

## Can soybean biomass addition optimize corn silage quality?

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**ABSTRACT:** Maize silage is the main conserved roughage used in animal feed in Brazil and improving its quality has great relevance. The aim of this experiment was to evaluate the characteristics of maize silage, containing different percentages of soybean biomass. In this way, different percentages of soybean green biomass added to maize ensilage (0, 10, 20, 30 and 40% - experiment 1 and 0, 10, 20, 30, 40 and 50% in experiment 2) were evaluated. Experiments were laid out as a completely randomized. Variables were submitted to analysis of variance and when it present significance was applied regression analysis. Silage ashes increased as soybean biomass increased. Regarding to the neutral and acid detergent fiber and the amount of total digestible nutrients, there was no effect of the treatments. At experiment 1, silage crude protein increased from 7.5 to 12.6% from sole maize silage to the silage with 39.2% of soybean dry biomass, which represent an increase of 67.24%. At experiment 2, it increased from 6.77 to 12.09%, which represent 78.58% more protein at the treatment with 50% of soybean green biomass (41% dry matter of soybean) in relation to the sole maize silage. At experiment 2, for every 1% increase in soybean dry matter biomass addition, there was an increase of 0.1% of maize silage crude protein. The addition of soybean biomass to corn silage increases the ashes and crude protein content of silage.

**Key words:** ashes; crude protein; digestibility; *Glycine max*; *Zea mays*

## A adição de biomassa de soja pode otimizar a qualidade da silagem de milho?

**RESUMO:** A silagem é o principal volumoso conservado utilizado na alimentação animal no Brasil e melhorar sua qualidade, apresenta grande relevância. Objetivo deste estudo é avaliar características da silagem de milho, contendo diferentes porcentagens de biomassa de soja. Avaliaram-se diferentes frações de biomassa verde de soja, adicionadas a ensilagem de milho (0, 10, 20, 30 e 40% - experimento 1 e 0, 10, 20, 30, 40 e 50% no experimento 2). Utilizou-se delineamento inteiramente casualizado. As variáveis avaliadas foram submetidas à análise de variância e havendo efeito significativo aplicou-se análise de regressão. A matéria mineral da silagem aumentou de forma linear com a elevação das porcentagens de soja, entretanto não influenciaram fibra em detergente neutro e ácido, e a quantidade de nutrientes digestíveis totais. No experimento 1, o teor de proteína bruta da silagem aumentou de 7,5 para 12,6% da silagem de milho para a silagem com 39,2% de biomassa de soja, o que representa um aumento de 67,24%. No experimento 2, a proteína bruta aumentou de 6,77 para 12,09%, o que representa 78,58% a mais de proteína bruta no tratamento com 50% de adição de biomassa de soja verde (41% de matéria seca de soja) em relação à silagem de milho. No experimento 2, para cada 1% de acréscimo de biomassa seca de soja, tem-se elevação de 0,1% de proteína bruta. A adição de biomassa de soja a silagem de milho, eleva os teores de matéria mineral e proteína bruta da silagem.

**Palavras-chave:** matéria mineral; proteína bruta; digestibilidade; *Glycine max*; *Zea mays*

## Introduction

Animal feeding is increasingly discussed in the farming and scientific field (Jobim et al., 2010). According to the same researchers, further studies aiming at the use of technologies that ally efficiency and economy in animal production, are essential. Belel et al. (2014) emphasized that forage production improvement has great relevance for animal production, being a fundamental factor in the current production systems. In addition, the use of conserved silage is a well-known option in Brazil, especially in periods of lack of green fodder (Stella et al., 2016).

For the ensiling process, maize (*Zea mays* L.) stands out as the main used forage crop, due to its high dry matter yield per area, sufficient amount of sugars for the production of lactic acid, good fermentation, providing a feed with approximately 7.7% of crude protein (Goes et al., 2013). Despite that, studies have been showing that the addition of legume biomass, including soybean, has the potential to increase silage yield (Baghdadi et al., 2016; Tsujimoto et al., 2016) and enhance the quality of corn silage, especially the crude protein content (Keplin 2004; Stella et al., 2016; Kim et al., 2018; Batista et al., 2018; Batista et al., 2019).

According to Stella et al., (2016), maize silage presents 7.31% of crude protein, and this value increased to 10.5 and 13.6%, with the addition of 25 and 50% of soybean dry matter biomass, respectively. Likewise, Keplin (2004) points out that soybean silage may produce 2.5 times more crude protein per kilogram of dry matter compared to corn silage.

