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## Determining the effects of different bypass fat supplementation on performance, milk yield, and milk composition of Anatolian buffaloes

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**Abstract:** This study aimed to explore the effects of animal bypass fat (ABF) and palm bypass fat (PBF) added to the rations of Anatolian buffaloes on dry matter consumption, feed conversion ratio, milk yield, some milk components, and parameters. A total of 21 heads of buffalo in the early lactation period; milk yield averages, lactation periods, lactation numbers, live weights, and ages were selected and divided into three experimental groups, each consisting of 7 buffaloes. The experiment was conducted according to the crossover trial pattern. There was no statistically significant difference between the groups in terms of dry matter consumption and feed conversion ratio ( $p > 0.05$ ). There were no significant differences between the groups in terms of milk yield and corrected milk yield ( $p > 0.05$ ). In addition, there were no statistically significant differences between the groups in terms of milk fat, solid not fat, protein, lactose, and milk energy yields ( $p > 0.05$ ). The better values of feed conversion ratio, higher milk yield, milk fat, milk protein and lactose were obtained in the group of ABF compared to the control and PBF groups. The milk pH, temperature, density, freezing point, ash, organic matter ratio and milk urea nitrogen values obtained at the end of the experiment were insignificant between the groups ( $p > 0.05$ ). In conclusion, to increase milk yield and components of buffalo rations, it has been determined that ABF and PBF can be added to 250 g per animal per day and will not cause any side effects. When the groups to which bypass fat sources were added were compared, it was concluded that ABF had more positive effects on performance and milk composition than PBF.

**Key words:** Buffalo, bypass fat, milk composition, performance

### 1. Introduction

Buffalo (*Bubalus bubalis*), which are classified into two groups as swamp and water buffalo, have been rearing all over the world but mainly in Asian countries [1]. They are mostly fed with low-quality roughages, crop residues, and industrial by-products in order to gain meat and milk [2]. While swamp buffaloes are mainly reared for draught power and meat production due to around 600 kg lactation milk yield, water buffaloes have been reared as dairy animals with almost 2000 kg lactation milk yield [1]. Despite not having been subjected to comprehensive breeding studies in terms of milk production like dairy cattle, buffaloes are the second highest milk producer (127.3 million t) in the world. Moreover, they are one of the most productive farm animals in terms of produced milk's ratio to its dry matter content [3,4].

The buffalo breed, rearing in Türkiye, named as Anatolian buffalo which has horns curved backwards, is black coat-colored and originated from the Mediterranean buffalo [1]. Mature live weight is 450–500 kg in females and 700–800 kg in males [5]. The Anatolian buffalo

population in Türkiye was about 1140 thousand heads in the 1960's but had reduced dramatically to 84 thousand heads by 2010 due to some socio-economic reasons like replacing poor yielded buffalo to high yielded Holstein-Friesian dairy cows, wide usage of mechanization instead of draught power of buffalo, and insufficient demand to the buffalo products [6]. The majority of buffalo farms in Türkiye comprise of small-scale (1–5 head) traditional farms in which buffalo graze at pasture whole year except for harsh weather conditions and milking ones [5]. Lactation milk yield and lactation length were reported as  $1087.49 \pm 5.91$  kg, and  $245.43 \pm 0.90$  d, respectively [7].

Bypass fats are included in ruminant rations for boosting energy density. However, higher inclusion of rations might reduce the degradability of cellulose in the rumen according to the type and amount of included fat [8]. Although the digestibility of dry matter and cellulose reduces when the fat level of the ration exceeds 3%, the protected fat level of the ration can reach 6%–7% since protected fats are degraded and absorbed at the lower gut which is the optimum part for milk and milk fat production

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at digestive tract. Calcium salts of fatty acids, hydrogenated protected fats, and fractionated fats are protected fats widely used in animal nutrition. Calcium salts of fatty acids have a soap taste that animals do not prefer [9].

Bypass fats positively affect milk yield and characteristics in cattle, sheep, goat. There are different studies in ruminants [10–14]. Therefore, this study is the first to compare the effects of both bypass fats in buffaloes reared in Türkiye. Based on this hypothesis, the effects of different bypass fats on feed consumption, feed efficiency, milk yield, and some milk parameters (fat, protein, lactose, solid not fat, urea nitrogen, pH, density, freezing point, ash) were determined.

## 2. Materials and methods

### 2.1. Animals and experimental rations

This study was carried out in a private farm, operating in the province of Bartın (coordinates; 41°35'12.6594"N and 32°23'31.272"E, altitude; 48.02 m)

A total of 21 Anatolian buffaloes with an average live weight of  $445 \pm 65$  kg, found during the 1<sup>st</sup>, 2<sup>nd</sup>, and 3<sup>rd</sup> parity and averaging  $81 \pm 42$  days milked, were selected and divided into three groups. The equal distribution of animals regarding their lactation number to each group is considered. All seven buffaloes in each experimental group were placed in the prepared paddocks of 22.3 m<sup>2</sup> area. All procedures were approved by the Local Ethical Committee of the Experimental Research on Animals (No: 2016-15-148).

