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Effects of Wafer Addition on Fermentation Properties, Aerobic Stability and Feed Value of Alfalfa Silages

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Abstract

This study was planned to determine the effects of waste wafer addition on fermentation, aerobic stability, and in vitro digestibility of alfalfa silages. Experimental groups consisted of the G20:20 g kg^{-1} wafer, G30:30 g kg^{-1} wafer, G40:40 g kg^{-1} wafer, G50:50 g kg^{-1} wafer, and the control (CONT) group without any additives. The wafer was added to 1 kg of wilting alfalfa. Except for the G20 group, the addition of wafers increased the amount of crude protein (CP) and decreased the neutral detergent fiber (NDF) (P<0.01). While lactic acid bacteria (LAB) counts increased in the wafer groups compared to the control group (P<0.01), yeast counts also increased. Adding wafers increased the relative feed value (RFV) of alfalfa silages, the amount of organic matter dissolved in the enzyme, and the ME_{ESOM} content.

The study concluded that high-quality alfalfa silages could be made by adding wafers to alfalfa with low water-soluble carbohydrate content in regions where the food industry is dense.

Keywords: Alfalfa; Wafer; Waste; Silage

Introduction

It is of great importance that the feeds used in the rations of ruminants are of high quality and cheap the inclusion of roughage such as dry grass, green fodder, and silage in the rations increases the profitability of livestock enterprises by reducing the costs of the ration [1]. However, roughage supply is still an important problem today [2]. Alfalfa is defined as the queen of forage crops due to its nutrient content. In our country, alfalfa is mostly used as dry grass in animal feed. However, significant nutrient losses occur during drying [3]. In recent years, alfalfa silage has also gained importance. Especially in regions with abundant rainfall and without adequate drying opportunity, the final form of alfalfa is generally considered as silage [4]. As a silage green fodder, alfalfa is classified as difficult ensiled feeds due to its high protein level and buffer capacity, and low water-soluble carbohydrate (WSC) content [5].

For this reason, it is sometimes necessary to use additives to ensure fermentation during the silage of forage crops rich in protein and mineral substances and poor in carbohydrates [6]. For this purpose, different additives (bacterial inoculants, enzymes, acids, urea, and ammonia) are used, but mainly additives aiming at filling the insufficient carbohydrate deficit in the environment are used. Raw materials such as cereal grains, molasses, grape pomace, and sugar are mainly used as a source of carbohydrates [7].

Food industry wastes as an alternative feed source have attracted the attention of animal nutritionists, and many studies have been conducted on evaluating food industry wastes in ruminant feeding. Food industry wastes are generally destroyed by incineration. Apart from this, it causes environmental pollution by being left in piles directly to nature. During wafer production, when the wafers are cut into rectangles or cubes, or when they are on the market shelves, the wafers break down and form crumbs due to the placement of the packages on the shelves, their displacement, and the consumers touching the packages. Since consumers do not prefer these packages, crumb wafer packages are separated as waste material. However, before using new products such as wafers in animal nutrition, it is necessary to examine them in terms of content and limiting factors, and studies should be conducted to reveal their feed values [8]. Waste crumb wafers can be used in animal feed to increase the water-soluble carbohydrate content in alfalfa's first and last cutting. However, no studies on this have been found.

This research was planned to determine the effects of crumb wafer addition on fermentation, aerobic stability, and feed value of alfalfa silage.

Materials and Methods

Experimental Design and Ensiling Process

The research material consisted of alfalfa (fifth cutting) harvested at the beginning of flowering in the last week of October and crumb wafer. Crumb wafer is made of cocoa wafers, which are produced in cubes and become crumbs (consumers do not prefer this form) because they stay on the market shelf for a long time. The remaining big pieces of the wafer were thoroughly crushed with the help of a pestle and homogenized. After harvesting, alfalfa was withered under laboratory conditions for 18 hours and then chopped into approximately 1.5-2.0 cm in size with a silage machine. Experimental groups consisted of the G20:20 g kg-1 wafer, G30:30 g kg-1 wafer, G40:40 g kg-1 wafer, G50:50 g kg-1 wafer, and the control (CONT) group without any additives. The additives were added to 1 kg of wilting alfalfa.

