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EFFECTS OF COMMERCIAL BACTERIAL INOCULANTS ON FERMENTATION AND NUTRITIVE QUALITY OF WHEAT AND ANNUAL LEGUME MIXED SILAGES

IBRAHIM ERTEKIN*

Department of Field Crops, Faculty of Agriculture, Hatay Mustafa Kemal University, Hatay, 31060, Turkey

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Abstract

In the present study fermentation quality, microbial populations and nutritive value of wheat and some cool season annual legume (forage pea, grass pea and vetch) mixed silages (25:75, 50:50 and 75:25) using various commercial bacterial inoculants (Pioneer 1188, Pioneer 11A44 and Pioneer 11CFT) were determined. Wheat and legume species were sown in separate plots and these plants were harvested when wheat reached the dough stage about 180 days after plant emergence. The present study was conducted in a factorial (3 legume species×3 inoculants×5 mixtures) arrangement design. Results showed that effects of legume species on ADF, crude ash, pH, lactic acid bacteria, *enterobacteria*, yeast and mold, lactic acid, acetic acid, propionic acid, butyric acid and ammonia nitrogen were significant ($P < 0.05$). Crude protein, silage fermentation features and microbial populations were affected ($P < 0.05$) by inoculants. Effects of mixture ratios on dry matter, chemical compositions, relative feed value, fermentation parameters of silages were significant ($P < 0.05$). It was determined that ensiling the 50 % wheat with 50% grass pea with the use of inoculant 1188 as an additive could give a good result both in terms of nutritive value and fermentation quality.

Introduction

In developing countries such as Turkey, ruminant animals cannot be fed adequately with roughage. These countries carry out the culture of summer industrial plants in most of the agricultural lands within the scope of agricultural production. For this reason, industrial plants production fields stay empty during the winter season (Kaplan and Gökkuş 2018). However, during this period, especially annual legumes and wheat crops can be cultivated effectively on these fallow fields and the fresh crops obtained from these plants can be made into silage.

For more than 3000 years, ensiling is a method that has been practiced to preserve fresh forage plants and by-products (Woolford 1984). Ensiling means the fermentation of fresh plants by storing them in an anaerobic environment, and the product obtained this process is called silage. Silage production has been increasing in many countries recently compared to hay production. It offers advantages over hay making because silage making is less dependent on weather conditions and allows producers to conserve high-quality forage (Copani *et al*. 2016). The success of ensiling fresh forages in silos depends on the plant type, harvest time, silo type, silo management and additives (Ertekin and Kızılşimşek 2020, Ertekin *et al.* 2021). Lactic acid bacteria are some of the most widely used additives. Adding lactic acid bacteria to silages improves silage fermentation and helps to obtain a healthier silage (Ertekin and Kızılşimşek 2020, and Kung *et al.* 2018). Some lactic acid bacteria inoculants are commercially used to reduce the losses during ensiling (Contreras-Govea and Muck 2006). Lima *et al*. (2010), Reich and Kung (2010) reported that the acid LAB is a useful compound as it stimulates ~~a~~ decrease in pH during ensiling, inhibits the growth of fungi or yeasts that can cause aerobic deterioration, and protects roughage.

*Author for correspondence: <ibrahim.ertkn@hotmail.com>.

Although cereals can be ensiled more easily than legume forage crops (Kung *et al.* 2018), the digestible protein content of cereals is lower than that of legume crops. In addition, ensiling legumes together with cereals improves the ability of these plants to be stored as silage (Arslan-Duru *et al.* 2021). For these reasons, cereals + legume mixed silages have been the subject of scientific studies intensively in recent years.

In the present study, the ensiling potential of wheat and some annual legume forage crops, which have the potential to produce roughage in developing countries such as Turkey, especially during the winter season, with various lactic acid bacteria inoculants at different mixing ratios was investigated.