### 2.2. Experimental design

This study was designed as feeding trials with rations supplemented with different bypass fats in 3 different treatment groups in 3 different periods. Three groups, which are one control and two trials (PBF and ABF) were formed. Each experimental period consisted of a 15-day of transition period and a 5-day of milk sampling period. The experiment was continued 60 days in 3 periods of 20 days according to the  $3 \times 3$  Latin square trial order. The rations were prepared according to the daily nutritional needs of the animals, taking into account the formula for large-breed dairy cows and the nutritional needs of buffaloes in lactation [15]. The amount of feed the animals could consume daily during the adaptation period was determined, and they were fed to increase by 10% during the experiment. At the beginning of the experiment, 6 kg of corn silage, 6 kg of wheat straw, and 4 kg of concentrated feed were given per animal, while silage and wheat straw were increased to 6.5 kg in the third period of the experiment. The roughage to concentrate ratio was set to approximately 70:30. The feeding was done daily as two meals in the morning and evening. Additionally, clean water and licking-block containing minerals were always available for the animals. Bypass fats were

gradually increased and added to the concentrate feed in the morning, as 250 g per animal daily, during the adaptation period.

The feeds used in the study were prepared as total mixed ration (TMR). Palm bypass fat (PBF) and calcium salt of animal bypass fat (ABF) were used as bypass fat sources. The combination of PBF and ABF used in the research is given in Table 1.

The amounts of dry matter (DM), crude protein (CP), ether extract (EE), ash, crude fiber (CF) nutrient amounts of the concentrate and roughage used were determined according to AOAC [16]. Additionally, neutral detergent fiber (NDF) and acid detergent fiber (ADF) analyses were implemented according to the regarding method [17]. Metabolizable energy in concentrate feed, amounts of corn silage and wheat straw and TMR were calculated following the instructions previously reported and given in Table 2 [18–20]. The amount of non-fiber carbohydrates (NFC) was calculated by difference:  $100 - (CP + \text{ash} + EE + \text{NDF})$  [21].

The feed was weighed and given as two meals. Each morning before feeding, the residual feed left by animals was weighed. Thus, the amount of feed consumed per day was calculated. Feed conversion ratio (FCR); 4% fat corrected milk (FCM) (FCR1), FCM (FCR2) for fat and protein, FCM (FCR3), fat, protein, and lactose, and solid not fat FCM (FCR4) according to the substance were calculated by dividing the daily dry matter use, respectively (Table 3) [22–24].

Animals were milked once a day in the morning with automatic milking machines and their milk yields were determined as the group average. Milk yields, fat-corrected milk (FCM), fat and protein, energy, and solid not fat (SNF) yields were calculated, respectively [22, 25, 26]

Samples were collected by weighing the collected milk in the last five days of each trial period. Dry matter, fat, protein, lactose, density, freezing point, and ash analyses were carried out in the milk samples using an automatic milk analyzer (Lactostar, FUNKE GERBER, Article No. 3510, Berlin, Germany). The pH levels in milk were measured with a pH meter (Testo 205). Ammonia in milk was determined by the modified indophenol method [27–29].

### 2.3. Statistical analyses

The study was carried out according to the crossover trial design. The statistics obtained to analyze of the data of buffaloes are expressed as mean and standard error ( $\bar{x} \pm S_x$ ). Continuous variables; comparing changes according to groups and periods, Repeated Measured Analysis of Variance was performed. The Duncan Multiplicity Comparison Test was used to determine the differences between the groups following the analysis of variance [30]. The statistical efficiency level was taken as 5% in the calculations and the SPSS statistical software was used.

### 3. Results

The effect of bypass fat in the feeding of Anatolian buffaloes on performance parameters such as total dry matter intake (DMI) and feed conversion ratio is provided in Table 3. The effect of parameters such as daily milk yield, FCM

according to fat, FCM according to fat and protein, FCM according to energy, and FCM according to solid nonfat on milk yield are shown in Table 4. Effect on some milk components such as milk fat, milk fat yield, milk DM, solid non-fat and yield, protein ratio, milk protein yield, lactose

**Table 1.** The composition of the PBF and ABF used in the experiment.

Component	PBF*	ABF*
Metabolisable energy (MJ/kg)	17.41	16.46
Ether extract (%)	84	85
Crude ash (%)	14	12.2
Moisture (%)	3.5	3.5
Aroma (%)	0.4	-
Butylated hydroxytoluene (mg/kg)	125	125
Calcium (%)	9	9
Fatty acid profile determined by analysis, (%)		
Lauric acid (C <sub>12</sub> )	2.67	0.35
Myristic acid (C <sub>14</sub> )	1.18	3.25
Palmitic acid (C <sub>16</sub> )	49.31	29.58
Palmitoleic acid (C <sub>16-1</sub> )	0.22	2.50
Stearic acid (C <sub>18</sub> )	3.63	24.44
Oleic acid (C <sub>18:1</sub> cis-9)	38.82	36.36
Linoleic acid (C <sub>18-2</sub> )	3.04	2.82
Linolenic acid (C <sub>18-3</sub> )	0.10	0.07
Arachidic acid (C <sub>20</sub> )	0.32	0.00
Others	0.20	0.2
Total SFA*	57.11	57.62
Total MUFA*	39.04	38.86
Total PUFA*	3.14	2.89
Total UFA*	42.18	41.75
Unsaturated/ Saturated	0.74	0.70

