

# Removal of solids and chemical oxygen demand in poultry litter anaerobic digestion with different inocula

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# ABSTRACT

Population growth has contributed to increasing poultry production, entailing a high waste loading, mainly poultry litter. One of the alternatives to treat such residues is anaerobic digestion, in which digester startup and generated-digestate quality are related to the material to be digested and to operation conditions, wherein inoculum use is one of the factors. This study therefore aimed to investigate how digestates, such as inocula, influence poultry litter (PL) anaerobic digestion, as well the reduction of solids and chemical oxygen demand (COD). For this, two inocula (bovine and swine digestates) were tested in the digestion process. The inocula were added at loads of 0.67, 1.00 and 1.67 gVS.L<sup>-1</sup>day<sup>-1</sup>. A split-plot design was developed and data underwent analysis of variance with means compared by the Tukey's test at 5% significance. Concerning bovine and swine inocula, it was concluded that both are indicated in the process. However, swine inoculum is better indicated because it had a better removal of total solids (TS), volatile solids (VS) and COD.

Keywords: biodegradability, bovine inoculum, poultry waste, swine inoculum.

# Remoção de sólidos e da demanda química de oxigênio na biodigestão anaerobia da cama de aviário com diferentes inóculos

# **RESUMO**

O crescimento populacional contribuiu para o aumento da produção avícola, o que implica na geração de resíduo de cama de aviário. Uma das alternativas para tratar esses resíduos é a digestão anaeróbia, na qual a partida do digestor e a qualidade do digestato gerado estão relacionadas ao material a ser digerido e às condições de operação, nas quais o uso do inóculo é um dos fatores. Portanto, este estudo teve como objetivo investigar como os inóculos influenciam na digestão anaeróbia da cama de aves, bem como na redução de sólidos e da demanda química de oxigênio (DQO). Para isso, foram testados dois inóculos (bovinos e suínos) no processo de digestão, com cargas de alimentação diárias de 0.67; 1.00 e



1.67 gSV L<sup>-1</sup> dia<sup>-1</sup>. Para a análise estatística foi considerado o delineamento em parcelas subdivididas e os dados foram submetidos à análise de variância com as médias comparadas pelo teste de Tukey com 5% de significância. Em relação aos inóculos de bovinos e suínos, concluiu-se que ambos são indicados ao processo. No entanto, o inóculo suíno é mais indicado porque teve uma melhor remoção de sólidos totais (TS) e DQO.

Palavras-chave: biodegradabilidade, inóculo bovino, inóculo suíno, resíduo de aviário.

### **1. INTRODUCTION**

The United States of America, Brazil, and China were responsible for the greatest production of poultry meat worldwide in 2017. Among them, Brazil reached a production of 13.3 million tons, with the largest values achieved by the states of Paraná, Santa Catarina and Rio Grande do Sul (ABPA, 2018).

Within this background, the broiler industry is responsible for a large consumption of natural resources and, consequently, waste generation, including chicken litter, within an average value of 1,262 kg/(bird year). This value depends on substrate type, season, number of flocks, cycle length and bird density (Rinaldi *et al.*, 2012; Migliavacca and Yanagihara, 2017).

Improper disposal of waste may engender large-scale contaminations of land, water, and air (Scherhaufer *et al.*, 2018). Because of this, environmental problems have intensified recently, raising the need for alternatives to minimize impacts and to add value to generated wastes. As the poultry industry has expanded, in addition to producing large quantities of meat, waste production has also increased proportionately (Silveira *et al.*, 2014).

Anaerobic treatment of agricultural waste is not only an alternative for energy recovery but also for pollution mitigation (Alice *et al.*, 2014). This technique is well established for renewable energy production and includes waste treatment as an additional benefit.

Although anaerobic digestion has proven to be a good tool for degrading several pollutants and organic wastes, it is a complex process and requires monitoring of pH, alkalinity, temperature, volatile fatty acids, organic load, hydraulic retention times, biogas composition, etc. (Mao *et al.*, 2015; Amaral *et al.*, 2014).

Sanchez Rubal *et al.* (2012) determined the influence of temperature, stirring, sludge concentration, and solid retention time (SRT) on biodegradable organic matter acquisition during primary fermentation, clarifying each operational parameter effect on hydrolysis.