Materials and Methods

In the present study, grass pea, forage pea and common vetch plant species, which are in the cool climate forage crops class, and bread wheat were used as plant material. These plant species were sown in separate plots (20 m^2) at the Hatay Mustafa Kemal University Faculty of Agriculture, Field 49 Research Area, with 20 cm spacing among rows and 450 plants m^{-1} density. The field soil has a clay loam structure and its organic carbon content is quite low. When the wheat plant reached the dough stage about 180 days after plant emergence, the plants in all plots were harvested and chopped with 2-3 cm via plant shredder for silage production. Samples of 500 g were taken from the chopped plants and the nutritive value of the silage and the epiphytic microorganism contents were examined in these samples. The chopped plants were ensilaged in pure (pure wheat, pure grass pea, pure forage pea and pure common vetch) and mixed form (25:75, 50:50, 75:25) with 5 replicates. At the ensiling stage, lactic acid bacteria inoculants of Pioneer 1188 (*Lactobacillus plantarum* and *Enterococcus faecium*), Pioneer 11A44 (*Lactobacillus buchneri*) and Pioneer 11CFT (*Lactobacillus buchneri* and *Lactobacillus casei*) were inoculated (10^5 cfu g^{-1} fresh matter) into silage materials. Fresh plants were ensilaged to vacuum bags as a laboratory type mini silo in the amount of 400 ± 50 g via an industrial type vacuum device. All silages were fermented at room conditions for 120 days and opened at the end of this period.

Silage fresh materials and fresh samples separated from silages were dried in an oven with hot air blowing at 105^{o}C for 72 hrs and dry matter ratios were determined by making use of the weight difference. In addition, neutral detergent fiber (NDF), acid detergent fiber (ADF), crude protein and crude ash contents of silages and fresh plant materials were analyzed according to AOAC (2019). Analysis of water-soluble carbohydrates was done according to the method reported by Dubois *et al.* (1956). Silage pH was determined with a table-type pH meter. The ammonia nitrogen contents of the silages were analyzed according to the Kjeldahl distillation and titration procedures. Lactic acid, acetic acid, propionic acid and butyric acid contents of silages were determined using the RID detector in the HPLC device according to the method reported by de Quiros *et al.* (2009). According to Yan *et al.* (2019), 20 g of fresh sample from each treatment was homogeneously blended in 180 ml of sterile Ringer's solution for 60 sec with the help of a blender. Then the obtained samples were filtered with Whatman no 54 filter paper. In the dilution series (from 10^{-1} to 10^{-10}) made from these samples. Lactic acid bacteria (LAB), *enterobacteria* and yeast and mold populations were determined using the MRS (DE MAN, RAGOSA, SHARPE) Lactobacillus agar, VRB-G (Violed Red Bile Glucose) and Malt Extract Agar (MEA), respectively. For this purpose, microorganisms isolated from fresh samples according to a certain procedure were inoculated to agar media sterilized in an autoclave and kept in a water bath under a sterile cabinet on agar media which were kept in a water bath. Pouring of the agar media and inoculating of the microorganism were made into disposable sterile plastic Petri dishes. MRS and MEA media prepared to determine the LAB and yeast and mold numbers, respectively, were

incubated in anaerobic conditions at 37°C for 48 hrs in a climate cabinet. In addition, samples containing VRB-G prepared to determine the number of *enterobacteria* were incubated at 33°C for 18 hrs. A maximum of 300 colonies were counted in each Petri dish. The relative feed values of the silages were calculated according to the formulas given below.

$$DDM\% = 88.9 - (0.779 \times ADF\%) \quad [1]$$

$$DMI\% = 120/NDF\% \quad [2]$$

where DDM is % of dry matter, DMI is % of animal body weight

$$RFV = DDM \times DMI \times 0.775 \quad [3]$$

The present study was conducted in a factorial (3 legume species × 3 inoculants × 5 mixtures for silages and 4 plant species × 3 inoculants × 5 mixtures for silage beginning materials) arrangement design. Variance analysis (ANOVA) for all values obtained from the present study were made according to factorial arrangement model. Significance among the means was evaluated by using the TUKEY pairwise test ($P < 0.05$). It was given main effect (legume species, inoculants and mixtures) results.

