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Yield and silage quality of soybean-maize intercrop under different mixing ratios and harvest stages

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Abstract

The research was carried out to determine the effects of mixing ratios and harvest stages on dry matter yield and silage quality of intercropped soybean (*Glycine max*) and maize (*Zea mays*). A split plot design with four replications was conducted in Eastern Mediterranean Agricultural Research Institute Experiment Area, Adana-Turkey. Five treatments (sole maize (M) and sole soybean (S), 100:100 MS; 50:100 Ms, 100:50 Ms %) and two harvest stages (milk stage and dough stage) were evaluated in 2011 growing second crop season. As a result of the research, dry matter (DM) yield, crude protein ratio (CPR), pH, neutral detergent fiber (NDF), acid detergent fiber (ADF), ash, dry matter intake (DMI), relative feed value (RFV), and digestible dry matter ratio (DDMR) ranged from 7.6 to 19.6 t ha^{-1}; 56.0 to 49.7 g kg^{-1}; 3.81 to 4.95; 411.8 to 539.6 g kg^{-1}; 272.8 to 366.7 g kg^{-1}; 52.0 to 111.9 g kg^{-1}; 22.4 to 30.0 g kg^{-1}; 109.8 to 145.8; 603.3 to 676.5 g kg^{-1}, respectively. Hence, intercrop of maize (100%) + soybean (100%) at dough stage can be suggested for high dry matter yield and forage quality in a crop rotation following winter wheat.

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Introduction

Maize (*Zea mays*), which is the third most important cereal crop of world, is an important dual purpose crop used in human diet and animal feed. Maize silage has a relatively high dry matter content, low buffering capacity and adequate water soluble carbohydrates for fermentation to lactic acid and is regarded as an ideal crop for silage. Also, maize silage is the main silage fed to dairy cattle in Turkey and plays a vital role in supplying large amounts of digestible fiber and energy-rich forage for animal diets. In spite of its high energy content, its protein content is low (88 g kg^{-1}) (NRC, 2001) compared to legumes silage (Anil *et al*., 2000) and needs supplementation with protein for better feed quality (Stoltz *et al*., 2013). It is well documented that legumes have higher buffering capacities compared to grasses, which extends duration of fermentation, slows drops in pH and increases proteolysis (Albrecht and Muck, 1991; McDonald *et al*., 1991). Moreover, it is also well documented that all legume types does not ferment equally when planted in monoculture (Owens *et al*., 1999; Albrecht and Beauchemin, 2003; Mustafa and Seguin, 2003) or in a mixture (Dawo *et al*., 2007).

Physiological and morphological differences between intercrop components affect their ability to use resources; especially cereals with legumes, have several advantages such as higher total yield, better land use efficiency (Dhima *et al*., 2007), yield stability of the cropping system (Lithourgidis *et al*., 2006), better utilization of light, water, and nutrients (Javanmard *et al*., 2009), improved soil conservation (Anil *et al*., 1998), soil fertility through biological nitrogen fixation, increases soil conservation through greater ground cover compared to sole cropping, and provides better lodging resistance for crops that are susceptible to lodging when grown in monoculture (Lithourgidis *et al*., 2006) and better control of pests and weeds (Banik *et al*., 2006; Vasilakoglou *et al*., 2008).

Atmospheric nitrogen fixation by legumes can reduce the competition for nitrogen in the legume-cereal intercropping system, allowing the cereals to use more soil nitrogen (Eskandari *et al*., 2009). This can affect forage quality of intercrop components because protein content is directly related to the nitrogen content of the forage (Putnam *et al*., 1985).

The hypothesis of present study it would provide valuable information about the contribution of intercropping maize with soybean for better silage; (i) the making of silage under Eastern Mediterranean field condition during second crops season (after harvest wheat) with both crops simultaneously sown and harvested; (ii) to determine the effect of two different mixture and harvest stage on forage yield and silage quality by increasing protein contents.

Material and methods

Experiment site

The field experiment was established in the experiment area of Eastern Mediterranean Agricultural Research Institute in Adana/Turkey (36°51´ 18" latitude N, 35°20´ 49" longitude E, and 15 m above sea level). The experiment was carried out during 2011 second crop growing season after wheat harvest. The climate of the region is typically Mediterranean, with a mild rainy winter and a dry, high relative humidity and hot summer. According to the long-term average from four decades of records, there is early total precipitation of 625 mm and mean temperature of 18.7 °C. According to climatic data of growing season (from June to September), mean temperature and relative humidity were 27.03 °C, and 69.5 %, respectively. There is not any precipitation.