\* PBF: Palm bypass fat; ABF: Animal bypass fat; SFA: Saturated fatty acids; MUFA: Monounsaturated fatty acids; PUFA: Polyunsaturated fatty acids; UFA: Unsaturated fatty acids

**Table 2.** Chemical composition of feeds.

	Concentrate feed	Corn silage	Wheat straw	TMR*
Dry matter (%)	87.00	25.00	92.00	63.00
Crude protein (%)	20.10	8.50	4.40	10.88
Ether extract (%)	3.85	3.40	0.99	1.95
Crude fiber (%)	10.10	29.30	45.35	20.50
Ash (%)	6.32	3.30	2.77	3.80
Neutral detergent fiber (%)	28.50	47.10	78.56	62.98
Acid detergent fiber (%)	11.60	32.00	48.33	34.55
Nonfiber carbohydrates (%)	41.23	37.70	13.28	20.39
Metabolizable energy (MJ/kg)	11.40	8.80	6.25	8.75

\* TMR: Total Mixed Ration

**Table 3.** Effect of bypass fat in feeding of Anatolian buffaloes on some performance parameters.

	Control	PBF*	ABF*	p-values
Total DMI* (kg/day)	10.30 ± 0.097	10.22 ± 0.176	10.31 ± 0.15	0.875
FCR*1 (Milk yield 4% fat-corrected /DMI)	0.38 ± 0.028	0.42 ± 0.021	0.44 ± 0.014	0.257
FCR2 (Fat and protein corrected milk yield/DMI)	0.39 ± 0.028	0.42 ± 0.022	0.44 ± 0.021	0.305
FCR3 (Energy-corrected milk yield/DMI)	0.59 ± 0.052	0.65 ± 0.038	0.71 ± 0.049	0.210
FCR4 (Solid not fat milk yield/DMI)	0.58 ± 0.051	0.64 ± 0.037	0.70 ± 0.048	0.223

\* PBF: Palm bypass fat; ABF: Animal bypass fat; DMI: Dry matter intake; FCR: Feed Conversion Ratio  
 No significant differences among groups (p > 0.05).

**Table 4.** Effect of bypass fat in feeding of Anatolian buffaloes on some milk yield parameters.

	Control	PBF*	ABF*	p-values
Daily milk yield (kg/day)	2.53 ± 0.138	2.69 ± 0.116	2.81 ± 0.079	0.225
Milk yield 4% fat-corrected (kg/day)	3.91 ± 0.977	4.27 ± 0.723	4.54 ± 0.816	0.136
Fat and protein corrected milk yield (g/day)	3.95 ± 0.254	4.25 ± 0.203	4.57 ± 0.203	0.235
Energy-corrected milk yield (kg/day)	6.04 ± 0.474	6.65 ± 0.356	7.36 ± 0.494	0.123
Solid not fat corrected milk yield (kg/day)	5.89 ± 0.466	6.49 ± 0.350	7.19 ± 0.488	0.123

\* PBF: Palm bypass fat; ABF: Animal bypass fat  
 No significant differences among groups (p > 0.05).

**Table 5.** Effect of bypass fat in the feeding of Anatolian buffaloes on some milk yield components.

	Control	PBF*	ABF*	p - values
Milk fat (%)	7.57 ± 0.157	7.91 ± 0.182	8.03 ± 0.258	0.269
Milk fat yield (g/day)	300 ± 24.658	339 ± 17.838	372 ± 27.692	0.115
Milk DM* (%)	17.25 ± 0.345	17.32 ± 0.240	17.94 ± 0.390	0.286
Solid, not fat (%)	9.68 ± 0.231	9.41 ± 0.201	9.91 ± 0.192	0.254
Solid not fat yield (g/day)	380 ± 28.547	400 ± 24.152	450 ± 25.731	0.157
Protein (%)	4.30 ± 0.110	4.17 ± 0.093	4.40 ± 0.087	0.248
Milk protein yield (g/day)	170 ± 12.692	180 ± 10.843	200 ± 11.419	0.155
Lactose (%)	4.65 ± 0.110	4.51 ± 0.094	4.77 ± 0.092	0.210
Milk lactose yield (g/day)	180 ± 13.827	190 ± 11.730	220 ± 12.551	0.156
Milk energy yield (g/day)	290 ± 1.774	300 ± 1.517	300 ± 2.466	0.281

\* PBF: Palm bypass fat; ABF: Animal bypass fat; DM: Dry matter  
 No significant differences among groups (p > 0.05).

ratio, and yield and milk energy yield are shown in Table 5. Finally, its effects on some milk characteristics such as density, freezing point, ash, organic matter, and milk urea nitrogen are given in Table 6.