In addition to biogas production, Orrico Junior *et al.* (2010) evaluated potential production and quality of biofertilizers derived from anaerobic digestion of pre-composted poultry litter and carcasses. These authors reported sharp reductions in VS contents, on average 44.05%, showing that the process is effective in degrading resistant compounds, such as bird litter.

Recent studies have prospected growth medium for microalgae using effluents from poultry litter anaerobic digestion (Singh *et al.*, 2011). Moreover, digestates, widely used as biofertilizers, can also be employed as inoculum in anaerobic digestion, playing a balancing role on microorganism populations at the beginning of this process (Shah *et al.*, 2014).

Onofre *et al.* (2015) have tested the biofertilizer from previous poultry litter anaerobic processes as inoculum, in order to minimize the start of the treatment.

Cattle, poultry, and swine biofertilizers may serve as inocula, mainly for difficult to digest materials, because of the high contents of cellulose and lignin, as seen in poultry litter. Costa *et al.* (2012) investigated such issues and observed that biological co-treatment and thermochemical pretreatments improved poultry litter hydrolysis but led to an accumulation of metabolites, inhibiting methanogenesis.

The inoculum source can affect the amount of biogas produced in anaerobic digestion as well as influence the process speed (Swiatczak *et al.*, 2017). Furthermore, several dynamic and microbial factors contribute to the efficiency of organic waste treatment.



It is worthy to note that different inocula lead to different biogas production, with significant effect of the inoculum on the Biochemical Methane Potential (BMP) (Vrieze *et al.*, 2015). Another paper (Rico *et al.*, 2017) shows the influences of inocula on the anaerobic digestion of pig slurry in UASB reactor.

Therefore, this technology must be optimized to reach maximum benefits (Gyenge *et al.*, 2014). Understanding the microbial community and its role in the process is critical to the process control (Shen and Zhu, 2016).

Given this background, this study tested two types of inocula, bovine and swine, in the anaerobic digestion of poultry litter, quantifying reductions of solids and COD.

### 2. MATERIAL AND METHODS

#### 2.1. Substrate and Inocula

The substrate consisted of poultry litter gathered from a farm in the city of Quatro Pontes - PR, Brazil. Table 1 shows the characterization of the material with values of hydrogenation potential (pH), total solids (TS), fixed solids (FS), volatile solids (VS), moisture, N and chemical oxygen demand (COD). Prior to biodigestion, residue underwent a pre-treating process with 8 mm mesh sieving to increase its contact surface and facilitate anaerobic digestion.

Material	рН	TS (%)	FS (%)	VS (%)	Moisture (%)	N (%)	COD (mg.L <sup>-1</sup> )
Poultry litter	7.90 (±0.00)	85.00 (±1.10)	36.00 (±1.00)	79.00 (±1.00)	12.30 (±0.3)	1.8 (±0.1)	7306.36 (±50.0)
Inocula Bovine	7.34 (±0.01)	5.44 (±0.05)	2.65 (±0.03)	2.79 (±0.02)	-	-	-
Inocula Swine	7.65 (±0.00)	2.46 (±0.02)	1.78 (±0.03)	0.68 (±0.05)	-	-	-

 Table 1. Poultry litter and inocula physicochemical characterization.

As inoculum (IN), two types of fresh material were used: bovine (B) and swine (S), both collected from field biodigesters and kept at room temperature for characterization. The previous table shows the physicochemical characterization of both inocula.

#### 2.2. Poultry litter digestion

The trial was carried out at the Laboratory of Bioreactors of the Research Group on Water Resources and Environmental Sanitation of the Western Paraná State University.

Two biodigesters built in polyvinyl chloride (0.60 x 0.10 m) were operated. The equipment had an inlet port and a digestate outlet arranged horizontally, with 4 L useful volume, as shown in Figure 1.



Figure 1. Experimental module.

The two biodigesters were inoculated with 30% of their useful volume and 70% with a mixture of water and chicken litter at a ratio of 50 grams of litter for each mixture liter. Twenty-four hours after inoculation, removal of oxygen traces was completed and daily feeding was started, in a semi-continuous flow model, applying loads of 0.67 (L1), 1.00 (L2) and 1.67 (L3) gVS.L<sup>-1</sup>.day<sup>-1</sup>, at a concentration of 2% volatile solids. These loads were defined according to previous work (Alcantara *et al.*, 2016).

