48	0.22	0.38
C12:1 trans	0.17	0.2	0.18	0.09	0.94
C13:0	0.07	0.1	0.46	0.29	0.41
C14:0	12.2	13.7	14.85	0.52	0.07
C15:0	1.22	1.3	1.18	0.31	0.2
C16:0	33.5^{a}	29.65^{b}	24.55^{c}	0.17	0.002
C17:0	0.43	0.39	0.36	0.31	0.42
C18:1 9cis	16	14	12.85	1.2	0.41
C18:0	9.65	8.35	7.1	1.21	0.43
C20:0	0.25	0.21	0.29	0.09	0.67
C22:0	0.09	0.1	0.1	0.06	0.93
C6:1	0.05	0.5	0.57	0.15	0.1
C10:1 cis	0.16	0.24	0.2	0.09	0.7
C14:1 cis	1.71	1.73	1.66	0.08	0.45
C14:1 trans	0.39	0.45	0.5	0.09	0.4
C15:1 cis	0.56	0.54	0.52	0.06	0.92
C15:2	0.04	0.045	0.045	0.03	0.96
C16:1 cis	3.35	2.39	2.59	0.2	0.46
C16:1 trans	0.15^{b}	0.11^{c}	0.50^{a}	0.01	0.02

C17:1	0.14	0.2	0.13	0.54	0.62
C18:1 cis	0.74	1.76	2.26	0.19	0.015
C18:2 cis	1.78	1.68	1.6	0.52	0.92
C18:3 cis	0.13^{b}	0.22^{b}	0.40^{a}	0.008	0.0004
C20:1	0.46	0.39	0.39	0.08	0.29
C22:1	0.05	0.05	0.09	0.29	0.59
C18:1 11 cis	0.14	0.16	0.17	0.26	0.81
C18:1 12 cis	0.24	0.245	0.375	0.07	0.43
CLA:8cis 10cis	0.08^{b}	0.06^{b}	1.07^{a}	0.02	0.0002
CLA:10cis 12cis	0.015^{c}	0.165^{b}	0.56^{a}	0.04	0.006
CLA:10trans 12cis	0.025^{b}	0.06^{b}	0.23^{a}	0.01	0.008
CLA:cis9 11tra	0.08	0.05	0.32	0.42	0.086
CLA:cis9 11cis	0.035^{b}	0.065^{b}	0.32^{a}	0.01	0.003
CLA:cis10 trans12	0.09^{b}	0.14^{b}	0.59^{a}	0.03	0.002
CLA:cis12 trans10	0.065^{b}	0.085^{b}	0.13^{a}	0.009	0.029
other	6.93^{b}	7.36^{a}	5.40^{c}	0.26	0.0265
ΣPUFA	2.3^{c}	3.065^{b}	5.22^{a}	0.02	0.01
ΣUFA	27.07	26.55	28.685	0.5	0.86
ΣSFA	65.82	65.75	67.78	1.4	0.6
ΣMUFA	24.77^{a}	23.49^{c}	23.47^{b}	0.1	0.003
AI	3.12^{a}	3.29^{b}	3.07^{b}	0.36	0.01
TI	3.97^{a}	3.72^{b}	3.02^{c}	0.01	0.02

Note: MUFAs: The Monounsaturated Fatty Acids; SFA: Saturated Fatty Acids; PUFA: Polyunsaturated Fatty Acids.
[1] Olive Pomace (OP) substituted with wheat flour at levels 0% (OP0), 7.5% (OP7.5), and 15% (OP15).
[ab] in each row showed a significant difference in the error level of $p<0.05$.

Inclusion of vegetal fats rich in unsaturated fatty acids often reduces the proportion of saturated average chain fatty acids in the milk as cis-9 18: 1, 18: 2n-6 and 18: 3n-3 relatively similar changes. More changes were in the proportion of fatty acids 18: 2n-6. In addition, it has been well confirmed that fat supplement 18: 2n-6 increases concentrations of cis-9, trans-11 CLA in cow's milk to a greater extent compared with 18: 1 cis-9 and 18: 3n-3 sources.