Thus, its inclusion/addition level should be carefully evaluated to avoid problems in the fermentation process and bromatological quality of silage caused by the amount of water present in plants (Ghizzi et al., 2017). Evangelista et al. (2003) reported bad silage fermentation when soybean was ensiled alone due to its higher water content. Due to it, further investigations are necessary to explore the effect of different proportions between maize and soybean biomass and their effects on silage quality.

It is worth noting that among legume species, soybean is one of the best options to be used as intercrop specie with maize for silage production. There are many transgenic events for pest and weed management, as well as aspects that allow cultivation in several regions, with a great supply and low cost of seeds when compared to other legumes.

Considering these facts, the aim was to evaluate the effect of different percentages of soybean biomass addition on the Bromatological traits of corn silage.

## Materials and Methods

This experiment was carried out during 2017/2018 growing season at Agricultural Research Station and the Bromatological Analysis Laboratory of the Federal Technological University of Paraná (UTFPR), campus of Dois Vizinhos, located at 25° 42' 52" latitude south, 53° 03' 94" longitude west, and at 510 m above sea level. The experimental area has a consolidated no-tillage system with the climate classified as Cfa (Alvares et al., 2013). The soil is classified as a Clayey Oxisol (Bhering et al., 2009). The average rainfall ranges from 1,800 to 2,200 mm per year (IAPAR, 2019).

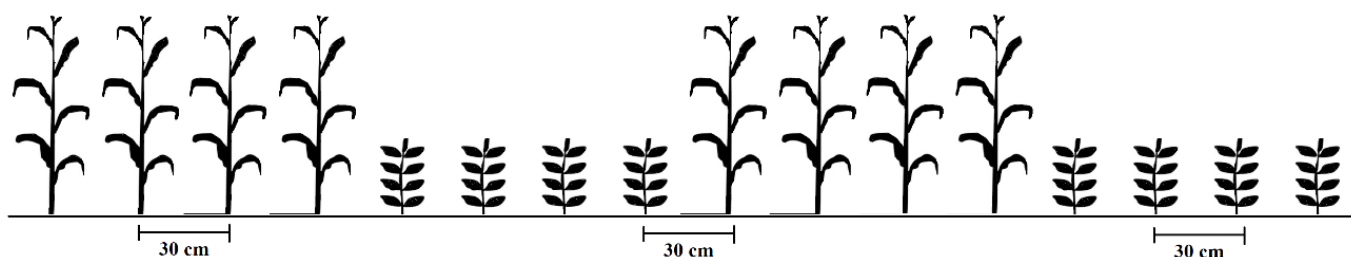
Two studies in a completely randomized design with four replications were conducted. Treatments were composed by adding percentages of soybean green biomass into maize ensiling. At experiment 1, soybean green biomass addition of 0, 10, 20, 30, and 40% into maize silage were evaluated and at experiment 2 were kept the same levels adding a treatment with 50% of soybean green biomass.

At the first experiment, maize (P30F53 hybrid) and soybean (TMG 7062 Intacta RR2 Pro) biomass samples were collected from commercial crop fields grown as the UTFPR experimental station. Maize kernel milk line stage (½ milk ½ dough stage) was used as an indicator of when to harvest maize fields to silage. Soybean phenological stage of 7.0- beginning maturity (one normal pod on the main stem has reached its mature pod color) was determined at the right moment to harvest soybean for silage, which according to Leonel et al. (2008), allows to obtain the higher silage crude protein levels.

Considering the results from the first experiment and the benefits provided by maize and soybean intercropped reported in the literature research (Baghdadi et al., 2016; Tsujimoto et al., 2016), the second experiment was carried out in an intercropping system. Maize hybrid 2B533PW and soybean cultivar TMG 7062 Intacta RR2 Pro was used as plant material.

On October 02, 2017, the intercrop of maize and soybean was sown simultaneously, using a seed drill with maize seed disc of 28 holes and soybean seed disc of 100 holes. Seed drill regulation was set up to sow 62.000 maize seeds ha<sup>-1</sup> and consequently 221.429 soybean seeds ha<sup>-1</sup>. Intercropping was obtained by alternating four maize rows with four soybean rows, with 30 cm row spacing (Figure 1).