Before sowing, soil samples were collected for chemical and physical analysis. The soil of the research area has salinity 0.026 %, pH 7.72, calcium carbonate ($CaCO_3$) 20 %, organic matter 2 %, loam 27.8 %, clay % 31.2 % and silt 41.0 %. The soil of the research area is clay-loam-silt in texture, non-saline and rich in calcium carbonate and slightly alkaline in pH (Source: University of Cukurova, Soil Science Dept. Lab, 2011).

Experimental design, description of cropping system and crop management

The treatments were evaluated in a split plot design with five levels of mixing ratios 100:100 (M:S), 50:100 (M:S), 100:50 (M:S), 0:100 (S) and 100:0 % (M) in main plots and at two levels of maturity stages of maize (milk stage and dough stage) in sub plots with four replications.

The descriptions for treatments in the experiment were summarized in Table 2. Yesilsoy soybean (*Glycine max*) variety and Pioneer P31Y43 maize (*Zea mays)* varieties were used as the material.

Individual plot size was 2.8 m × 5 m. The seeds of maize and soybean varieties were mixed and sown by hand in 2011. Sole crop densities were 28 and 9 plants m^{-2} for soybean and maize, respectively. Inner-row spacing were sole soybean and maize as well as M:S, m: S, M:s, 5, 15, 3-4, 6, and 4-4.5 cm, respectively with an inter-row spacing of 70 cm.

Soybean and maize seeds were mixed before sowing than sown on the same rows and determined inner row space for intercropping applications. Descriptions of experimental treatments were given in Table 1. All plots were fertilized with the same amount of fertilizer before sowing; containing 70 kg P_2O_5 ha^{-1} and 70 kg N ha^{-1} in the form of compose (20:20:0). The rest of the nitrogen (70kg N ha$^{-1)}$, in the form of urea was applied at stem elongation stage of maize. Thinning and weed control were applied uniformly and five times irrigation was applied for all the treatments during growth period.

Table 1. Description of experimental treatments.

Factor A	Description	Plant Density (plants ha^{-1})	Botanical Composition (%) Maize-Soybean
M	Sole maize (100 % maize + 0 % soybean)	95.240	100 – 0*
S	Sole soybean (100 % soybean + 0 % maize)	285.710	0 - 100
M:S	Intercrop of maize (100 %) + soybean (100 %)	95.240 +285.710	87.4 - 12.6
M:s	Intercrop of maize (100 %) + soybean (50 %)	95.240 +142.850	86.0 - 14.0
m:S	Intercrop of maize (50 %) + soybean (100 %)	47.620 +285.710	92.9 - 7.1
Factor B			
H1	Harvest at milk stage		
H2	Harvest at doughy stage		

(*) each treatment was silaje based on botanical composition.

Plant sampling and silage preparation

Harvested sole and mixture materials were sub sampled, separated into each species by hand and weighted based on botanical composition to calculate mixture ratios at harvest time. According to table data, each treatment was silage based on botanical composition. Besides, a random sample of 500 g fresh forage was collected from each plot for each species, weighed, and dried for 48 h in Owen at 70 ºC for calculating the dry matter yield.

Secondary sub samples from the harvested materials were taken and used for silage preparation. For this aim, 3 kg of fresh mixture samples from each plot were taken, chopped mechanically with dimensions 1-3 cm. The samples without additives were pressed by a special apparatus (Petterson, 1988) into plastic bottles of 3 liters capacity for each treatment. The bottles were tightly sealed and kept and ensiled for 60 days at ambient temperatures (17-25 ºC). Five hundred gram fresh sub sample of ensiled was taken for dry matter and initial characterization of corn-soybean mixture. Silos were frozen to -20 ºC to stop fermentation and remained frozen until silages were analyzed.

Chemical analyses

Fresh and ensiled forage were analyzed for pH by placing a 20 g sample in a blender jar, diluting with deionizer distilled water to 200 g, and blending for 30 s in a high-speed blender (Chen *et al.*, 1994). The diluted sample was filtered through four layers of cheesecloth and pH was measured immediately with a pH meter. A 20 ml aliquot was centrifuged at 25.000 xg for 20 min at 4 ºC, and the supernatant was decanted into 20 mL scintillation vials and stored at 20 ºC for later analysis of fermentation products. Organic acids (lactic, acetic, propionic, butyric and ethanol) concentration was measured by HPLC as reported by Erten (1998) and Cordieretal (2007).