Similar values were determined in 3 different groups and there was no statistically significant difference between the groups when daily dry matter intake of buffaloes was evaluated. Correlatively, it was determined that the FCRs did not make a significant difference between the groups. Still, it was reflected more positively in the groups to which bypass fat was added to their feed compared to the control group. There was statistically no significant difference

between the groups regarding milk yield, milk dry matter, milk fat and fat yield, solid not fat and dry matter yield, milk protein and protein yield, milk lactose and lactose yield, and its effect on milk yield. Likewise, no differences were found between the energy efficiency of animals with bypass fat added to their rations (p > 0.05). Regarding this issue, higher values were obtained in the groups fed with a ration containing bypass fat compared to the control group. Similarly, the groups with bypass fat added to the rations had no remarkable effect on milk parameters such as pH, density, freezing point, minerals, organic matter, and urea-nitrogen (p > 0.05).

**Table 6.** Effect of bypass fat in the feeding of Anatolian buffaloes on some milk characteristics.

	Control	PBF*	ABF*	p-value
pH	6.68 ± 0.016	6.72 ± 0.021	6.69 ± 0.023	0.392
Density (g/cm <sup>3</sup> )	1.028 ± 0.0008	1.026 ± 0.0008	1.028 ± 0.0009	0.186
Freezing point (°C)	0.67 ± 0.011	0.66 ± 0.008	0.67 ± 0.013	0.511
Ash (%)	0.56 ± 0.007	0.56 ± 0.005	0.56 ± 0.027	0.961
Organic matter (%)	16.69 ± 0.346	16.77 ± 0.240	17.38 ± 0.389	0.284
Urea-N (mg/dL)	17.02 ± 0.629	16.61 ± 0.593	16.55 ± 0.328	0.332

\* PBF: Palm bypass fat; ABF: Animal bypass fat

No significant differences among groups ( $p > 0.05$ ).

#### 4. Discussion

There was no statistically significant difference between the groups regarding total daily DMI for the control, PBF, and ABF-added groups (Table 3). Different reports have been made that adding bypass fat to buffalo rations does not affect DMI [31–35]. Similarly, there are reports that adding bypass fat to dairy cow rations does not affect DMI [11, 36–38]. This situation could be explained by the fact that DMI may decrease or not change of due to the suppression of cellulose digestion in the rumen by adding fat to ruminant rations and reducing rumen fermentation [38].

Since buffalo milk contains different levels of fat, protein, and lactose compared to cow's milk, it is observed in the literature review that feed conversion ratios (FCR) are calculated using different corrected milk yields (CMY). This ratio, which is also expressed as milk production efficiency, or feed efficiency ratio, shows the milk yields obtained with 1 kg feed intake.

There was no noticeable difference between the groups regarding the ratio of CMY to DMI according to fat (FCR1) and fat and protein (FCR2). However; a trend towards an increase in FCRs was detected (Table 3) with the addition of bypass fat to the rations. It can be stated that this situation increases due to the increase in milk yield. Accordingly, the study conducted by adding bypass fat to the rations of Murrah buffaloes [24] found that the higher FCR determined by using FCM compared to fat was found to be higher than the control group.

There was no statistically significant difference between the groups regarding FCR3 obtained by the ratio of FCM to DMI according to energy. The findings of a study showed that PBF supplementation in dairy cows with high milk yield is similar to the control group calculated using FCM values according to energy, which is similar to the results of our research [39].

There was no considerable difference between the groups in terms of FCR obtained by the ratio of FCM to DMI according to solid not fat (FCR4). Similarly, more milk yield was obtained in the group ABF with 1 kg feed consumption compared to the other groups. It is thought

that the reason for this is that individual milk yield records cannot be obtained depending on the milking system and the milk of the study group is collected in a common boiler.

##### 4.1. Effects of bypass fat on milk yield and corrected milk yield

There was no notable difference between the groups regarding daily milk yields and FCM. The FCMs by fat, fat and protein, energy and solid not fat; 9.02%, respectively, in the groups to which PBF was added compared to the control group; 7.57; 10.17 and 10.20; in the groups to which ABF was added, 16.10% compared to the control group; 15.72; An increase of 21.91 and 22.08 was observed. These increases were found to be higher %6.49; %7.58; %10.65, and %10.78 in groups ABF, respectively, compared to groups PBF. Although there was a proportional difference in the results, it was not statistically significant. This might be related to the milking system of the farm and getting milk yields as a group.

Milk yield in buffaloes shows significant differences according to breeds [40]. The average annual milk yield of Anatolian buffaloes is 1000 kg [7,40], 1800–2400 kg in Nili-Ravi buffaloes, 2000–2800 kg in Italian buffaloes, 1800–2500 kg levels in Murrah breeds. Breeding studies in Anatolian water buffaloes have gained momentum in recent years while breeding studies in Italian buffaloes have been carried out for more than 40 years [41]. The milk yields of the Anatolian buffaloes used in the study were lower than the literature reports. It is thought that this is because milking occurs once a day on the farm and the newborn calves are fed with milk until the age of 4 months.