The research was carried out in 5 groups, four in parallel. The alfalfa, mixed with the crumb wafer, was placed in plastic bags, and the air inside was removed by vacuum and covered with stretch film (16-17 layers). Then, a layer of tape was passed and made it completely airtight. A total of 20 packs of silage (10-16°C) were left to ferment for 60 days under laboratory conditions.

Physical and Chemical Analysis

Three different observers scored the silages on the day they were opened (60th day) in terms of color, odor, and structure (Deutsche Landwirtschafts Gesellschaft: DLG) [5]. Evaluation according to DLG, 16-20: excellent; 10-15: moderate; 5-9: medium; 0-4: poor. The pH of the silages was determined with a digital pH meter, the buffer capacity according to Playne and McDonald [9], and LA by the spectrophotometric method [10]. Ammonia nitrogen (NH_3-N) and WSC contents were determined according to Anonymous [11]. An eight-days aerobic stability test was carried out on samples developed by Ashbell, et al. [12]. Flieg score was calculated from the dry matter and pH values of silages according to the formula below [5].

Flieg score = 220 + (2 x % DM - 15)-40 x pH.

According to this index, silage was considered "poor" when it had a score of <20; to be "low" with a score between 21 and 40; to be "medium" with a score between 41 and 60; to be "good" quality with a score between 61 and 80; and to be "excellent" when it had a score between 81 and 100 [5].

Microbial Populations

Total mesophilic aerobic bacteria (TMAB), Lactobacilli, yeast, and mold analyzes were determined by the method developed by Seale, et al. [13]. MRS agar (de Man Rogosa and Sharpe agar, Merck, Darmstadt, Germany) was used to detect lactobacilli. In the enumeration of the yeast, malt extract agar, and for enterobacter, violet red bile agar was used. The plates were incubated for 3 days at 30°C. The Lactobacilli, mold, and yeast numbers of the silages were converted into logarithmic colony form units (cfu g-1).

Nutrient Analysis and in Vitro Digestibilty

The DM, crude ash (CA), and crude protein (CP) contents of the starting alfalfa and silages were determined according to the Weende analysis method [14]. NDF, acid detergent insoluble fiber (ADF), and acid detergent insoluble lignin (ADL) contents, were determined according to the methods reported by Van Soest, et al. [15]. Pepsin-cellulase digestibility was determined according to a modification of De Boever, et al. method [16]. In the technique [17], pre-treatment with the pepsin-hydrochloric acid solution followed an incubation in water at 80°C for 45 minutes before the treatment by cellulase (Onozuka R 10 from Trichoderma viride, Merck). The solubility of the organic matter in cellulase (ELOS) was derived as follows [17]:

ELOS (%) = DM - CA – G

G= Loss upon ashing

Metabolizable Energy Value Estimating

In vitro ME contents in examples were calculated using crude nutrition components (CNC), NDF, ADF, ADL and ELOS determined because of chemical analysis according to the equation given below:

ME_{CNC} kcal/kg OM*= 3260 + (0.455 x CP* + 3.517 x EE*) – 4.037 x CF* [18].

(*in Organic matter (OM) g/kg).

ME_{NDF}, kcal/kg DM=3381.9-19.98 x NDF* [19].

ME_{ADF}, MJ/kg DM= 14.70-0.150 x ADF* [20].

ME_{ADL}, kcal/kg DM=2764.4-102.73 x ADL* [19].

* NDF, ADF and ADL in %, ME contents were translated into

kilocalories.

ME_{ESOM}, MJ/kg DM=12.6 CP+22.5 CF+11.2 NFE + 0.3975 CA x EE – 0.1993 CA x CF + 0.2449 ELOS2 –150) x 10-3 [21].