The operating times for each load were 74, 69, and 66 days, respectively, including stabilization time, monitored by the ratios VA/TA (Volatile Acidity/Total Alkalinity) and IA/PA (Intermediate Alkalinity/Partial Alkalinity). The temperature was room temperature, daily monitored.

The volume of biogas and methane produced was measured with a Mariotte device filled with sodium hydroxide (NaOH) solution to dissolve carbon dioxide (CO<sub>2</sub>) (Bertin *et al.*, 2004; Buitrón and Carvajal, 2010; Scoma *et al.*, 2013). However, these results are shown and discussed in another paper.

#### 2.3. Monitoring pH, Ammoniacal Nitrogen and Ratios VA/TA and IA/PA

The pH was determined using a benchtop pH meter (Model 3MP) and ammonia nitrogen concentration was monitored following the 4500-NH<sub>3</sub> titration method (APHA, 1998).

The values for the ratios VA/TA and IA/PA were measured through titration, observing the threshold values reported by Méndes-Acosta *et al.* (2010) and Ripley *et al.* (1986).

The total alkalinity in anaerobic systems is determined by titration of the sample to pH 4.30, measuring the buffer capacity due to bicarbonate and volatile acids, defined by Ripley *et al.* (1986) as partial alkalinity (PA) and intermediate alkalinity (IA) respectively.

Ripley *et al.* (1986) established that the ratio IA/PA with values greater than 0.30 indicates the occurrence of disturbances in the process of anaerobic digestion.

According to Méndez-Acosta *et al.* (2010), the VA/TA ratio should be between 0.10 and 0.35, to assure the stability of anaerobic digestion.

#### 2.4. Removal of solids, COD Removal and characterization of digestate and sludge

Measurements of TS, FS, and VS were performed based on method 2540 of APHA (2005). In this case, COD quantification was made following method 5220D of APHA (2005). For soluble COD, samples were filtered, using the blank test prepared with distilled water.

To determine the micro (Zn, Fe, Cu and Mn) and macronutrients (K, N, Mg, Na, Ca) of the digestate, a nitro-perchloric digestion (3:1) of the samples was performed and an aliquot was taken for reading with atomic absorption equipment.

#### 2.5. Statistics

Statistical analyses were performed following a split-plot design, wherein inoculum factor was the main plot (with two levels: B and S). The secondary plot consisted of feeding load factor (with three levels: L1, L2, and L3). The analyses were performed using SISVAR software v. 5.3 (Ferreira, 2010). As response variables, both ratios VA/TA and IA/PA were considered, as well as removal rates of total solids, total volatile solids, total fixed solids, COD total and COD soluble.

Data presenting normality by the Kolmogorov-Smirnov's test and homoscedasticity by the Bartlett or Levene's test had the variances submitted to analysis of variance, at 5% significance. Tukey's test was used for mean comparison at a 5% significance level.

### **3. RESULTS AND DISCUSSION**

#### 3.1. Stability through pH, ammonia N, IA/PA ratio e VA/TA ratio

With respect to pH, biodigesters remained stable for all treatments, with values near



neutrality, as shown in Figure 2A. The highest mean values of pH occurred at loads of 1.00 and 1.67 gVS.L<sup>-1</sup>.day<sup>-1</sup>, for bovine and swine inocula, respectively.

These results corroborate the findings of Onofre *et al.* (2015), who evaluated the pH of the biofertilizer from poultry litter and pointed out that the pH ranged from 6.6 to 8.0, and determined that the anaerobic digestion process was suited.

Although no variations in pH were observed, the presence of ammonia was also monitored throughout the process, and the highest concentration of ammonia (280.51 mg L<sup>-1</sup>) occurred with a load of 1.67 gVS. L<sup>-1</sup> day<sup>-1</sup> swine inoculum. According to Chen *et al.* (2008), ammonia concentrations greater than 1000 mg.L<sup>-1</sup> were harmful to the process. Since the presented value is below the concentration threshold reported in the literature, it was concluded that there was no effect of ammonia on anaerobic digestion.