In the study of Faye, et al., on the effect of crude olive pomace on fatty acid compositions in camels, olive pomace was replaced by barley in the diet. Increase in palmitic c16 and linoleic acid c18: 3w-6 was observed. The concentration of C4, C6, C8, C10, C12, C14, C16iso, C16: 01w-7, C17: 0, C17: 0iso, C18: 0, C18: 0iso, C18: 1w-9, C18: 2iso, C18: 2w-6, C18: 3w-3, C20: 1w-9, C20: 5w-3, C22: 6w-3 not changed and there was a substantial difference at C15: 0, C15: 0iso, C16: 0, C16: 0iso, C17: 1, C18: 1w-7 and C18: 3w-6 gamma-linolenic acid and in general no significant differences were observed in short and medium fatty acids and long chain fatty acids.

Some researchers observed that by adding 6% olive oil to the diet of sheep, the amount of medium-chain saturated fatty acids decreased while stearic, oleic acid, and c18: 1 trans trans-10 isomers increased. The isomer of C18: 2 trans 11 and cis 9 decreased, but the trans 9 cis 11 and cis 9 trans 7 increased

significantly. Using dried stoned olives pomaces in buffaloes decreased saturated fatty acids and increased unsaturated higher in mozzarella.

Using olive cake in the dairy camel diet, decreased medium-chain fatty acids, and palmitic acid and gamma-linoleic acid increased. In an experiment on the effect of olive oil on dairy ewes; oleic and vaccenic acids increased, and saturated fatty acids decreased. Generally, adding olive oil increased monounsaturated fatty acids and decreased saturated fatty acids.

It is stated that using oilseeds in the diet cause to increase in the milk CLA. The CLA production occurs by lactic acid family and linoleic acid isomerase in the presence of a source of linoleic acid, and the number of CLA increases after fermentation of milk products. In the current study, due to the high production of milk and possibly "high feed intake," there was not enough opportunity for complete biohydrogenation, and part of the oleic acid was contained in the milk. The researchers reported that the amount of linoleic acid (C18: 2cis) in milk fat has decreased with increased roasted canola seed in the diet (a fat resource with high fiber same to olive pomace). Reducing of polyunsaturated fatty acids in the milk was due to the lower concentration of this fatty acid in canola seeds. The atherogenic and thrombogenic indices are interesting parameters by which to measure FA profile. Throughout these indices it is possible to evaluate the different effects that some single fatty acids might have on human health, with the probability of increasing or not the incidence of cardiovascular pathologies as atheroma, thrombus formation. In fact, the atherogenic index is correlated to arteries damages leading to cell adhesion of the immune and circulatory systems, so a lower atherogenic index prevents the occurrence of coronary diseases and is correlated to healthy lipid indices for human consumption. Based on our results, both AI and TI are lower with the addition of olive pomace ($P<0.05$), giving a better health profile to obtained products. Obviously, further studies are needed to evaluate sensory properties as well as clinical performance to verify the existence of a benefit for consumer health.

CONCLUSION

Replacing olive pomace with wheat flour in the diet of lactating buffaloes did not affect the milk production and dry matter intake but increased milk fat. Also, the concentration of short and medium-chain fatty acids (C4:0, C8:0, C10:0, C12:0), linolenic acid, and conjugated fatty acids increased in buffaloes fed with OP15 olive pomace. This experiment showed that replacing olive pomace with wheat flour had no adverse effect on the production performance of lactating buffaloes. Also, due to the reasonable price of olive pomace, its replacement with wheat flours up to 15% level in the diet of lactating buffaloes is recommended.

ANIMAL CARE

All animal management and sampling procedures were conducted according to The Care and Use of Agricultural Animals in Research and Teaching guidelines (FASS). All procedures and guidelines involving animals were approved by the Animal Experiment Committee at Agricultural Sciences and Natural Resources University of Khuzestan, Iran.

AUTHOR CONTRIBUTIONS

T.M: Conceptualization, methodology, writing the original draft, editing and reviewing manuscript; M.C. and S.A: Conceptualization, methodology, and characterization, editing and reviewing manuscript.

All authors have read and agreed to the published version of the manuscript.

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CONFLICTS OF INTEREST

The authors declare no conflict of interest.