*(CP, NFE, EE, CF, CA g/kg; ELOS in g/kg DM).

The equities stated below, which were developed by Van Dyke and Anderson [22], were used for relative feed value detection of silages.

Digestible Dry Matter (DDM), % =88.9 – (0.779 x %ADF)

Dry Matter Intake (DMI), %= 120 / %NDF

Relative Feed Value (RFV) =DDM% x DMI% x 0.775

Statistical Analyses

The statistical analyses were performed using SPSS software v.18 suite [23]. The fermentation characteristics and microbial quantity of silage from ensiling to aerobic conditions were analyzed via one-way ANOVA. Duncan's test was used to compare the differences between group averages [24].

Results and Discussion

According to the physical evaluation of the silages opened on the 60th day of ensiling (Table 1), the addition of crumb wafers resulted in the formation of silages with green color (except G30), a pleasant and slightly acidic odor, and intact stem and leaf integrity. When the obtained scores were compared with the total scores of the silages made by Malhatun-Çotuk and Soycan-Önenç [25] by adding bran and pudding to alfalfa, it was seen that the scores of the crumb wafer groups were higher.

Item	CONT	W20	W30	W40	W50	*p*-Value
Smell	14	14	14	14	14	-
Structure	4.00	4.00	4.00	4.00	4.00	-
Colours	1.00	2.00	1.00	1.00	2.00	-
DLG point	19	20	19	19	20	-
Quality	Excellent	Excellent	Excellent	Excellent	Excellent	
Flieg Point	75.40±1.13^{c}	94.44±1.24^{ab}	96.43±1.32^{ab}	91.07±2.42^{b}	98.51±1.20^{a}	<0.001
Quality	Good	Excellent	Excellent	Excellent	Excellent	-

CONT: Control, W20:20 g wafer, W30:30 g wafer, W40:40 g wafer, W50:50 g wafer, a-c: Means with different letters in the same line are statistically significant (p<0.01). According to Flieg score, silage was considered "poor" when it had a score of <20; to be "low" with a score between 21 and 40; to be "medium" with a score between 41 and 60; to be "good" quality with a score between 61 and 80; and to be "excellent" when it had a score between 81 and 100.

Table 1: The effects of different wafer levels on alfalfa silage qualities (n=4).

The browning seen in the G30 group may have been caused by withering. Browning in color may occur in withered silages [5]. While Flieg scores were higher than Malhatun-Çotuk and Soycan-Önenç [25], it was found that the silages that Yayla and Soycan Önenç [26] added jam to alfalfa were between Flieg scores.

Item	CONT	W20	W30	W40	W50	P-Value
OM	89.31±0.33^{c}	89.41±0.10^{c}	89.58±0.03^{bc}	89.82±0.07^{b}	90.18±0.07^{a}	<0.001
CA	10.69±0.33^{a}	10.59±0.10^{a}	10.41±0.03^{ab}	10.18±0.07^{b}	9.81±0.06^{c}	<0.001
CP	24.40±0.05^{c}	24.35±0.06^{c}	25.37±0.04^{b}	25.12±0.07^{b}	25.79±0.10^{a}	<0.001
EE	3.13±0.06^{d}	5.06±0.08^{c}	5.32±0.05^{c}	5.76±0.08^{b}	7.02±0.06^{a}	<0.001
CF	23.86±0.17^{b}	21.07±0.13^{d}	21.58±0.05^{d}	23.19±0.10^{c}	25.83±0.14^{a}	<0.001
NFE	37.20±0.18^{a}	37.49±0.08^{a}	37.31±0.05^{a}	36.47±0.01^{b}	32.98±0.13^{c}	<0.001
NDF	33.55±0.12^{a}	33.67±0.12^{a}	31.53±0.07^{c}	31.35±0.06^{c}	32.79±0.12^{b}	<0.001
ADF	26.19±0.08^{b}	23.91±0.10^{d}	23.15±0.07^{e}	24.69±0.06^{c}	27.62±0.09^{a}	<0.001
ADL	5.33±0.13^{a}	5.33±0.06^{a}	5.08±0.08^{ab}	4.51±0.06^{c}	4.79±0.06^{bc}	<0.001