The experimental site of the second experiment has been cultivated for years in a no-tillage system, with crop rotation. Soil showed the following traits: 57% clay, 33% silt and 10%



**Figure 1.** Row arrangement of maize and soybean intercropped for silage. UTFPR – Dois Vizinhos-PR, Brazil (2019).

sand, and chemical properties in the 0-20 cm layer of 3.43, 4.30, 0.90, and 0.90  $\text{cmol}_c \text{ dm}^{-3}$  for H + Al, Ca, Mg, and K respectively. It was also observed values of 21.63  $\text{mg dm}^{-3}$  of P, organic matter 44.23  $\text{mg dm}^{-3}$ , pH 5.20, and base saturation 64.08%.

Temperature and precipitation data observed during the second experiment are shown in Figure 2.

Black oat (*Avena strigosa*) was used as prior crop and it was desiccated with glyphosate (1.080  $\text{g ha}^{-1}$  of active ingredient) 30 days before intercrop establishment.

At sowing, fertilization was used for both species adding 450  $\text{kg ha}^{-1}$ , of chemical fertilizer 5-20-10 (N-P<sub>2</sub>O<sub>5</sub>-K<sub>2</sub>O). Nitrogen (N) was applied as urea (45% of N) at the rate of 180  $\text{kg N ha}^{-1}$ . Half of the N dose was applied at V4 (collar of fourth leaf visible) (three weeks after sowing) and the remaining half at V8 (collar of 8<sup>th</sup> leaf visible), all by side placement manually along the rows. Insecticide *imidacloprid + beta-cyfluthrin* at the dose of 1 L  $\text{ha}^{-1}$  was applied, shortly after maize emergence to control stink bug (*Dichelops melacanthus*). Weed control was achieved by applying *glyphosate* (1.2  $\text{g a.i. ha}^{-1}$ ) on maize at V3 stage (collar of third leaf visible). Fungicide application was done at maize blister (R2 stage) with a systemic ready mixture product containing *estrobilurina + pirazol carboxamida* at a commercial dose of 300  $\text{g ha}^{-1}$ . Along with the fungicide, vegetable oil was added at a dose of 0.5 L  $\text{ha}^{-1}$  and spray volume of 160 L  $\text{ha}^{-1}$  applied with a self-propelled sprayer.

Maize ensiling point occurred on 01/30/2018, which happened 120 days after its emergence. At that point, soybean cultivar was at 5.3 phenological stage (seeds are between 25 and 50% developed at one of the four upper nodes on the main stem).

At the corn silage point, in both studies, corn and soybean plants were harvested by hand cutting the plants at 25 cm above soil surface. Plants from experimental unit were harvested in four sampling area of 4.5  $\text{m}^2$  (5.0 x 0.9 m) for experiment 1 and 4.8  $\text{m}^2$  (4.0 x 1.2 m) for experiment 2. The

samples were weighed to determine maize and soybean biomass yield, being the values extrapolated to hectare ( $\text{kg ha}^{-1}$ ) (green biomass (GB)).

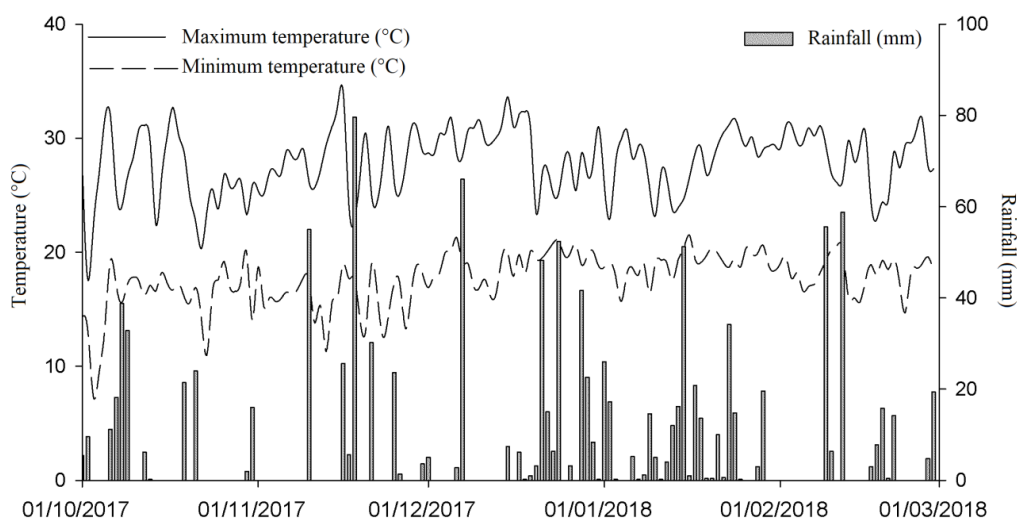
Then, samples of both crops of each experimental unit (EU) were ground separately on a forage harvester coupled to a tractor with an average particle size of 1.5 cm. Four subsamples of soybean and corn ground biomass with 300 g each were placed in paper bags and oven-dried with forced air circulation at 65 °C until constant mass to determinate its dry matter content of each experiment.