At the time of opening, 500 g silage sample was taken and dried at 60-65 ºC for 48 h and ground in a mill and passed through a 1mm screen for forage quality analysis. Ground samples were analyzed for Kjeldahl N (Association of Official Analalytical Chemists, 1984).

Crude protein (CP) was calculated as Kjeldahl N x 6.25. The content of Acid detergent fiber (ADF), neutral detergent fiber (NDF), and ash content were determined according to the methods of Van Soest (1982) with Ankom fiber analyzer (Ankom Technology Corporation, Fairport, NY). Digestibility of dry matter (DDM) ratio, Dry Matter Intake (DMI), and Relative Feed Value (RFV) were determined by method of Jaranyama and Garcia (2004) and used formulas: DDM (Digestible Dry Matter)= 88.9 - (0.779 x ADF %), DMI = Dry Matter Intake (% of BW) = 120/(NDF %), RFV = (DDM x DMI)/1.29.

Statistical analyses

The data on growth, yield, and quality parameters were analyzed by JMP statistical analysis package with variance technique and SEM test probability level, in a split plot experiment with four replicates (Yurtsever, 1984).

Results and discussion

The results of DMY, CP, NDF, ADF, Ash content, DMI and RFV values presented in Table 2.

Table 2. Yield and some silage quality parameters (CP, NDF, ADF, Ash, DMI and RFV) in different cropping systems and harvest stages.

	Treatments	DM Yield (t ha^{-1})	CP (g kg^{-1})	NDF (g kg^{-1})	ADF (g kg^{-1})	Ash (g kg^{-1})	DMI (g kg^{-1})	RFV
Milk Stage	S	7.6	149.7	411.8	366.7	111.9	29.1	136.3
	M	13.6	56.0	489.1	313.0	67.9	24.6	123.2
	M:S	16.4	65.6	525.2	347.7	64.8	22.9	109.8
	m:S	17.8	70.6	507.6	350.6	78.0	23.7	113.0
	M:s	16.5	60.0	539.6	331.6	64.1	22.4	110.0
Dough Stage	S	11.5	142.3	504.4	345.0	89.0	30.0	144.5
	M	17.5	61.5	452.1	272.8	52.0	26.6	139.6
	M:S	19.6	72.0	428.5	290.0	65.4	28.3	145.8
	m:S	19.5	71.0	480.7	311.3	58.7	25.1	126.0
	M:s	16.7	65.1	468.4	298.0	65.6	25.6	130.6
Mean of treatments	S	9.5 c	147.0 a	408.1 b	352.8 a	99.9 a	29.5 a	140.4 a
	M	15.6 b	58.8 d	470.6 a	291.9 c	60.0 b	25.6 b	131.4 ab
	M:S	18.0 a	68.8 bc	482.8 a	318.8 bc	65.1 b	25.6 b	127.8 ab
	m:S	18.6 a	70.8 b	494.2 a	331.0 ab	68.4 b	24.4 b	119.5 b
	M:s	16.6 ab	62.6 cd	504.0 a	314.8 bc	65.3 b	24.0 b	120.3 b
Harvest Stage	H1	14.4 B	80.4	494.7 A	341.5 A	77.4 A	24.5 B	115.0 B
	H2	17.0 A	82.8	446.8 B	303.4 B	66.1 B	27.1 A	137.3 A
P-value	HS	<0.01	NS	<0.01	<0.01	<0.01	<0.01	<0.01
	Treatments	<0.01	<0.01	<0.01	<0.05	<0.01	<0.01	<0.05
	HSxT Int	NS	NS	NS	NS	NS	NS	0.38
SEM	HS	0.01	1.37	8.02	5.86	2.63	0.50	3.17
	Treatments	0.68	2.17	13.0	9.22	4.15	0.78	5.00
	HSxT Int	0.98	3.07	18.40	13.04	5.87	1.10	7.08

*Dry Matter (DM); Crude Protein (CP); Neutral Detergent Fiber (NDF); Acid Detergent Fiber (ADF); Dry Matter Intake (DMI), Relative Feed Value (RFV).