CONT: Control, W20:20 g wafer, W30:30 g wafer, W40:40 g wafer, W50:50 g wafer, OM: Organic matter, CA: Crude ash, CP: Crude protein, EE:Ether extract, CF:Crude fiber, NFE: Nitrogen-free extract NDF: Neutral detergent fiber, ADF: Acid detergent fiber, ADL: Acid detergent lignin, a-c: Means with different letters in the same line are statistically significant (p<0.01).

Table 2: Chemical compositions of the alfalfa silages (% in DM).

In the study determined that the amount of CA decreased in the W40 and W50 groups (Table 2) compared to the CONT group. Şakalar and Kamalak [27] reported that increasing the addition of dried molasses beet pulp to alfalfa reversed the rate of increase of CA due to the CA content of dried molasses beet pulp. The decrease in the amount of CA in the W40 and W50 groups inthis study was similar to that of Şakalar and Kamalak [27] and was caused by the low amount of CA in the wafer.

Withering in good weather conditions can improve the protein quality of silage by preventing protein hydrolysis [28]. In this study, adding 30 g kg-1 WA and above crumb wafer to withered alfalfa prevented CP fragmentation, especially in G50; the CP amount was found to be 25.79%. Increasing the WSC content with the addition of wafers caused a rapid increase in the Lactobacilli counts, lowering the pH and inhibiting proteolysis in low pH silages (4.43-4.67). Proteolytic enzymes can reduce the feed value of the silage material. These enzymes convert protein nitrogen into non- protein nitrogen (NPN) forms, such as peptides and free amino acids. However, it is known that microbial activity in silage plays a significant role in the degradation of ammonia and amines [28]. In the study, NH_3-N levels decreased significantly by adding 40 and 50 g kg-1 WA crumb wafers, indicating that the wafer successfully prevents proteolysis. It has been reported that alfalfa silage causes 85% of the total N in alfalfa to become NPN [28]. Also, when large amounts of proteolysis occur, additional protein supplements may have to be used even if the total CP of the ratio appears sufficient to achieve optimum milk production. Therefore, proteolysis in silage-making can seriously affect the cost of milk production [6].

The EE content was higher in the groups with wafers added than in the control group (Table 2). However, the highest value in the G50 group was due to the high amount of added wafers. However, the fact that the wafer contained 23.13% EE in DM explains the increase in EE in the G50 group.

In the study, adding 30 and 40 g of the wafer decreased NDF and ADF while adding 50 g increased them (Table 2), and this may be related to the fact that the high WSC in the wafer was used together with Lactobacilli by yeasts and the relaxant effect of LA on the cell wall could not be revealed. The fact that the pH levels of the wafer-added groups are close to each other and above 4.5 also supports this situation. However, in Özaslan›s [29] study, it was reported that the cell wall components decreased with the addition of corn syrup, and the silage pH was around 4.22.

Item	CONT	W20	W30	W40	W50	P-Value
DM,%	36.53±0.13^{d}	38.05±0.06^{c}	38.38±0.02^{c}	39.03±0.06^{b}	40.09±0.08^{a}	<0.001
pH	5.07±0.03^{a}	4.67±0.03^{b}	4.63±0.03^{b}	4.80±0.06^{b}	4.67±0.03^{b}	<0.001
WSC, g kg^{-1} DM	28.41±0.11^{e}	41.53±0.04^{d}	51.96±0.08^{c}	60.14±0.07^{b}	77.39±0.10^{a}	<0.001
LA, g kg^{-1} DM	32.16±0.13^{e}	80.20±0.10^{d}	101.03±0.03^{c}	111.32±0.11^{b}	126.08±0.05^{a}	<0.001
NH_3-N, g kg^{-1} TN	7.70±0.13^{a}	7.60±0.11^{a}	7.18±0.21^{a}	6.10±0.11^{b}	4.42±0.10^{c}	<0.001
DM loss,%	0.86±0.03^{a}	0.83±0.01^{a}	0.72±0.01^{b}	0.70±0.01^{b}	0.69±0.01^{b}	<0.001