Figure 2. Monitoring of the pH, IA/PA ratio and the VA/TA ratio.

It should be pointed out that the temporal scale shown in Figure 2A is different from the scales of the other figures because the pH monitoring was registered daily, a fact that did not occur with the monitoring of the ratios VA/TA and IA/PA, solids and COD removals (Figure 2B and 2C).

According to Chernicharo (2007), the monitoring of the partial or bicarbonate alkalinity is more useful than the pH monitoring due to scales characteristics: while the pH scale is logarithmic, the scale for alkalinity is linear. It is fundamental to maintain a low concentration of volatile fatty acids and pH in the interval 6.6 to 7.4 (Lahav and Morgan, 2004).

The main buffer in anaerobic digesters is the ion bicarbonate, at pH from 5.3 to 7.3, expressed in mg of CaCO<sub>3</sub> L<sup>-1</sup>, varying with the waste type (Lahav and Morgan, 2004).

According to Ripley *et al.* (1986), IA/PA values greater than 0.30 may indicate instability problems in digestion. Here, it was observed that IA/PA values for a load of 0.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> exceeded 0.30; however, decreasing over time and remaining stable for the other applied loads.

Figure 2B denotes that the highest mean values of IA/PA were observed for loads of 1.67 and 0.67 gVS.L<sup>-1</sup>.day<sup>-1</sup>, respectively for bovine and swine inocula; however, these values are still below the recommendation of Ripley *et al.* (1986).

Martín-Gonzalez *et al.* (2013) asserted a possible process stability for values different from 0.3 due to variations in the characteristics of each effluent.

Values for the ratio VA/TA were similar for both inocula, presenting slightly higher values in a load of 0.67 gVS.L<sup>-1</sup>.day<sup>-1</sup>, as seen in Figure 2C, ranging from 0.252 to 0.228 for bovine and swine inocula, respectively.

Table 2 shows the statistical breakdown of VA/TA ratio means. It can be observed that, by fixing the load factor and evaluating its variation in all inoculum levels, a significant difference occurred between loads. The highest values occurred for the loads of 0.67 and 1.67 gVS.L<sup>-1</sup>.day<sup>-1</sup>, considered statistically equal for bovine inoculum. By fixing the inoculum factor, there was no statistically significant difference between loading levels.

**Table 2.** Statistical breakdown of the means of VA/TA ratio as function of inoculum and load.

Inoculum	VA/TA										
	L1-0.67 gVS.L <sup>-1</sup>	.day <sup>-1</sup>	$L2 - 1.00 \text{ gVS.L}^{-1}$	.day <sup>-1</sup>	L3 – 1.67 gVS.L <sup>-1</sup> .day <sup>-1</sup>						
Bovine	0.2523(±0.09)	Aa	0.1186(±0.02)	Ab	0.1865(±0.04)	Aa					
Swine	$0.2286(\pm 0.08)$	Aa	$0.1286(\pm 0.02)$	Ac	$0.1749(\pm 0.04)$	Ab					

Means followed by the same uppercase letters within columns (inoculum) and lower case within lines (loads) do not differ from each other by the Tukey's test at 5% significance.

The greatest stability time was observed for a load of 1.00 gVS.L<sup>-1</sup>.day<sup>-1</sup>; however, at 0.67 gVS.L<sup>-1</sup>.day<sup>-1</sup>, the highest means were reached. Nevertheless, these values are within the optimum range for operating levels. The results corroborate those of Kuczman *et al.* (2011), who evaluated biogas production from cassava wastewater with increasing organic loads and feeding volumes. The microbial adaptation was analyzed by monitoring VA/TA ratio, which remained within a range between 0.14 and 0.30.

It is important to note that the operational conditions of the process affect the response of the indicators (Li *et al.*, 2014), indicating the unbalance between acid production and consumption and warning about a possible digestion failure.

The temperature mean values were from  $16^{\circ}$ C to  $25^{\circ}$ C and values below  $20^{\circ}$ C occurred only for a load of 1.00 gVS. L<sup>-1</sup> day<sup>-1</sup>, caused by climatic interference. From the mean and standard deviation, the variation in load was 2.1°C and, as reported by Deublein and Steinhauser (2011), methanogenic Archaea are able to change rapidly ideally but the temperature should not vary abruptly by  $\pm 2^{\circ}$ C.