Samples of 3 kg of biomass were prepared (corn green biomass + fraction of soybean green biomass), mixed for total homogenization and packed compactly into Laboratory silos made of PVC pipes, measuring 100 mm in diameter and 500 mm in length. At the time of ensiling, the silos were sealed with PVC caps and stored for 60 days for fermentation.

Upon the opening of the silos, the determination of pH was carried out in the silage in accordance with the methodology described by Silva & Queiroz (2002). Samples collected (300 g) after the opening of the silos were placed in paper bags and oven-dried with forced air circulation at 65 °C until constant mass. Dried samples were ground in a 'Willey' type mill with a 1mm mesh sieve, and the samples taken to the Bromatological Analysis Laboratory of the UTFPR.

Further analysis of dry matter, ashes (%) (Silva & Queiroz, 2002), neutral detergent fiber (NDF), acid detergent fiber (ADF) (%) were determined by the methodology described in the Ankom (2009) manual. TDN = 87.84 - (0.7 x ADF) (Pionner, 2019) was used to estimate the total digestible nutrients (TDN). Silage crude protein (SCP) ( $\text{g kg}^{-1}$ ) analysis was performed by quantifying the N present in the samples, with the total N being determined in Kjeldhal semi-micro steam distillation methodology (Tedesco et al. 1995).

Dry mass and biomass yield values for crop silage are presented in a descriptive way. The other variables were submitted to analysis of variance (ANOVA) ( $p < 0.05$ ) and



Source: IAPAR (2019).

**Figure 2.** Maximum and minimum temperature (°C) and rainfall (mm) recorded during the intercropping Maize and soybean for silage. UTFPR – Dois Vizinhas-PR, Brazil (2019).

when it present significance was applied linear and quadratic regression analysis. For analysis of data, Sisvar 5.6 (Ferreira, 2008) software was used.

## Results and Discussion

At the ensiling point (Experiment 1), maize and soybean crop showed a dry matter (DM) value of 34.1 and 29.7% respectively. At the second experiment, these values were of 35.8 and 24.9% respectively. The difference between the studies for soybean biomass (4.8% DM) is related to the phenological stage at the time of cutting and/or ensiling. At the first experiment, the legume was at the 7.0 phenological stage and in the second at 5.3. Because of this difference in DM contents of the crops, it was observed that the DM proportions in the silage, changed in relation to the proportion of green biomass (GB) placed into the silos (Table 1).

It is worth to comment that when a silo is made, the farmer is ensiling green biomass, and thus, as a research source, the green mass content of the silage should be considered.

The analysis of variance (ANOVA) showed a significant effect of the evaluated treatments on ashes and crude protein

from experiment 1, and for pH, ashes, and crude protein from experiment 2 (Table 2).

The pH of silage was not influenced by the percentages of soybean biomass in experiment 1, presenting average values of 4.49. However, it was observed in experiment 2 that pH was influenced by the addition of soybean percentages, being the quadratic model, which best represents the results (Figure 3A). According to the mathematical model ( $y = -0.00006x^2 + 0.0066x + 4.3611$ ), the highest pH values (4.54) are recorded in the silage composed of 55.00% of DM of soybean in the amount of ensiled biomass.

This increase in pH in the second experiment may be related to the higher percentage of moisture content in the soybean biomass. Evangelista et al. (2003), observed that the high moisture in the silo may compromise the microbial fermentation of the ensiled biomass and, consequently, changing the pH values of the silage.

It was observed that the pH values increased from 4.35 in the maize monocrop (100% M) to 4.52 in silage composed of equal fractions of maize and soybean (50% M + 50% S) (experiment 2 - Figure 3A). The pH variation among treatments of only 0.17 allows to infer that it did not interfere in the silage quality.