The data related to milk yield obtained in the study showed that adding bypass fats to buffalo rations did not have a statistically significant impact on milk yield, as observed by previous studies [31–33]. It is consistent with research conducted by several researchers indicating that the addition of bypass fats to dairy cow rations did not significantly affect ECM yield statistically [37,38,42]. Furthermore, it is in line with various studies which found no significant differences in milk yield, milk protein, and

lactose content as a result of adding bypass fats to lactating dairy cow rations [36,38,43]. There are also different demonstrations showing that the addition of bypass fats to buffalo rations increases milk production of twice-daily milking animals [34,44,45]. Other researchers also reported that using the Ca salts of long-chain fatty acids at 300 g per animal per day in twice-daily milking Mediterranean buffaloes with high milk yield, bypass fat applications resulted in increased milk yield and FCM significantly [44]. In summary; the aforementioned studies differ from this study on Anatolian buffaloes. It has been evaluated that there are different feed additives as well as different milking structures.

#### 4.2. Effect of bypass fat on some milk components

The level of milk fat obtained from the groups was not influenced statistically by the addition of PBF and ABF to the rations. However, compared to the control group with PBF added 4.46%; ABF sources yielded 6.05% higher fat. The group to which ABF was added resulted in 1.52% more milk fat than the group to which PBF was added.

Evaluating that adding bypass fat to the ration did not statistically affect milk fat; found coherent with studies conducted by different researchers [12,24,25,31,32,39,44,46]. However, it was not found to be compatible with several studies previously performed [33,47]. A study used choline chloride with bypass fat and reported that since choline chloride is used in phospholipid synthesis, it supports milk fat synthesis via facilitating additional lipid absorption and distribution [47]. Contrary to the study on Anatolian buffaloes, another research determined that milk fat ratio was considered to be significant since twice-daily milking and individual milk measurements were made [33].

While there was no significant difference between the daily milk fat yield groups collectively ( $p > 0.05$ ); 39 g of more milk fat was obtained from the group PBF compared to the control. Seventy-two g of more milk fat was obtained from the group PBF compared to control; 33 g of more milk fat was obtained from the group ABF compared to PBF per day. Regarding the obtained data for milk fat yields, it was found to be consistent with the results of different studies [12,25,31,37,43,45]. Various studies reported that a statistically significant increase ( $p < 0.01$ ) was procured with an average of 60 g daily milk fat yield in the twice-daily milking farms. It is evaluated that this process is due to the inability to get individual milk records due to the milking system.

There was no significant difference between the groups in terms of milk dry matter ratio and solid not fat ratio. These results were compatible with similar studies conducted on buffalo, dairy cow, and sheep species [31–34, 43, 46].

While similar values were attained between the dry matter values of milk solid not fat, it was observed that the yields of solid not fat were 20 g and 70 g higher per day,

respectively in the groups that added PBF and ABF to their rations compared to the control group. Even though the dry matter values were close to each other, this difference was due to the higher corrected milk yields in the bypass fat-added groups than in the control group.

There was no statistically significant difference between the groups in terms of milk protein and daily milk protein yield. These data were found to be correlated with different studies [24,25,34,39]. However, another study reported that milk protein levels increased significantly due to the inclusion of bypass fats in buffalo rations [32]. Researchers have reported that this may be because including bypass fats in the ration reduces the use of amino acids as an energy source and enables them to be used for casein synthesis.

There was no statistically significant difference between the groups in terms of milk lactose level and daily lactose yield similar to [12,25,34,36,37,43,46].

Nevertheless, the difference between the trial periods was significant in terms of milk fat ratio, milk fat yield, milk dry matter ratio, solid not fat yield, milk protein and lactose yields, and milk energy yield. It is thought that this situation is because although the buffaloes are divided into groups based on their similar characteristics, individual yields cannot be determined as a result of milking as a group and collecting the milk in a standard boiler. Moreover, one group has the higher milk yield.

#### 4.3 The effect of bypass fat on some milk parameters

Milk pH was not statistically affected by bypass fat supplementations, and the data obtained by a research study was found to be consistent with the values reported. Similarly, the density of milk was not statistically affected by bypass fat supplementations [48]. In fact; according to the Turkish Food Codex, the density of buffalo milk and cow milk is 1.028; sheep milk is reported to be 1.030, and goat milk is 1.026 [4]. The density values obtained were compatible with the values (1.026–1.029) obtained in a previous study [49].

The freezing point, organic matter, and mineral content of milk were not affected by bypass fat treatments ( $p > 0.05$ ). Accordingly, the urea-nitrogen of the milk was not statistically affected by the bypass fat treatments. It was found to be consistent with various former studies [38,39,50]. Milk-urea nitrogen was found to be the highest in the control group, and similar values were obtained in the groups containing bypass fat when the data of the groups fed with control and bypass fat rations were evaluated. The data obtained were found to be compatible with recent research that there is a negative correlation between the energy level in bovine rations and milk urea-nitrogen [51]. It has been evaluated that this situation is due to the decrease in the milk urea-nitrogen value of the increase in the ration energy amount since the energy level affects the protein quality and the NPN compounds used by the rumen microorganisms.