** Harvest at milk stage (H1); Harvest at doughy stage (H2); Harvest x Treatment Interaction (HS x TInt); Non Significant (NS); Standard Error of the Mean (SEM)

***Means in the same row with different letters differ significantly (P<0.05).

Dry Matter Yield (DMY)

Analysis of variance showed that harvest stage and treatments had a significant effect on DM yield at the levels of 1%, whereas no significantly differences in interaction between harvest stage and treatment were found. The value of DM yield at dough stage (17.0 t ha^{-1}) was higher than that at milk stage (14.4 t ha^{-1}). Harvesting at dough stage has longer vegetative time than milk stage harvest so DM accumulation increased from milk stage to dough stage. The DM yield varied from 9.5 t ha^{-1} to 8.6 t ha^{-1}. Intercrop of maize (100 %) + soybean (100 %) and intercrop of maize (50 %) + soybean (100 %) treatments were ranked the first group and followed by intercrop of maize (100 %) + soybean (50 %).

Similar results have been reported by Dahmardeh *et al.* (2009). On the other hand, the lowest DM yield value was obtained from sole soybean. It was reported that maize-soybean intercrops produced higher DM yield than either species sole (Tansı and Sağlamtimur, 1992; Geren *et al.*, 2008; Eskandari, 2012; Eslamizadeh, 2015). One possible explanation for the higher yields of intercrops is the ability of the crops to exploit different soil layers without competing with each other. Besides, higher consumption of environmental resources, agronomic practices, crop genotypes, photosynthetic ally active radiation and soil moisture by intercropping raining period can affect the yield and potential use of the intercropping system (Ofori and Stern, 1987; Anil *et al.*, 1998; Lithourgidis *et al.*, 2006). Geren *et al.*, (2008); Htet *et al.*, (2016) indicated that, legume contribution to corn in mixtures was significant and increased the total biomass yield of mixtures. Our findings are in accordance with these researches.

Crude Protein Ratio (CPR)

As seen in Table 2, Analysis of variance showed that treatments had a significant effect on CPR at the levels of 1%, whereas no significantly differences in harvest stage and interaction between harvest stage and treatment were found. The means of treatment CPR varied between 58.8 and 147.0 g kg^{-1}. The highest CPR was obtained from sole soybean, while the lowest value was obtained from sole maize. The mean CP concentration of sole soybean was considerably greater than that of sole maize. These results showed that an increased proportion of legumes in intercrops increased the crude protein contents in mixture. Results in the present study were in agreement with other studies where legumes also increased CP concentration when in a mixture with corn (Dawo *et al.*, 2007; Dahmardeh *et al.*, 2009; Baghdadi *et al.*, 2016; Erdal *et al.*, 2016; Htet *et al.*, 2016). It could be due to higher nitrogen availability for maize in intercropping compared with the sole crop (Eskandari *et al.*, 2009; Eskandari, 2012). The results are in agreement with other studies where legumes also increased CP concentration when in mixture with corn (Dawo *et al.*, 2007; Geren *et al.*, 2008; Htet *et al.*, 2016).

Neutral Detergent Fiber (NDF)

Analysis of variance showed that harvest stage and treatments had a significant effect on NDF at the levels of 1 %, whereas no significantly differences in interaction between harvest stage and treatment were found. The value of NDF at milk stage (494.7 g kg^{-1}) was higher than that at dough stage (446.8 g kg^{-1}). Similar results have been reported by Dahmardeh *et al.* (2009) and Htet *et al.* (2016). The mean NDF declined from milk stage to dough stage of harvest (Table 2). The means of treatment NDF value varied between 408.1 and 504.0 g kg^{-1} and except for sole soybean, all treatments ranked as first. The NDF content is important in ration formulation because it reflects the amount of forage that can be consumed by animals (Lithourgidis *et al.*, 2006). As NDF percentage increases, DMI decreases (Van Soest, 1994). A decline in fiber concentration with increasing maturity can be attributed to the dilution effect created by increasing content of grain as maize get matured (Coors *et al.*, 1997). High quality forages have low concentration of both NDF and ADF and high digestibility (Peterson *et al.*, 1994). The ADF concentration values, consisting of cellulose and lignin, are important because they describe the ability of an animal to digest the forage. As the ADF increases, the digestibility of the forage usually decreases. The NDF value refers to the total cell wall and is composed of the ADF fraction plus hemicelluloses.