Results and Discussion

Effects of plant species on silage beginning dry matter, chemical compositions and epiphytic flora contents except for yeast and mold number were significant. The highest dry matter, NDF, ADF, water soluble carbohydrate and lactic acid bacteria were determined in wheat. Although the highest crude protein was in grass pea, other legume species were in the same statistical group. Crude protein, crude ash and *enterobacteria* of wheat was lower than legume species while NDF, ADF and water soluble carbohydrate contents of legume species were higher than wheat. The highest crude ash was in vetch species.

In the present study, the initial contents of wheat, fodder pea, grass pea and vetch species used as silage material were generally different from each other (Table 1). Similarly, it has been reported in many studies that legumes have higher crude protein and crude ash contents and lower NDF, ADF and water soluble carbohydrate contents compared to cereals (Kung *et al.* 2018, Turan 2020, Arslan-Duru *et al.* 2021, Xu *et al*. 2022). In parallel with the results of the lactic acid bacteria count obtained from the present study. Kung and Shaver (2001) reported that the epiphytic flora of cereals can contain higher lactic acid bacteria compared to legumes.

Effects of legume species on dry matter, crude protein, NDF and relative feed value were insignificant while the effects on other parameters were significant (Table 2). The highest ADF and crude ash were in vetch silages but the lowest were in grass pea silages. The lowest silage pH was determined in grass pea silage. Similarly, the lactic acid bacteria, lactic acid and acetic acid contents were in grass pea silages. In addition, *enterobacteria*, yeast and mold, propionic acid, butyric acid and ammonia nitrogen of grass pea silages were lower than silages of vetch and forage pea. Forage pea and vetch silages gave similar results for properties of *enterobacteria*, yeast and mold, propionic acid and butyric acid. The highest ammonia nitrogen was obtained from forage pea silage.

Although there were no generally significant differences among the nutritive value parameters in legume silages, significant differences occurred among the fermentation and microbial population parameters (Table 2). Xu *et al.* (2022) reported that there were significant differences among legume silages in terms of fermentation quality and microbial population. On the other hand, the fermentation end product data obtained from the current study were similar to the data

reported by Kung and Shaver (2001). Dewhurst *et al.* (2003) found that there were no differences between the fermentation end products of some legume species, unlike the results of this study. However, Hymes-Fecht *et al.* (2013) reported that there were significant differences between the fermentation end products of some legume species.

Table 1. Chemical compositions and epiphytic flora contents of silage initial materials.

Variables	Plant species				P values
	Forage pea	Grass pea	Vetch	Wheat	
Dry matter g kg^{-1} FM	257.2±1.2 ^{c}	268.5±1.4 ^{b}	254.6±1.3 ^{c}	329.5±1.2 ^{a}	<.0001
Crude protein g kg^{-1} DM	205.2±2.2 ^{a}	200.7±8.3 ^{a}	199.5±4.4 ^{a}	108.4±3.0 ^{b}	<.0001
NDF g kg^{-1} DM	415.8±6.4 ^{b}	420.6±5.7 ^{b}	448.6±12.5 ^{b}	587.0±5.4 ^{a}	<.0001
ADF g kg^{-1} DM	211.6±6.1 ^{b}	215.0±7.8 ^{b}	232.5±6.4 ^{b}	324.7±5.6 ^{a}	<.0001
Water soluble carbohydrate g kg^{-1} DM	74.4±1.9 ^{b}	85.7±2.4 ^{b}	76.4±2.6 ^{b}	122.5±3.8 ^{a}	<.0001
Crude ash g kg^{-1} DM	124.1±5.8 ^{a}	102.1±3.1 ^{b}	134.5±1.9 ^{a}	96.2±4.5 ^{b}	0.0005
Lactic acid bacteria log_{10}cfu g^{-1} DM	3.8±0.1 ^{b}	4.4±0.1 ^{b}	4.0±0.1 ^{b}	5.4±0.2 ^{a}	0.0002
Enterobacteria log_{10}cfu g^{-1} DM	6.4±0.2 ^{a}	5.8±0.1 ^{a}	6.1±0.1 ^{a}	4.9±0.2 ^{b}	0.0015
Yeast and mold log_{10}cfu g^{-1} DM	4.3±0.2	4.0±0.1	4.4±0.2	3.7±0.1	ns