CONT: Control, W20:20 g wafer, W30:30 g wafer, W40:40 g wafer, W50:50 g wafer, DM:Dry matter, WSC: Water-soluble carbohydrates, LA: Lactic acid, NH_3-N: Ammonia nitrogen, TN: Total nitrogen a-c: Means with different letters in the same line are statistically significant (p<0.01).

Table 3: Fermentation quality of the alfalfa silages.

In a study [30], in which 5% and 10% dried molasses sugar beet pulp (DMSBP) was added to alfalfa [30], it was reported that the DM content increased depending on the added amount of DMSBP. In our study, similar to Yakışır and Aksu [30], DM contents of silages increased due to the increase in the amount of wafers. However, the reason for the high DM content of the control group was due to the high DM of the starting materials (37.03% of alfalfa and 98.24% of the crumb wafer) (Table 3). Kurtoğlu [31] reported that with the withering up to 37% DM, the desired quality of silage fermentation would be achieved. In the study, the starting DM content of alfalfa with withering increased to 37.03%; therefore, as Kurtoğlu [31] reported, the DM content of the control group silages was also found to be high. However, it is thought that the effect of the wafer may be more pronounced in alfalfa with low DM.

In order to obtain quality silage from legume forage crops, it is very important to do the withering process before silage, to choose the most suitable silage additive, and to use it at sufficient levels. It is difficult to obtain quality silage without additives from legumes [31]. For this purpose, alfalfa was first withered in laboratory conditions for 18 hours after cutting, and then crumb wafer with high WSC content (Table

3) was added. In silage production, there must be LAB in the environment so that the silage material does not deteriorate, and WSC content must be sufficient for them to produce lactic acid [32]. In the study, the addition of wafers showed an encouraging effect on the development of Lactobacilli counts (Table 4); while the Lactobacilli counts increased, the yeast counts increased due to the increase in the wafer level. It is reported that carbohydrate sources activate the proliferation of some anaerobic bacteria, primarily LAB, in silage [33]. The increase in Lactobacilli and yeast counts was due to the addition of crumb wafers, a carbohydrate source, as reported by Bolsen, et al. [33]. Although an increase in LAB counts is desired in silages, an increase in yeast counts is not desired. However, due to the high WSC content of the added wafer in this study, the yeast counts were increased along with the Lactobacilli, which is not an expected result.

Item	CONT	W20	W30	W40	W50	*P*-Value
TMAB	5.98±0.04^{b}	6.42±0.14^{a}	6.42±0.06^{a}	6.27±0.02^{ab}	6.11±0.09^{ab}	<0.001
Lactobacilli	5.72±0.07^{b}	6.23±0.05^{a}	6.23±0.06^{a}	6.20±0.08^{a}	6.00±0.06^{ab}	<0.001
Yeast	4.20±0.10^{b}	4.59±0.06^{a}	4.62±0.09^{a}	4.68±0.10^{a}	4.69±0.05^{a}	<0.001
Mould	ND	ND	ND	ND	ND	-
Enterobacter	ND	ND	ND	ND	ND	-

CONT: Control, W20:20 g wafer, W30:30 g wafer, W40:40 g wafer, W50:50 g wafer, TMAB:Total mesophilic aerobic bacteria, ND: Not detected, a,b: Means with different letters in the same line are statistically significant (p<0.01).
Table 4: Microbiological analysis results of alfalfa silages, log10 cfu g-1.

The addition of wafers to alfalfa silages promoted the development of yeast counts on the 3rd, 5th, and 7th days of aerobic stability (Table 5).