REFERENCES

1. Devendra C. Perspectives on animal production systems in Asia. Livest Sci. 2007;106(1):1-8.
2. Besharati M, Palangi V, Salem AZ, de Palo P, Lorenzo JM, Maggiolino A. Substitution of raw lucerne with raw citrus lemon by-product in silage: *In vitro* apparent digestibility and gas production. Front Vet Sci. 20223;9:1006581.
3. de Bellis P, Maggiolino A, Albano C, de Palo P, Blando F. Ensiling grape pomace with and without addition of a *Lactiplantibacillus plantarum* strain: Effect on polyphenols and microbiological characteristics, *in vitro* nutrient apparent digestibility, and gas emission. Front Vet Sci. 2022;9:808293.
4. Maggiolino A, Bragaglio A, Salzano A, Rufrano D, Claps S, Sepe L, et al. Dietary supplementation of suckling lambs with anthocyanins: Effects on growth, carcass, oxidative and meat quality traits. Anim Feed Sci Technol. 2021;276:114925.
5. Maggiolino A, Lorenzo JM, Quinones J, Latorre MA, Blando F, Centoducati G, et al. Effects of dietary supplementation with *Pinus taeda* hydrolyzed lignin on *in vivo* performances, *in vitro* nutrient apparent digestibility, and gas emission in beef steers. Anim Feed Sci Technol. 2019;255:114217.
6. Sansoucy R. Olive by-products for animal feed. Food Agr Org. 1985.
7. Zilio DM, Bartocci S, Di Giovanni S, Servili M, Chiariotti A, Terramoccia S.. Evaluation of dried stoned olive pomace as supplementation for lactating Holstein cattle: Effect on milk production and quality. Anim Prod Sci. 2014;55(2):185-188.
8. Vossen P. Olive oil: History, production, and characteristics of the world's classic oils. HortSci. 2007;42(5):1093-1100.

9. Molina-Alcaide E, Yanez-Ruiz DR. Potential use of olive by-products in ruminant feeding: A review. Anim Feed Sci Technol. 2008;147(1-3):247-264.
10. Berbel J, Posadillo A. Review and analysis of alternatives for the valorisation of agro-industrial olive oil by-products. Sustainability. 2018;10(1):237.
11. Sadeghi H, Yansari AT, Ansari-Pirsarai Z. Effects of different olive cake by products on dry matter intake, nutrient digestibility and performance of Zel sheep. Int J Agric Biol. 2009;11(6).
12. Servili M, Esposto S, Fabiani R, Urbani S, Taticchi A, Mariucci F, et al. Phenolic compounds in olive oil: Antioxidant, health and organoleptic activities according to their chemical structure. Inflammopharmacol. 2009;17:76-84.
13. Terramoccia S, Bartocci S, Taticchi A, Di Giovanni S, Pauselli M, Mourvaki E, et al. Use of dried stoned olive pomace in the feeding of lactating buffaloes: Effect on the quantity and quality of the milk produced. Asian-Australas J Anim Sci. 2013;26(7):971.
14. Covas MI. Bioactive effects of olive oil phenolic compounds in humans: Reduction of heart disease factors and oxidative damage. Inflammopharmacol. 2008;16:216-218.
15. Alcaide EM, Ruiz DY, Moumen A, Garcıa IM. Chemical composition and nitrogen availability for goats and sheep of some olive by-products. Small Rumin Res. 2003;49(3):329-336.
16. Difonzo G, Troilo M, Squeo G, Pasqualone A, Caponio F. Functional compounds from olive pomace to obtain high added value foods–a review. J Sci Food Agric. 2021;101(1):15-26.
17. Paz A, Karnaouri A, Templis CC, Papayannakos N, Topakas E. Valorization of exhausted olive pomace for the production of omega-3 fatty acids by *Crypthecodinium cohnii*. Waste Manag. 2020;118:435-444.
18. Castellani F, Vitali A, Bernardi N, Marone E, Palazzo F, Grotta L, et al. Dietary supplementation with dried olive pomace in dairy cows modifies the composition of fatty acids and the aromatic profile in milk and related cheese. J Dairy Sci. 2017;100(11):8658-8669.
19. Taticchi A, Bartocci S, Servili M, Di Giovanni S, Pauselli M, Mourvaki E, et al. Effect on quanti-quality milk and mozzarella cheese characteristics with further increasing the level of dried stoned olive pomace in diet for lactating buffalo. Asian-Australas J Anim Sci. 2017;30(11):1605.
20. Samadi, F. Effects of different levels of barely substitution by olive cake on Dalagh lamb fattening. J Agric Sci. 2009;16.