Acid detergent fiber (ADF)

Analysis of variance showed that harvest stage and treatments had a significant effect on ADF at the levels of 1 and 5 %, whereas no significantly differences in interaction between harvest stage and treatment were found. The value of ADF at dough stage (303.4 g kg^{-1}) was lower than that at milk stage (341.5 g kg^{-1}). The mean ADF declined from milk stage to dough stage of harvest (Table 2). Similar results have been reported by Dahmardeh *et al.* (2009) and Htet *et al.* (2016). It is well known that cell wall components such as cellulose and lignin contents are greater in stems than leaves (Aman, 1993) so the ADF concentration decreased with the delay in harvest time.

Besides, the highest ADF value was obtained from sole soybean (352.8 g kg^{-1}), but the lowest ADF value was obtained from sole maize (291.9 g kg^{-1}). The increase in the soybean ratio in the mixture increase ADF significantly. Generally, NDF concentration is greater for grasses than for legumes (NRC, 2001; Dahmardeh *et al.*, 2009). On the other hand, Armstrong *et al.*, 2008 reported that intercropping climbing beans with corn increased NDF concentration and decreased digestibility compared to monoculture corn.

In soybean, the rate of cell wall structural constituents, such as cellulose and lignin is increasing as the harvest time is delayed. Because of this reason in this research, monoculture soybean had the highest ADF value. Forage quality in terms of ADF content can be improved by intercropping as compared with sole soybean. Thus addition of cereal to soybean reduced the ADF concentrations.

Ash Content

Analysis of variance showed that harvest stage and treatments had a significant effect on ash content at the levels of 1 %, whereas no significantly differences in interaction between harvest stage and treatment were found. The value of ash content varied between 77.4 g kg^{-1} and 66.1 g kg^{-1} also, the mean ash content declined from milk stage to dough stage of harvest (Table 2). Similar results have been reported earlier by Dahmardeh *et al.* (2009) and Ht et *et al.* (2016). In terms of treatment, ash content value varied between 60.0 and 99.9 g kg^{-1}. The highest ash value was obtained from sole soybean whereas other treatments had the lowest value with no significantly differences.

Dry Matter Intake (DMI)

Analysis of variance showed that harvest stage and treatments had a significant effect on DMI at the levels of 1%, whereas no significantly differences in interaction between harvest stage and treatment were found. The value of DMI varied between 24.5 g kg^{-1} and 27.1 g kg^{-1} also, the DMI value increased from milk stage to dough stage of harvest (Table 2).

Dry matter intake (DMI) varied between 24.0 and 29.5 g kg^{-1} and the highest value were obtained from sole soybean, while there were no significantly differences among other treatments. The mean DMI concentration of sole soybean was higher than intercropping. Oba and Allen (1998) stated that, enhanced NDF digestibility of forage significantly increased DMI and milk yield of dairy cows. As NDF percentages increase, the DM intake will generally decrease (Joachim and Jung, 1997). High quality forages have low concentration of both NDF and ADF and high digestibility (Hatfield, 1993; Peterson *et al.*, 1994). Huhtanen *et al.*, 2002 showed that variation in fermentation quality affects voluntary feed intake of cattle.

Relative Feed Value (RFV)

Analysis of variance showed that harvest stage and treatments had a significant effect on RFV at the levels of 1 and 5 %, whereas no significantly differences in interaction between harvest stage and treatment were found (Table 2). The value of RFV at dough stage (137.3) was higher than that at milk stage (115.0). The RFV varied from 140.4 to 119.5. While the highest value was obtained from sole soybean, sole maize and intercrop of maize (100 %) + soybean (100 %) shared same statistical group with sole soybean. On the other hand, both intercrop of maize (100 %) + soybean (50 %) and intercrop of maize (50 %) + soybean (100 %) had statistically the lowest group. The results of DDMR and fermentation products values presented in Table 3.

Digestible Dry Matter Ratio (DDMR)

Analysis of variance showed that harvest stage and treatments had a significant effect on DDMR at the levels of 1 %, whereas no significantly differences in interaction between harvest stage and treatment were found (Table 3). The value of DDMR at dough stage (653.0 g kg^{-1}) was higher than that at milk stage (623.0 g kg^{-1}). The DDMR varied from 611.8 to 661.7 g kg^{-1}.While the highest value was obtained from sole maize, the lowest DDMR value was obtained from sole soybean. Fermentation products such as pH, lactic acid, acetic acid, propionic acid, butyric acid and ethanol given in Tables 3.