In conclusion, the addition of PBF or ABF to the rations of Anatolian buffaloes did not affect their daily dry matter intake. The inclusion of bypass fat in the rations resulted in advancements in feed conversion ratios calculated using the corrected milk yields. In particular, in the ABF group, more milk yield was obtained with each kilogram of feed dry matter intake per day. Although the daily milk yield of the groups was not statistically different ( $p > 0.05$ ), adding bypass fat to the rations positively affected the corrected milk yields. In terms of daily milk yield, the inclusion of ABF was more effective than the addition of PBF. The higher data were obtained from the groups to which bypass fat was added compared to the control group. At the same time, there was no significant difference between the groups in terms of milk fat, protein, lactose, dry matter, and solid nonfat ratios ( $p > 0.05$ ). Adding ABF to their diets was more effective. It was observed that there was no significant difference between the groups in terms of parameters such as pH, density, freezing point, ash, organic matter ratio, and milk-urea nitrogen of milk obtained by adding bypass fat to the ration ( $p > 0.05$ ). In summary, of the bypass fats added to the rations of Anatolian buffaloes of 250 g per animal per day, especially ABF; it was observed that performance, milk yield and composition improved. Starting from here; it can be said that the addition of

bypass fat to the ration has a positive effect in buffaloes and dairy cattle. However, it is thought that it would be beneficial to determine these effects by adding bypasses at different levels, including more buffaloes in the study, and diversifying them with detailed studies in more extensive scale and professional farms.

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#### Conflict of interest

The authors declare no conflict of interest.

#### References

- Atasever S, Erdem H. Manda yetiştiriciliği ve Türkiye'deki geleceği. *Ondokuz Mayıs Üniversitesi Ziraat Fakültesi Dergisi* 2008; 23 (1): 59-64 (in Turkish).
- Pasha T. Prospect of nutrition and feeding for sustainable buffalo production. *Buffalo Bulletin* 2013; 32 (Special Issue 1): 91-110.
- Aneja RP, Mathur BM, Chandan RC, Banerjee, AK. Principles of Processing, Section 2. Technology of Indian Milk and Milk Products, Dairy India Yearbook. Delhi, India; 2002
- Anonymous. Çiğ Süt ve Isıl İşlem Görmüş İçme Sütleri Tebliğinde Değişiklik Yapılması Hakkında Tebliğ. Resmi Gazete: 06.02.2009-14; 2009 (in Turkish).
- Soysal MI. Anatolian water buffalo husbandry in Turkey. *Buffalo Bulletin* 2013; 32 (Special Issue 1): 293-309.
- Borghese A. Development and perspective of buffalo and buffalo market in Europe and Near East. *Revista Veterinaria* 2010; 21: 20-31.
- Koçak S, Tekerli M, Çelikeloğlu K, Erdoğan M, Bozkurt Z et al. An investigation on yield and composition of milk, calving interval and repeatabilities in riverine buffaloes of Anatolia. *The Journal of Animal & Plant Sciences* 2019; 29 (3): 650-656.
- Purushothaman S, Kumar A, Tiwari DP. Effect of feeding calcium salts of palm oil fatty acids on performance of lactating crossbred cows. *Asian-Australian Journal of Animal Science* 2008; 21 (3): 376-385. <https://doi.org/10.5713/ajas.2008.60505>
- Ayaşan T, Karakozak E. Korunmuş yağların hayvan beslemede kullanımı. *Atatürk Üniversitesi Veteriner Bilimleri Dergisi* 2010; 6 (1): 85-94 (in Turkish).
- Sirohi SK, Walli TK, Mohanta RK. Supplementation effect of bypass fat on production performance of lactating crossbred cows. *Indian Journal of Animal Sciences* 2010; 80 (8): 733-736.
- Tyagi N, Thakur SS, Shelke SK. Effect of bypass fat supplementation on productive and reproductive performance in crossbred cows. *Tropical Animal Health and Production* 2010; 42 (8): 1749-1755. <https://doi.org/10.1007/s11250-010-9631-1>
- Obeidat BS, Awawdeh MS, Titi HH, Abu Ghazaleh AA, Al-Lataifeh FA et al. Effect of feeding calcium salts on performance of nursing awassi ewes. *Tropical Animal Health Production* 2011; 43 (6): 1211-1217. <https://doi.org/10.1007/s11250-011-9827-z>
- Rotunno T, Sevi A, Di Caterina R, Muscio A. Effects of graded levels of dietary rumen-protected fat on milk characteristics of comisana ewes. *Small Ruminant Research* 1998; 30 (2): 137-145. [https://doi.org/10.1016/s0921-4488\(98\)00096-0](https://doi.org/10.1016/s0921-4488(98)00096-0)
- Sampelayo MRS, Alonso JJM, Pe'Rez L, Extremera FG, Boza J. Dietary supplements for lactating goats by polyunsaturated fatty acid-rich protected fat. Effects after supplement withdrawal. *Journal of Dairy Science* 2004; 87 (6): 1796-1802. [https://doi.org/10.3168/jds.s0022-0302\(04\)73335-4](https://doi.org/10.3168/jds.s0022-0302(04)73335-4)