#### 3.2. Removal of TS and VS

The highest means of TS removal were related to the load 0.67 gVS.L<sup>-1</sup>.day<sup>-1</sup>, with swine inoculum (59.46%), as shown in Figure 3A.

In addition, bovine inoculum promoted increase in TS removal as loads were raised, with the highest value at the load of 1.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> (57.36%).





Figure 3. Monitoring of total solid removal and of total volatile solid removal.

Table 3 displays the statistical breakdown of the means for TS removal. It is shown that by fixing load factor there was a significant difference as inoculum level increased, with the removal by bovine inoculum statistically equal for 0.67 and 1.00 gVS.L<sup>-1</sup>.day<sup>-1</sup>. The greatest value was observed for 1.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> (57.36%). Yet for swine inoculum, loads of 0.67 and 1.00 gVS.L<sup>-1</sup>.day<sup>-1</sup> were statistically similar to each other. On the other hand, loads of 0.67 and 1.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> differed from each other at 5% significance, with the largest removal observed for 0.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> (59.46%).

By fixing the inoculum factor, and assessing it according to the loads, there was a significant difference between both inocula at  $0.67 \text{ gVS}.\text{L}^{-1}.\text{day}^{-1}$ , being the greatest TS removal for the swine one (59.46%).

Inoculum	TS removal (%)										
	L1 – 0.67 gVS.L <sup>-1</sup>	.day <sup>-1</sup>	L2 – 1.00 gVS.L	<sup>-1</sup> .day <sup>-1</sup>	L3 – 1.67 gVS.L <sup>-</sup>	<sup>1</sup> .day <sup>-1</sup>					
Bovine	44.66(±10.51)	Aa	47.65(±6.02)	Ba	57.36(±6.39)	Bb					
Swine	59.46(±14.34)	Bb	48.30(±4.41)	Bba	52.57(±8.60)	Ba					

**Table 3.** Tukey's test for means of TS removal at 5% significance.

Means followed by the same uppercase letters within columns (inoculum) and lower case within lines (loads) do not differ from each other by the Tukey's test at 5% significance.

The highest VS removal values referred to a load of 1.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> using bovine inoculum, as shown in Figure 3B. Additionally, there was an increase in VS removal as loads increased, with bovine inoculum, being the greatest mean for a load of 1.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> (78.21%).

Table 4 displays the statistical breakdown of the means for VS removal. It is shown that by fixing load factor there was significant difference as inoculum level increased, with the greatest removal for bovine inoculum at a load of 1.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> (78.21%). Regarding swine inoculum, loads of 0.67 and 1.00 gVS.L<sup>-1</sup>.day<sup>-1</sup> are statistically equal, as well as 0.67 and 1.67 gVS.L<sup>-1</sup>.day<sup>-1</sup>. Conversely, loads of 1.00 and 1.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> differed at the 5% significance from each other, with the greatest removal for swine inoculum at a load of 1.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> (71.71%).

By fixing the inoculum factor and assessing it according to the loads, there was a significant difference between both inocula at 0.67 and 1.67 gVS.L<sup>-1</sup>.day<sup>-1</sup>. The greatest VS removal was found for swine inoculum at 0.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> (67.85%), and for bovine one at 1.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> (78.21%).

Inoculum	VS removal (%)										
	L1 – 0.67 gVS.L	<sup>1</sup> .day <sup>-1</sup>	$L2 - 1.00 \text{ gVS.L}^{-1}$	l.day <sup>-1</sup>	L3 – 1.67 gVS.L <sup>-1</sup> .day <sup>-1</sup>						
Bovine	50.45(±13.25)	Bc	65.07(±10.07)	Bb	78.21(±5.38)	Aa					
Swine	67.85(±13.64)	Aba	63.82(±9.53)	Bb	71.71(±8.54)	Ba					

Table 4. Tukey's test for means of VS removal at 5% significance.

Means followed by the same uppercase letters within columns (inoculum) and lower case within lines (loads) do not differ from each other by the Tukey's test at 5% significance.