Data shown with different superscript letters on the same line are different from each other. FM: fresh matter, DM: dry matter.

Table 2. Effects of legume species on dry matter, chemical compositions, fermentation characteristics and microbial populations of silages.

Variables	Legume species			P values
	Forage pea	Grass pea	Vetch	
Dry matter g kg^{-1} FM	294.8±3.7	302.1±5.1	293.1±3.5	0.0717
Crude protein g kg^{-1} DM	150.2±4.3	148.9±4.1	151.2±5.7	0.8541
NDF g kg^{-1} DM	506.1±7.9	508.7±7.6	513.3±10.1	0.5610
ADF g kg^{-1} DM	275.0±5.2 ^{b}	273.5±5.3 ^{c}	282.9±4.4 ^{a}	<.0001
Crude ash g kg^{-1} DM	111.9±1.3 ^{b}	100.95±0.3 ^{c}	116.7±1.8 ^{a}	<.0001
Relative feed value	126.6±2.8	126.0±2.7	136.9±16.0	0.6477
pH	4.35±0.04 ^{a}	4.26±0.04 ^{c}	4.34±0.04 ^{b}	<.0001
Lactic acid bacteria log_{10}cfu g^{-1} DM	5.9±0.1 ^{b}	6.0±0.1 ^{a}	5.9±0.1 ^{b}	<.0001
Enterobacteria log10cfu g^{-1} DM	2.6±0.1 ^{a}	2.5±0.0 ^{b}	2.6±0.1 ^{a}	<.0001
Yeast and mold log10cfu g^{-1} DM	2.0±0.1 ^{a}	1.8±0.1 ^{b}	2.0±0.1 ^{a}	<.0001
Lactic acid g kg^{-1} DM	25.3±0.8 ^{b}	27.4±0.7 ^{a}	25.4±0.8 ^{b}	<.0001
Acetic acid g kg^{-1} DM	16.6±0.8 ^{b}	17.5±0.9 ^{a}	15.8±0.8 ^{c}	<.0001
Propionic acid g kg^{-1} DM	6.1±0.1 ^{a}	6.0±0.1 ^{b}	6.2±0.1 ^{a}	<.0001
Butyric acid g kg^{-1} DM	3.7±0.0 ^{a}	3.6±0.0 ^{b}	3.7±0.0 ^{a}	<.0001
Ammonia nitrogen g kg^{-1} TN	91.4±1.2 ^{a}	88.9±1.0 ^{c}	90.5±1.1 ^{b}	<.0001

Data shown with different superscript letters on the same line are different from each other. FM: fresh matter, DM: dry matter.

Effects of inoculant on dry matter, crude protein, NDF, ADF and relative feed value were insignificant while the effects on crude protein, silage fermentation features and microbial populations were significant (Table 3). Crude ash content of inoculant treatments was higher than control treatment. Using inoculants caused a lower pH level than control. Similarly, higher lactic acid bacteria population was determined in silages treated inoculant. On the contrary, *enterobacteria* and yeast and mold populations of inoculant treatments were lower than control. Inoculant treatments improved silage lactic acid and acetic acid contents. The highest lactic acid (22.5 g kg^{-1} DM) was determined in inoculant 1188 but the lowest value (23.6 g kg^{-1} DM) in control treatment. The highest acetic acid (19.8 g kg^{-1} DM) was recorded in inoculant 11CFT but inoculant 11A44 gave statistically similar results. The highest propionic acid (5.6 g kg^{-1} DM) and the lowest butyric acid (3.9 g kg^{-1} DM) were determined in control treatment. Similar to the propionic acid results, the highest ammonia nitrogen (93.8 g kg^{-1} TN) was recorded in control. The lowest ammonia nitrogen was in inoculant 11CFT and inoculant 11A44 gave statistically similar results.