Item	CONT	W20	W30	W40	W50	*P*-Value
3.day						
DM	37.50±0.05^{e}	39.13±0.11^{d}	40.48±0.11^{c}	41.97±0.05^{b}	42.81±0.09^{a}	<0.001
pH	5.07±0.03^{a}	4.66±0.03^{b}	4.63±0.03^{b}	4.80±0.06^{b}	4.63±0.03^{b}	<0.001
CO_2 g kg^{-1} DM	3.13±0.16^{a}	2.84±0.15^{ab}	2.68±0.03^{ab}	2.93±0.01^{a}	2.45±0.01^{b}	<0.001
Yeast	5.38±0.06^{c}	5.52±0.04^{bc}	5.86±0.02^{a}	5.67±0.06^{ab}	5.74±0.02^{a}	<0.001
Mould	ND	ND	ND	ND	ND	-
5.day						
DM	37.21±0.08^{e}	39.18±0.09^{d}	40.14±0.07^{c}	42.75±0.04^{b}	43.10±0.02^{a}	<0.001
pH	5.13±0.03^{a}	4.67±0.03^{b}	4.63±0.03^{b}	4.70±0.06^{b}	4.66±0.03^{b}	<0.001
CO_2 g kg^{-1} DM	4.55±0.15^{a}	4.35±0.30^{a}	3.51±0.06^{c}	3.77±0.04^{ab}	4.25±0.13^{ab}	<0.001
Yeast	5.85±0.05^{ab}	5.94±0.02^{ab}	5.89±0.04^{ab}	5.77±0.01^{b}	5.98±0.05^{a}	<0.001
Mould	ND	ND	ND	ND	ND	-
7.day						
DM	36.10±0.15^{e}	38.57±0.05^{d}	39.34±0.04^{c}	42.10±0.02^{b}	43.74±0.05^{a}	<0.001
pH	5.13±0.03^{a}	4.87±0.03^{b}	4.73±0.03^{b}	4.83±0.03^{b}	4.73±0.03^{b}	<0.001
CO_2 g kg^{-1} DM	5.99±0.16^{a}	4.87±0.15^{b}	4.64±0.28^{b}	4.66±0.13^{b}	4.29±0.13^{b}	<0.001
Yeast	5.94±0.03^{bc}	6.12±0.03^{a}	6.17±0.04^{a}	5.81±0.04^{c}	6.01±0.04^{ab}	<0.001
Mould	ND	ND	ND	ND	ND	-

CONT: Control, W20:20 g wafer, W30:30 g wafer, W40:40 g wafer, W50:50 g wafer, DM:Dry matter, TMAB:Total mesophilic aerobic bacteria, ND: Not detected, a-e: Means with different letters in the same line are statistically significant (p<0.01).
Table 5: Aerobic stability test results of alfalfa silages.

In all groups, yeast counts were above the critical level (5 cfu g-1) during the aerobic period. In addition, withering is also reported to promote yeast growth [31]. There was no mold growth in the anaerobic and aerobic periods. The absence of mold growth in the aerobic period is associated with snowfall in the province of Tekirdağ and laboratory conditions after the silage is opened, with the air temperature falling below zero. This period's low DM, pH, and CO2 production (Table 5) are related to weather conditions. During the aerobic period, the air temperatures were low, the highest air temperature was 8 ºC, and the lowest air temperature was 0 ºC in laboratory conditions.

It is reported that alfalfa silages obtained by adding 3.0% corn syrup to the alfalfa harvested during the flowering period have a high degree of digestion and metabolic energy content; therefore, it is possible to produce high-quality alfalfa silage with the addition of corn syrup [29]. In the study, adding crumb wafers increased the ELOS content of alfalfa silages (Table 6), and accordingly, the amount of MEELOS increased.