15. NRC. Nutrient Requirements of Dairy Cattle. 7th rev ed. National Academy Press, Washington D.C.; 2001. <https://doi.org/10.17226/9825>
16. AOAC. Official Methods of Analysis. 17th ed. The Association of Official Analytical Chemists; Gaithersburg, MD, USA: 2000.
17. Goering HK, Van Soest PJ. Forage Fiber Analysis. Agriculture Handbook No.379, Washington D.C.; 1970. 829-835.
18. TSI. Animal Feeds-Determination of Metabolizable Energy (Chemical Method). Turkish Standards Institute. TS 9610. Ankara, Turkey; 1991 (in Turkish).
19. Ahmadi A, Robinson PH. PC Dairy for Windows: Ration Formulation and Analysis Programs for Dairy Cattle. EFITA/WCCA. University of California Davis, CA, US; 2005; 25-28.
20. Adams RS. Penn State Forage Testing Service Revised Regression Equations. Dairy Sci. Ext. Memo DSE-90-56, The Pennsylvania State Univ., University Park; 1980.
21. Weiss WP, Conrad HR, Pierre NR. A theoretically-based model for predicting total digestible nutrient values of forages and concentrates. *Animal Feed Science and Technology* 1992; 39 (1-2): 95-110. [https://doi.org/10.1016/0377-8401\(92\)90034-4](https://doi.org/10.1016/0377-8401(92)90034-4)
22. Nawaz H, Yaqoob M, Abdullah M. Effect of feeding supplemental tallow on the performance of lactating Nili-Ravi buffaloes. *Turkish Journal of Veterinary and Animal Sciences* 2007; 31 (6): 389-398.
23. Tanh LP, Suksombat W. Milk yield, composition, and fatty acid profile in dairy cows fed a high-concentrate diet blended with oil mixtures rich in polyunsaturated fatty acids. *Asian-Australasian Journal of Animal Sciences* 2015; 28 (6): 796-806. <https://doi.org/10.5713/ajas.14.0810>
24. Sharma S, Singh M, Roy AK, Thakur S. Effect of pre-partum prilled fat supplementation on feed intake, energy balance and milk production in Murrah buffaloes. *Veterinary World* 2016; 9 (3): 256-259. <https://doi.org/10.14202/vetworld.2016.256-259>
25. Hafez, YM. Enhancing milk production in periparturient buffalo fed protected fat with and without vitamin AD3E treatment. *Egyptian Journal of Animal Production* 2012; 49 (3): 249-256. <https://doi.org/10.21608/ejap.2012.94315>
26. Peterson SE, Rezamand P, Williams JE, Price W, Chanine M. et al. Effects of dietary betaine on milk yield and milk composition of mid-lactation Holstein dairy cows. *Journal of Dairy Science* 2012; 95: 6557-6562. <https://doi.org/10.3168/jds.2011-4808>
27. Chaney AL, Marbach EP. Modified reagents for determination of urea and ammonia. *Clinical Chemistry* 1962; 8 (2): 130-132. <https://doi.org/10.1093/clinchem/8.2.130>
28. Weatherburn MW. Phenol-hypochlorite reaction for determination of ammonia. *Analytical Chemistry* 1967; 39: 971-974. <https://doi.org/10.1021/ac60252a045>
29. Bressler LF, Steinbrink F. Stabilized Phenol Nitroprusside Reagent and Analysis of Nitrogen. United States Patent: 3769172. USA; 1973.
30. Dawson B, Trapp RG. Basic and Clinical Biostatistics. 3rd ed Lange Medical Books/McGraw-Hill Medical Publishing Division, New York; 2001.
31. Ranjan A, Sahoo B, Singh VK, Srivastava S, Singh SP et al. Effect of bypass fat supplementation on productive performance and blood biochemical profile in lactating Murrah (*Bubalus bubalis*) buffaloes. *Tropical Animal Health and Production* 2012; 44: 1615-1621. <https://doi.org/10.1007/s11250-012-0115-3>
32. Sarwar M, Khan MA, Nisa M. Influence of ruminally protected fat and urea treated corncobs ensiled with or without corn steep liquor on nutrient intake, digestibility, milk yield and its composition in Nili-Ravi buffaloes. *Asian-Australasian Journal of Animal Sciences* 2004; 17 (1): 86-93. <https://doi.org/10.5713/ajas.2004.86>
33. Savsani HH, Murthy KS, Gajbhiye PU, Vataliya PH, Bhadaniya AR et al. Milk yield and composition and efficiency of nutrients for milk production in Jaffrabadi Buffaloes on rations supplemented with varying levels of bypass fat. *Buffalo Bulletin* 2015; 34 (1): 113-123.
34. Shelke SK, Thakur SS, Amrutkar SA. Effect of feeding protected fat and proteins on milk production, composition and nutrient utilization in Murrah buffaloes (*Bubalus bubalis*). *Animal Feed Science and Technology* 2012; 171: 98-107. <https://doi.org/10.1016/j.anifeeds.2011.10.003>
35. Thakur SS, Shelke SK. Effect of supplementing bypass fat prepared from soybean acid oil on milk yield and nutrient utilization in Murrah buffaloes. *Indian Journal of Animal Science* 2010; 80: 354-357.
36. Chen KJ, Jan DF, Chiou PWS, Yang DW. Effects of dietary heat extruded soybean meal and protected fat supplement on the production, blood and ruminal characteristics of Holstein cows. *Asian-Australasian Journal of Animal Sciences* 2002; 15 (6): 821-827. <https://doi.org/10.5713/ajas.2002.821>
37. Duske K, Hammon HM, Langhof AM, Bellmann O, Losand B et al. Metabolism and lactation performance in dairy cows fed a diet containing rumen-protected fat during the last twelve weeks of gestation. *Journal of Dairy Science* 2009; 92 (4): 1670-1684. <https://doi.org/10.3168/jds.2008-1543>
38. Ganjkanlou M, Rezayazdia K, Ghorbanib GR, Banadaky DM, Morravega H et al. Effects of protected fat supplements on production of early lactation Holstein cows. *Animal Feed Science and Technology* 2009; 154: 276-283. <https://doi.org/10.1016/j.anifeeds.2009.09.008>
39. Rico DE, Ying Y, Harvatine KJ. Comparison of enriched palmitic acid and calcium salts of palm fatty acids distillate fat supplements on milk production and metabolic profiles of high-producing dairy cows. *Journal of Dairy Science* 2014; 97 (9): 5637-5644. <https://doi.org/10.3168/jds.2013-7723>
40. Borghese, A. Buffalo Production and Research. FAO ed REU Technical Series 2005; 67: (1-315).
41. Borghese A, Boselli C. Buffalo recording, udder physiology and milkability as factors of milk availability in the world economy. VIII. Asian Buffalo Congress 2015; Book of Abstracts, İstanbul; 2015.