Table 3. Effects of inoculants on dry matter, chemical compositions, fermentation characteristics and microbial populations of silages.

Variables	Control	Inoculants			*P* values
		1188	11A44	11CFT	
Dry matter g kg^{-1} FM	298.6±3.9	291.5±7.1	298.5±3.9	297.9±3.8	0.3838
Crude protein g kg^{-1} DM	146.6±4.8	148.6±4.7	155.8±7.3	149.3±4.7	0.2386
NDF g kg^{-1} DM	512.5±8.7	512.0±8.6	501.0±13.2	512.0±8.7	0.3852
ADF g kg^{-1} DM	277.4±5.8	277.4±5.7	276.5±5.8	277.2±5.8	0.0606
Crude ash g kg^{-1} DM	109.2±1.8 ^{b}	109.9±1.8 ^{a}	110.2±1.8 ^{a}	109.9±1.8 ^{a}	<.0001
Relative feed value	124.4±3.0	124.5±3.0	146.0±21.2	124.5±3.0	0.3897
pH	4.49±0.05 ^{a}	4.20±0.04 ^{d}	4.28±0.04 ^{c}	4.29±0.04 ^{b}	<.0001
Lactic acid bacteria log_{10}cfu g^{-1} DM	5.2±0.0 ^{b}	6.1±0.1 ^{a}	6.1±0.1 ^{a}	6.2±0.1 ^{a}	<.0001
Enterobacteria log10cfu g^{-1} DM	2.9±0.0 ^{a}	2.5±0.0 ^{b}	2.4±0.1 ^{c}	2.4±0.1 ^{c}	<.0001
Yeast and mold log10cfu g^{-1} DM	2.3±0.1 ^{a}	2.0±0.1 ^{b}	1.8±0.1 ^{c}	1.7±0.1 ^{d}	<.0001
Lactic acid g kg^{-1} DM	23.6±0.8 ^{c}	27.5±0.9 ^{a}	26.5±0.9 ^{b}	26.6±0.9 ^{b}	<.0001
Acetic acid g kg^{-1} DM	9.8±0.3 ^{c}	17.4±0.8 ^{b}	19.6±0.9 ^{a}	19.8±0.8 ^{a}	<.0001
Propionic acid g kg^{-1} DM	5.6±0.1 ^{d}	6.0±0.1 ^{c}	6.4±0.0 ^{b}	6.5±0.0 ^{a}	<.0001
Butyric acid g kg^{-1} DM	3.9±0.0 ^{a}	3.6±0.0 ^{b}	3.5±0.0 ^{c}	3.5±0.0 ^{c}	<.0001
Ammonia nitrogen g kg^{-1} TN	93.8±1.2 ^{a}	89.7±1.1 ^{b}	88.8±1.2 ^{bc}	88.7±1.3 ^{c}	<.0001

Data shown with different superscript letters on the same line are different from each other. FM: fresh matter, DM: dry matter. Control: no-inoculated, 1188: Pioneer 1188, 11A44: Pioneer 11A44, 11CFT: Pioneer 11CFT.