Table 3. Some quality parameters (DDMR and organic acids) in different cropping systems and harvest stages.

	Treatments	DDMR (g kg^{-1})	pH	LA (g kg^{-1})	AA (g kg^{-1})	PA (g kg^{-1})	BA (g kg^{-1})	Ethanol (g kg^{-1})
Milk Stage	S	603.3	4.95 a	37.0 a	10.7 a	0.88 c	0.85	2.80 bc
	M	646.7	3.81 f	36.7 a	5.0 de	1.19 ab	0.88	2.95 bc
	M:S	618.2	3.96 e	36.9 a	6.0 c	1.18 ab	0.88	1.76 d
	m:S	615.9	4.03 cde	33.6 ab	5.4 cd	1.09 ab	0.84	2.48 cd
	M:s	630.7	3.95 e	37.9 a	5.9 c	1.06 b	0.85	3.91 a
Dough Stage	S	620.3	4.85 b	20.8 d	7.7 b	1.21 a	0.87	3.19 abc
	M	676.5	3.98 de	28.9 bc	4.0 f	1.13 ab	0.90	3.50 ab
	M:S	663.1	4.07 cd	30.1 bc	3.1 g	1.17 ab	0.88	2.04 d
	m:S	646.5	4.09 c	36.3 a	6.2 c	1.20 a	0.85	3.89 a
	M:s	656.9	4.06 cd	26.6 c	4.2 ef	1.15 ab	0.89	3.25 ab
Mean of Treatments	S	611.8 c	4.90 a	28.9 b	9.18 a	1.05	0.86	3.00 b
	M	661.7 a	3.89 c	32.8 a	4.47 c	1.16	0.89	3.22 ab
	M:S	640.8 ab	4.02 b	33.5 a	4.53 c	1.18	0.88	1.90 c
	m:S	631.0 bc	4.06 b	34.9 a	5.76 b	1.15	0.85	3.18 ab
	M:s	643.8 ab	4.01 b	32.2 ab	5.06 c	1.10	0.87	3.58 a
Harvest Stage	H1	623.0 B	4.11 B	36.4 A	6.6 A	1.08 B	0.86	2.78 B
	H2	653.0 A	4.21 A	28.5 B	5.0 B	1.17 A	0.88	3.17 A
P-value	HS	<0.01	<0.01	<0.01	<0.01	<0.01	NS	<0.05
	Treatments	<0.01	<0.01	<0.05	<0.01	NS	NS	<0.01
	HS x T Int	0.74	<0.01	<0.01	<0.01	<0.01	NS	<0.05
SEM	HS	0.46	0.02	0.74	0.13	0.02	0.01	0.11
	Treatments	7.20	0.01	1.17	0.19	0.03	0.01	0.19
	HS x T Int	1.02	0.03	1.66	0.29	0.05	0.02	0.25

* Relative Feed Value (RFV), Digestible Dry Matter Ratio (DDMR), Lactic Acid (LA), Acetic Acid (AA), Propionic Acid (PA), Buthric Acid (BA)

** Harvest at milk stage (H1); Harvest at doughy stage (H2); Harvest x Treatment Interaction (HS x TInt); Non Significant (NS); Standard Error of the Mean (SEM)

** Means in the same row with different letters differ significantly (P<0.05).

pH

In terms of pH value there were significant differences between harvest stage, treatments and treatment x harvest stage interaction. The value of pH at dough stage (4.21) was higher than that at milk stage (4.11). The pH value varied from 3.89 to 4.90. While the highest value was obtained from sole soybean, the lowest pH value was obtained from sole maize. The interaction between treatment and harvest stage varied between 3.81 and 4.95. The highest pH value was obtained from sole soybean at milk and dough stages, while the lowest value was obtained from sole maize at both stages. The pH concentration averaged over all intercrop treatments was also lower than that of sole soybean (Table 3). Similar results have been reported by Erdal *et al.*, 2016. These results indicate that an increased proportion of soybean in intercrops resulted with an increased pH. Legumes have greater acetic acid concentrations than grasses; therefore, in general legume silages tend to have higher pH reasoning of higher buffering capacity caused by the organic acids (Albrecht and Beauchemin, 2003; Muck *et al.*, 2003). Silage made from the maize-soybean mixtures in our study had higher pH and contained greater lactic and acetic acid concentrations than monoculture maize. Ensiling fresh legume material is very difficult due to its high buffering capacity and low level of water soluble carbohydrate (Titterton and Maasdorp, 1997).