42. Alstrup L, Hellwing ALF, Lund P, Weisbjerg MR. Effect of fat supplementation and stage of lactation on methane production in dairy cows. *Animal Feed Science and Technology* 2015; 207 (9): 10-19. <https://doi.org/10.1016/j.anifeedsci.2015.05.017>
43. Kitessa SM, Gulati SK, Simos GC, Ashes JR, Scott TW et al. Supplementation of grazing dairy cows with rumen-protected tuna oil enriches milk fat with N-3 fatty acids without affecting milk production or sensory characteristics. *British Journal of Nutrition* 2004; 91 (2): 271-277. <https://doi.org/10.1079/BJN20031050>
44. Polidori F, Sgoifo Rossi CA, Senatore EM, Savoni G, Dellorto V. Effect of recombinant bovine somatotropin and calcium salts of long chain fatty acids on milk from Italian buffalo. *Journal of Dairy Science* 1997; 80 (9): 2137-2142. [https://doi.org/10.3168/jds.S0022-0302\(97\)76159-9](https://doi.org/10.3168/jds.S0022-0302(97)76159-9)
45. Vahora SG, Parnerkar S, Kore KB. Productive efficiency of lactating buffaloes fed bypass fat under field conditions: Effect on milk yield, milk composition, body weight and economics. *Iranian Journal of Applied Animal Science* 2013; 3 (1): 53-58.
46. Sampelayo MRS, Pe´Rez L, Alonso JJM, Amigo L, Boza J. Effects of concentrates with different contents of protected fat rich in PUFAs on the performance of lactating Granadina goats: Part II. Milk production and composition. *Small Ruminant Research* 2002; 43 (2): 141-148. [https://doi.org/10.1016/S0921-4488\(02\)00007-X](https://doi.org/10.1016/S0921-4488(02)00007-X)
47. Garg MR, Sherasia PL, Bhanderi BM. Effect of supplementing bypass fat with and without rumen protected choline chloride on milk yield and serum lipid profile in Jaffarabadi buffaloes. *Buffalo Bulletin* 2012; 31 (2): 91-97.
48. Barile VL, Tripaldi C, Pizzoferrato L, Pacelli C, Palocci G et al. Effects of different diets on milk yield and quality of lactating buffaloes: Maize versus sorghum silage. *Italian Journal of Animal Science* 2007; 6 (Suppl. 2): 520-523. <https://doi.org/10.4081/ijas.2007.s2.520>
49. Kanwal R, Ahmed T, Mirza B. Comparative analysis of quality of milk collected from buffalo, cow, goat and sheep of Rawalpindi/Islamabad Region in Pakistan. *Asian Journal of Plant Sciences* 2004; 3 (3): 300-305. <https://doi.org/10.3923/ajps.2004.300.305>
50. Wyngaard JDV, Meeske R, Erasmus LJ. Effect of palm kernel expeller as supplementation on production performance of Jersey cows grazing kikuyu-ryegrass pasture. *Animal Feed Science and Technology* 2015; 199: 29-40. <https://doi.org/10.1016/j.anifeedsci.2014.10.017>
51. Keser O, Alp M, Kutay HC, Demirel G, Kocabağlı N. Süt ineklerine uygulanan farklı besleme yöntemlerinin süt üre nitrojeni ve bileşimi yönünden değerlendirilmesi. *Lalahan Hayvancılık Araştırma Enstitü Dergisi* 2017; 57 (2): 83-87 (in Turkish).