Except for the crude ash content of the silages, other nutritive value parameters were not affected by the inoculant treatments (Table 3). Microbial populations and fermentation properties of silages were changed with inoculant applications (Table 3). Contreras-Govea and Muck (2006) reported that homofermentative and heterofermentative lactic acid bacterial inoculants cause a rapid pH decrease in silages and this situation increases especially lactic acid and acetic acid contents. It has been reported that *Lactobacillus buchneri* bacteria, which is heterofermentative and found in 11A44 and 11CFT inoculants, produces higher acetic acid than lactic acid bacteria species with homofermentative properties (Kung *et al.* 2018). Indeed, 11A44 and 11CFT

inoculants reduced protein loss in the silo (Table 3). The strong antifungal capacity of the acetic acid produced in the silo (Contreras-Govea and Muck 2006) may have revealed this situation.

Effects of mixture ratios on dry matter, chemical compositions, relative feed value, fermentation parameters of silages were significant (Table 4). As the wheat proportion in the silages increased, dry matter improved. The highest dry matter (325.3 g kg^{-1} FM) was determined in pure wheat (100 + 0) silage. Crude protein content of silages decreased as the wheat proportion in silages increased. The highest crude protein (190.3 g kg^{-1} DM) was recorded in pure legume (0 + 100) silage. As the legume proportion in the mixtures increased, NDF and ADF contents decreased. On the contrary, crude ash content increased. The highest relative feed value (153.9) was in pure legume (0 + 100) silage while the lowest value was recorded in pure wheat (100 + 0) silage. Silage pH level decreased as the wheat proportion in the mixtures increased. The mixture obtained as a result of mixing legumes with wheat had a lower pH level in silages than pure legume silage (0 + 100). The highest lactic acid bacteria (6.6 log_{10}cfu g^{-1} DM), the lowest *enterobacteria* (2.1 log_{10}cfu g^{-1} DM) and yeast and mold (1.4 log_{10}cfu g^{-1} DM) were determined in pure wheat (100 + 0) silages. Lactic acid and acetic acid contents of the silages improved as the legume proportion in the mixtures decreased. The highest lactic acid (34.7 g kg^{-1} DM) and acetic acid (22.4 g kg^{-1} DM) were recorded in pure wheat (100 + 0) silages. On the other hand, propionic and butyric acid of the silages decreased as the wheat proportion in the mixtures increased. The highest ammonia nitrogen (98.7 g kg^{-1} DM) was determined in pure legume (0 + 100) silage while the lowest value (76.8 g kg^{-1} DM) was in pure wheat (100 + 0) silage.

Table 4. Effects of mixture ratios on dry matter, chemical compositions, fermentation characteristics and microbial populations of silages.