**Table 1.** Green biomass (GB) and dry matter (DM) in maize silage with the addition of percentages of soybean biomass, UTFPR–Dois Vizinhas, Brazil (2019).

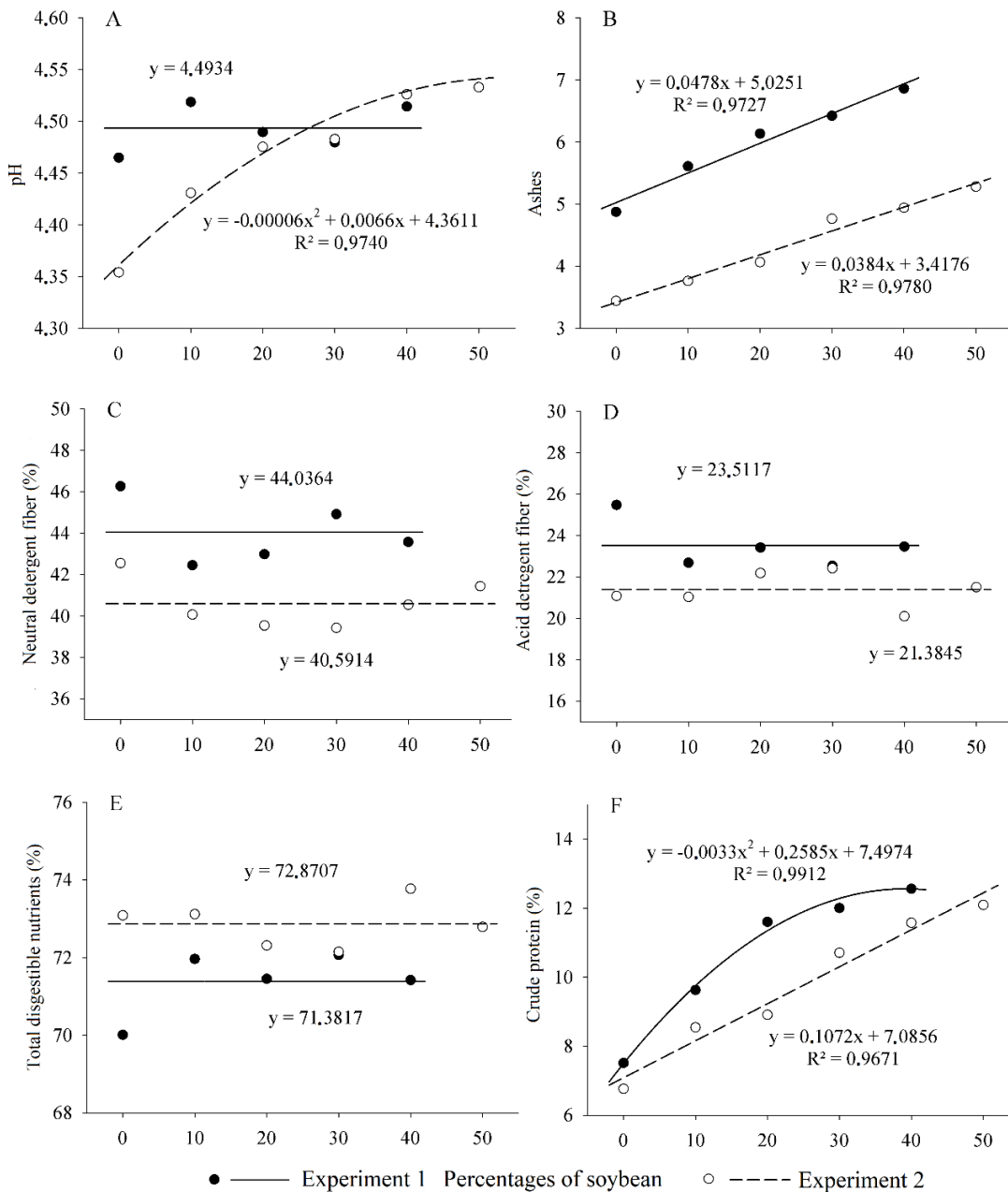
Treatments	GB (g)		DM (g)		Total DM (g)	Total DM (%)	
	Maize	Soybean	Maize	Soybean	Maize+Soybean	Maize	Soybean
Experiment 1							
100%M	3,000	0	1,023.0	0.0	1,023.0	100.0	0.0
90%M+10%S	2,700	300	920.7	89.1	1,009.8	91.2	8.8
80%M+20%S	2,400	600	818.4	178.2	996.6	82.1	17.9
70%M+30%S	2,100	900	716.1	267.3	983.4	72.8	27.2
60%M+40%S	1,800	1,200	613.8	356.4	970.2	63.3	36.7
Experiment 2							
100%M	3,000	0	1,074.0	0.0	1,074.0	100.0	0.0
90%M+10%S	2,700	300	966.6	74.7	1,041.3	92.8	7.2
80%M+20%S	2,400	600	859.2	149.4	1,008.6	85.2	14.8
70%M+30%S	2,100	900	751.8	224.1	975.9	77.0	23.0
60%M+40%S	1,800	1,200	644.4	298.8	943.2	68.3	31.7
50%M+50%S	1,500	1,500	537.0	373.5	910.5	59.0	41.0