Lactic Acid (LA)

As seen in Table 3, In terms of LA concentration, there were significant differences between harvest stage, treatments and treatment x harvest stage interaction. The value of LA at milk stage (36.4 g kg^{-1}) was higher than that at dough stage (28.5 g kg^{-1}). The LA value varied from 34.9 g kg^{-1} to28.9 g kg^{-1}. Sole maize, intercrop of maize (100 %) + soybean (100 %) and intercrop of maize (50 %) + soybean (100 %) treatments were ranked the first group and followed by intercrop of maize (100 %) + soybean (50 %). On the other hand, the LA concentration was found to be lower in sole soybean, but increasing with mixing maize in mixtures.

Anil *et al.* (2000) and Erdal *et al.* (2016) also reported an increase in lactic acid concentration when maize was ensiled in mixture with other legumes. Lactic acid (LA) concentration was effected by treatments x harvest stage interaction and varied between 20.8 and 37.9 g kg^{-1}. In terms of LA concentration, all treatments at milk stage and intercrop of maize (50 %) + soybean (100 %) at dough stage were ranked first group, whereas sole soybean at dough stage recorded the lowest value.

Acetic acid (AA)

In terms of AA concentration, there were significant differences between harvest stage, treatments and treatment x harvest stage interaction. The value of AA at milk stage (6.6 g kg^{-1}) was higher than that at dough stage (5.0 g kg^{-1}). The AA value varied from 4.47 g kg^{-1} to 9.18 g kg^{-1}. The highest acetic acid concentration was obtained from sole soybean. Sole maize, intercrop of maize (100 %) + soybean (100 %) and Intercrop of maize (100 %) + soybean (50 %) treatments had the lowest AA concentration. Acetic acid concentration was effected by treatment x harvest stage interaction and varied between 3.1 and 10.7 g kg^{-1}. The highest acetic acid concentration was obtained from sole soybean at milk stage and the lowest value was obtained from intercrop of maize (100 %) + soybean (100 %) at the dough stage.

Propionic acid (PA)

According to Table 3, results of statistical analysis indicated that harvest stage and treatment x harvest stage interaction had significant effect on propionic acid concentration. The value of propionic acid concentration at dough stage (1.17 g kg^{-1}) was higher than that at milk stage (1.08 g kg^{-1}). The mean propionic acid concentration declined from milk stage to dough stage of harvest. Propionic acid concentration varied between 0.88 and 1.21 g $kg^{-1}$and was at the first statistical group at sole soybean and intercrop of maize (50 %) + soybean (100 %) at the dough stage. On the other hand, sole soybean application at milk stage was at the last statistical group.

Butyric acid (BA)

In terms of butyric acid concentration, there were no significant differences between harvest stage, treatments and treatment x harvest stage interaction and varied from 0.86 to 0.88; from 0.85 to 0.89; from 0.84 to 0.90 g kg^{-1}, respectively. Ethanol concentration, there were significant differences between harvest stage, treatments and treatment x harvest stage interaction.

The value of ethanol concentration at dough stage (3.17 g kg^{-1}) was higher than that at milk stage (2.78 g kg^{-1}). The ethanol concentration varied from 1.90 g kg^{-1} to 3.58 g kg^{-1}. The highest ethanol concentration was obtained from intercrop of maize (100 %) + soybean (50 %) and followed by sole maize and of 50 % maize + 100 % soybean intercrop. The lowest value was obtained from 100 % maize + 100 % soybean intercrop.

Also, ethanol concentration was effected by treatment x harvest stage interaction and varied between 1.76 and 3.91 g kg^{-1} DM^{-1}. Intercrop of maize (100 %) + soybean (50 %) at milk stage and intercrop of maize (50 %) + soybean (100 %) at dough stage ranked as first, whereas intercrop of maize (100 %) + soybean (100 %) at both milk and dough stages ranked as last.

Conclusions

This study clearly brings out the beneficial effects of maize-soybean intercropping for forage yield and quality. Maize/soybean mixture was advantageous compared to both sole crops of maize and soybean so maize-soybean intercropping increased dry matter yield and forage quality of maize.

Hence, intercrop of maize (100 %) + soybean (100 %) at dough stage can be suggested for high dry matter yield and forage quality in a crop rotation following winter wheat.

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