Variables	Mixture ratios (Wheat + Legumes)					*P* values
	0+100	25+75	50+50	75+25	100+0	
Dry matter g kg^{-1} FM	262.3±2.2 ^{d}	281.9±1.4 ^{c}	299.7±1.6 ^{b}	314.0±0.6 ^{ab}	325.3±8.2 ^{a}	<.0001
Crude protein g kg^{-1} DM	190.3±1.1 ^{a}	172.1±0.2 ^{b}	149.9±0.2 ^{c}	126.7±0.4 ^{d}	111.4±8.4 ^{e}	<.0001
NDF g kg^{-1} DM	432.8±2.2 ^{e}	471.6±1.8 ^{d}	511.8±1.2 ^{c}	537.5±13.7 ^{b}	593.1±0.5 ^{a}	<.0001
ADF g kg^{-1} DM	223.3±1.6 ^{e}	251.6±1.1 ^{d}	276.3±0.7 ^{c}	302.7±0.4 ^{b}	331.8±0.3 ^{a}	<.0001
Crude ash g kg^{-1} DM	121.9±2.3 ^{a}	116.1±1.7 ^{b}	109.6±1.1 ^{c}	103.3±0.5 ^{d}	98.2±0.2 ^{e}	<.0001
Relative feed value	153.9±1.0 ^{a}	136.8±0.7 ^{ab}	122.5±0.4 ^{ab}	136.9±26.7 ^{ab}	98.9±0.1 ^{b}	0.0226
pH	4.67±0.03 ^{a}	4.56±0.03 ^{b}	4.37±0.02 ^{c}	4.06±0.02 ^{d}	3.91±0.01 ^{e}	<.0001
Lactic acid bacteria log_{10}cfu g^{-1} DM	5.3±0.1 ^{e}	5.6±0.1 ^{d}	5.9±0.1 ^{c}	6.2±0.1 ^{b}	6.6±0.1 ^{a}	<.0001
Enterobacteria log10cfu g^{-1} DM	2.9±0.0 ^{a}	2.8±0.0 ^{b}	2.6±0.0 ^{c}	2.4±0.0 ^{d}	2.1±0.1 ^{e}	<.0001
Yeast and mold log10cfu g^{-1} DM	2.4±0.0 ^{a}	2.3±0.0 ^{b}	2.0±0.0 ^{c}	1.7±0.0 ^{d}	1.4±0.0 ^{e}	<.0001
Lactic acid g kg^{-1} DM	18.7±0.4 ^{e}	22.3±0.3 ^{d}	25.5±0.3 ^{c}	29.1±0.6 ^{b}	34.7±0.4 ^{a}	<.0001
Acetic acid g kg^{-1} DM	11.3±0.3 ^{e}	13.5±0.5 ^{d}	16.8±0.9 ^{c}	19.3±1.0 ^{b}	22.4±1.2 ^{a}	<.0001
Propionic acid g kg^{-1} DM	6.5±0.1 ^{a}	6.1±0.0 ^{b}	6.1±0.0 ^{b}	6.0±0.1 ^{b}	5.8±0.1 ^{c}	<.0001
Butyric acid g kg^{-1} DM	3.9±0.0 ^{a}	3.9±0.0 ^{a}	3.7±0.0 ^{b}	3.5±0.0 ^{c}	3.2±0.0 ^{d}	<.0001
Ammonia nitrogen g kg^{-1} TN	98.7±0.6 ^{a}	96.0±0.4 ^{b}	92.6±0.5 ^{c}	87.1±0.5 ^{d}	76.8±0.6 ^{e}	<.0001

Data shown with different superscript letters on the same line are different from each other. FM: fresh matter, DM: dry matter.

Dry matter, chemical composition, microbial population and fermentation characteristics of silages were affected by mixing ratios (Table 4). Geren *et al.* (2008) found that crude protein and pH level of mixed silages (corn with cowpea and bean) were higher than pure corn silages.

Similarly, Contreras-Govea *et al.* (2009) determined that crude protein content improved and NDF and ADF contents decreased in mixed silages of maize and some legumes. In addition, it was highlighted that the pH level increased slightly with the addition of legume species in corn silage. Ozturk *et al.* (2006) and Contreras-Govea *et al.* (2011) found that pH levels of cereal + legume mixed silages were lower than those of pure legume silages. Also, it was reported that better fermentation was obtained from mixed silages compared to pure legume silages and the nutritive value of mixed silages was higher than pure cereal silages. Results of the scientific studies conducted with the legume and cereal mixed silages summarized above are similar to the results of the present study.

Results of current study indicated that increasing wheat proportion of legume + wheat mixed silages positively affected the chemical composition of mixed silages. In addition, a better nutritive value than pure wheat silages were obtained from legume + wheat mixed silages. Particularly the significant increase in protein content of mixed silages was considerable. On the other hand, fermentation parameters of mixed silages were greater than pure legume silages. Compared to control treatment, using the inoculant in pure legume and wheat silages and their mixture silages improved silages fermentation quality. Inoculant 1188 gave better fermentation quality than inoculants 11A44 and 11CFT. In addition, mixed silages of wheat and grass pea gave better results than vetch and fodder pea. From the above results may be said that ensiling the 50% wheat with 50% grass pea with the use of inoculant 1188 as an additive could give a good result both in terms of nutritive value and fermentation quality.

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