M = Maize. S = Soybean.

**Table 2.** Means and P-values of the bromatological traits of maize silage with the addition of different percentages of soybean biomass. UTFPR–Dois Vizinhas, Brazil (2019).

	pH	Ashes	NDF	ADF	TDN	CP
Experiment 1						
P-value (ANOVA)	0.6498 <sup>ns</sup>	0.0017*	0.1452 <sup>ns</sup>	0.3944 <sup>ns</sup>	0.3944 <sup>ns</sup>	0.0000*
Linear regression P-value	-----	0.0001*	-----	-----	-----	0.0000*
Quadratic regression P-value	-----	0.4471 <sup>ns</sup>	-----	-----	-----	0.0030*
Means	4.49	5.98	44.04	23.51	71.38	10.66
CV (%)	1.29	9.45	4.96	9.51	7.20	6.76
Experiment 2						
P-value (ANOVA)	0.0000*	0.0099*	0.6609 <sup>ns</sup>	0.9236 <sup>ns</sup>	0.9236 <sup>ns</sup>	0.0000*
Linear regression P-value	0.0000*	0.0003*	-----	-----	-----	0.0000*
Quadratic regression P-value	0.0362*	0.8988 <sup>ns</sup>	-----	-----	-----	0.2931 <sup>ns</sup>
Means	4.47	4.38	40.59	21.25	72.87	9.76
CV (%)	0.76	16.08	7.34	15.25	9.87	8.85

\* Significant at  $p \leq 0.05$  level and <sup>ns</sup> non-significant at the  $p > 0.05$  level. CV= Coefficient of variation; pH = potential hydrogen; NDF= neutral detergent fiber; ADF = acid detergent fiber; TDN = total digestible nutrients; CP = crude protein.



**Figure 3.** Bromatological traits of maize silage with the addition of different percentages of soybean biomass UTFPR – Dois Vizinhas, Brazil (2019).

According to Neves & Gai (2017), the pH values found in the present experiment characterize good quality silage and are similar to those observed in other studies using sole maize crop silage (Neves & Gai, 2017; Pauli et al., 2017) and composed silage (corn + soybean) (Stella et al., 2016; Batista et al., 2018; Batista et al., 2019). In this context, it can be inferred that the addition of soybean biomass does not affect the microbial fermentation of the ensiled biomass.

Regarding to the ashes, it was noticed that its silage content increased in a linear way as the proportion of soybean biomass increased (Figure 3B). A similar result was observed by Jobim et al. (2010) evaluating silage of maize grains with addition of soybean grains. According to the researchers, this performance of the ashes can be explained by the reduction of other components of the dry mass (carbohydrates), which

normally occur in the process of ensilage of corn, resulting in an increase in the concentration of the ashes with the addition of the legume.

No effects of soybean biomass on neutral detergent fiber (NDF), acid detergent fiber (ADF) and total digestible nutrients (TDN) were observed (Table 2), with observed NDF values of 44.04 and 40.59% (Figure 3C), ADF of 23.51 and 21.38% (Figure 3D), and TDN of 71.38 and 72.87% (Figure 3E), for studies 1 and 2, respectively. These values are similar to those observed by Batista et al. (2019) in silages containing approximately 90% corn biomass and 10% soybean biomass (NDF - 38.34%, ADF - 20.02% and TDN- 73.83%).