Item	CONT	W20	W30	W40	W50	*P*-Value
ELOS,% DM	60.38±0.12^{d}	61.21±0.20^{c}	61.58±0.05^{c}	65.22±0.12^{a}	64.37±0.14^{b}	<0.001
ME_{ELOS}	1162.8±7.9^{e}	1410.1±12.8^{d}	1482.9±10.6^{c}	1662.7±18.8^{b}	1932.0±19.2^{a}	<0.001
ME_{CNC}	2172.9±9.8^{d}	2359.5±4.6^{a}	2351.9±1.0^{a}	2305.6±4.9^{b}	2254.6±7.4^{c}	<0.001
ME_{NDF}	2711.6±2.5^{c}	2752.0±1.8^{a}	2751.9±1.3^{a}	2722.6±2.0^{b}	2717.0±1.9^{bc}	<0.001
ME_{ADF}	2574.6±3.0^{d}	2656.2±3.5^{b}	2683.5±2.6^{a}	2628.1±2.1^{c}	2523.1±3.1^{e}	<0.001
ME_{ADL}	2217.1±13.7^{c}	2216.9±6.7^{c}	2242.5±8.5^{bc}	2301.4±6.0^{a}	2272.2±6.5^{ab}	<0.001

CONT: Control, W20:20 g wafer, W30:30 g wafer, W40:40 g wafer, W50:50 g wafer, a-e: Means with different letters in the same line are statistically significant (p<0.01).
Table 6: ELOS and ME contents of alfalfa silages, kcal kg^{-1} DM.

Consistent with Özaslan›s [29] study, adding crumb wafers improved the feed value of alfalfa silages. It is known that changes in the CNC and cell wall contents of the feeds increase the ME content of the feeds and, thus, the feed value. In this study, the changes in the mentioned nutrients were also reflected in ME_{CNC}, ME_{NDF}, ME_{ADF}, and ME_{ADL}, and the addition of wafers increased the ME content of alfalfa silages. In ruminant animals, the NDF content of the roughage is an indicator of feed consumption, and the ADF and ADL contents are accepted as a measure of the digestibility of the roughage.

Item	CONT	W20	W30	W40	W50	*P*-Value
DDM,%	68.50±0.06^{d}	70.27±0.08^{b}	70.87±0.06^{a}	69.66±0.05^{c}	67.38±0.07^{e}	<0.001
DMI,%	3.58±0.01^{c}	3.56±0.01^{c}	3.81±0.01^{a}	3.83±0.01^{a}	3.66±0.01^{b}	<0.001
RFV	189.91±0.86^{c}	194.12±0.87^{b}	209.03±0.53^{a}	206.63±0.32^{a}	191.09±0.89^{bc}	<0.001

CONT: Control, W20:20 g wafer, W30:30 g wafer, W40:40 g wafer, W50:50 g wafer, DDM:Digestible dry matter, DMI:Dry matter intake, RFV:Relative feed value, a-e: Means with different letters in the same line are statistically significant (p<0.01).
Table 7: Dry matter digestibility, dry matter intake and relative feed value of alfalfa silages.

This study showed significantly higher DDM, DMI, and RFV contents in G40 and G50 groups (Table 7) increased alfalfa silage consumption and digestibility. The research's RFV contents were similar to the study of Malhatun-Çotuk and Soycan-Önenç [25].

Conclusion

In the study, adding crumb wafer to alfalfa at a WA level of 40 and 50 g kg-1 positively affected the chemical and microbiological properties of silages. The wafer added to increase the carbohydrate level increased the Lactobacilli counts and efficiency by encouraging the development of Lactobacilli. However, enzymes that break down proteins were inhibited, and the degradation of proteins into ammonia was also reduced. In addition, the amount of organic matter dissolved in the enzyme increased, resulting in a parallel increase in MEELOS content. Similarly, the relative feed value and dry matter consumption rate also increased. In the study, the high yeast counts were due to the high yeast counts at the beginning and the WSC content of the wafer. It is recommended that future studies be conducted with starting materials with low DM.

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