The higher concentration of NDF in the animal diet, the lower is the animal feed intake capacity (Ghizzi et al., 2017). Thus, similar fibers values observed between composed



silages (maize and soybean) and sole maize crop silage, is somehow desired, since other silage bromatological traits were increased. Van Soest (1994) reported that adequate levels of NDF and ADF of silage for animal intake should be below 60% and 40%, respectively, which are in accordance with those observed in the silages evaluated.

Regarding the crude protein, the evaluated treatments had significant effect on the percentage of crude protein of the silage (Table 2). In experiment 1, the quadratic model is the one that best represents the system, being observed maximum efficiency point (12.56% of CP) when using 39.17% of soybean biomass (Figure 3F). For the sole maize crop, the value of CP was 7.51%, while it was observed an increase of crude protein content of 67.24% when 39.17% of soybean dry matter biomass was added in the composed silage.

At the experiment 2, a linear increase of CP was observed as soybean biomass addition to maize silage increased (Figure 3F). Maize silage presented 6.77% of CP versus 12.09% in silage composed of 50% of corn and soybean green biomass (59 and 41% dry matter of corn and soybean, respectively), representing an increase of 78.58% on the protein content. Thus, each 1% increase in soybean biomass, there was an increase of 0.1% in crude protein contents (Experiment 2). This result shows that soybean biomass ensiling, along with maize biomass, has the potential to raise the protein content of the silage, improving its composition.

These results corroborate with other studies that evaluated these intercrop (Batista et al., 2018; Batista et al., 2019) with different proportions of ensiled biomass (Stella et al., 2016). According to Stella et al. (2016), from the standpoint of chemical composition, the ensiled corn biomass could be composed of up to 50% soybean plant, resulting in improved silage traits (CP) without affecting its fermentation quality (pH and N-NH<sub>3</sub> concentration).

Gobetti et al. (2011) emphasize that soybean biomass presents high nutritional value, compared to corn silage, and this fact may reduce the demand of concentrate in the animal diet. According to Stella et al. (2016), through the association of corn and soybeans, it is possible to reduce the ratio of roughage/concentrate in the animal diet, generating a reduction in the final cost of feeding due to the less need of protein source from a high-cost supplement, such as the soybean meal (Stella et al. 2016).

According to the results of these studies, the addition of soybean biomass in maize silage is an excellent alternative to raise the crude protein content of the silage.

Concerning to the biomass yield (Experiment 1) at the ensiling point, maize produced 63,449.01 kg ha<sup>-1</sup> of GB (21,636.11 kg ha<sup>-1</sup> DM), and soybean 18,281.62 kg ha<sup>-1</sup> of GB (5,429.64 kg ha<sup>-1</sup> DM). It is important to highlight that the crops were cultivated isolated. For experiment 2, crops were intercropped (4 row of maize and 4 row of soybean), and maize and soybean yielded 60,403.83 kg ha<sup>-1</sup> of GB (21,624.57 kg ha<sup>-1</sup> DM) and 10,849.44 kg ha<sup>-1</sup> of GB (2,701.51 kg ha<sup>-1</sup> DM), respectively.

Considering the biomass yield data from both studies (although the experiment happened at different growing seasons), it is possible to infer that the intercropping system is more advantageous in relation to the monocrop, since there was no reduction of maize biomass yield in the intercropping which added to the soybean biomass resulted in higher biomass yield, evidencing that the intercrop system increases biomass in relation to monocrop standing as a good option to optimize silage yield and quality. Moreover, soybean biomass yield in the intercropping presented considerable values of soybean biomass (2,701.51 kg ha<sup>-1</sup> DM), representing 11.1% of the ensiled dry matter, which can increase the amount of crude protein in the silage.

Maize biomass yield observed in both studies (1 and 2) were superior to those observed by Vieira et al. (2006), which evaluating 10 maize hybrids reported average yield of 45.59 and 18.17 t ha<sup>-1</sup> of GB and DM, respectively. Higher biomass yield from studies 1 and 2 is possibly related to the good climatic conditions observed during the growing period of the crops (Figure 1), and soil fertility, associated to the productive potential of the used hybrid.

## Conclusions

The addition of soybean biomass to corn silage provides pH changes and increases in ashes and crude protein content but does not affect digestibility (neutral and acid detergent fiber content and total digestible nutrients).

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