The highest removal means, 59.46% (0.67 gVS.L<sup>-1</sup>.day<sup>-1</sup>) and 78.21% (1.67 gVS.L<sup>-1</sup>.day<sup>-1</sup>), were found for TS and VS, respectively. These findings corroborate Orrico Júnior *et al.* (2010), who evaluated the technical feasibility of anaerobic digestion of pre-composted poultry litter and carcasses. These authors noted the anaerobic digestion efficiency in degrading resistant compounds, such as chicken litter. Despite the sharp reductions in VS, around 44.05%, this value was lower than that obtained here, evidencing microbial action in the inoculum.

This way, the results encountered here are in agreement with those of Silva *et al.* (2013), who concluded that anaerobic digestion is an efficient procedure to treat organic residues. They also ascertained that biofertilizer addition not only favored system performance regarding pH, but also influenced biogas production and reduced total solids by 40.85%.

#### 3.3. Removal of COD (total and soluble)

The major means of COD<sub>total</sub> removal were registered at loads of 0.67 and 1.00 gVS.L<sup>-1</sup>.day<sup>-1</sup>, using both swine and bovine inocula, respectively, as shown in Figure 4A.



removal.



As seen in Figure 4B, the load of  $1.00 \text{ gVS.L}^{-1}$ .day<sup>-1</sup> provided the major means of  $\text{COD}_{\text{soluble}}$  removal. Since data regarding  $\text{COD}_{\text{total}}$  removal showed no normality, results did not undergo variance analysis.

Table 5 presents the statistical breakdown of means for  $\text{COD}_{\text{soluble}}$  as a function of inoculum and loads. When load factor is fixed, it is noteworthy seeing a significant difference of loads using bovine inoculum, with the emphasis placed at the load of 1.00 gVS.L<sup>-1</sup>.day<sup>-1</sup>, which provided the highest mean (61.31%). For swine inoculum, there was a significant difference among loads, with the highest removal reached by 1.00 gVS.L<sup>-1</sup>.day<sup>-1</sup> (53.29%), being statistically equal at 5% significance for a load of 0.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> (48.91%).

When fixing the inoculum factor and evaluating it in relation to the load levels, we could note a statistically significant difference between both inocula at 0.67 and 1.67 gVS.L<sup>-1</sup>.day<sup>-1</sup>.

COD soluble was effectively removed by using a load of 0.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> (48.91%) for swine inoculum, and at 1.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> (46.32%) for bovine.

The results found in this study are superior to those reported by Blanco *et al.* (2014), who studied anaerobic digestion of dairy cattle manure with the addition of poultry litter. These authors reported a low COD removal efficiency, on average 36%, which resulted in a low stability for the biofertilizer.

Inoculum	% COD soluble removal									
	L1 – 0.67 gVS.L <sup>-1</sup>	.day <sup>-1</sup>	L2 – 1.00 gVS.L <sup>-1</sup>	.day <sup>-1</sup>	L3 – 1.67 gVS.L <sup>-1</sup>	.day <sup>-1</sup>				
Bovine Swine	37.23(±21.07) 48.91(±13.75)	Bb Ab	61.31(±12.66) 53.29(±8.64)	Ba Bb	46.32(±14.11) 28.48(±15.32)	Ab Ba				

Table 5. Tukey's test for means of COD soluble removal at 5% significance.

Means followed by the same uppercase letters within columns (inoculum) and lower case within lines (loads) do not differ from each other by the Tukey's test at 5% significance.

The results obtained corroborate Lynch *et al.* (2013), who presented a review detailing advances in the three main alternative disposal routes for poultry litter, which are composting, anaerobic digestion, and direct burning. These authors concluded that such technologies open up opportunities to marketing both energy and nutrients in poultry litter, mainly for anaerobic digestion, once operation conditions are monitored.

It is known that the presence of inoculum influences the methanogenic activity and can present significant differences for different inoculum sources (Vrieze *et al.*, 2015; Rico *et al.*, 2017). In this study, it is observed that different types of inoculum may also influence the removal of COD and solids.

#### 3.4. Characterization of digestate and sludge

The quality of the digester and its potential for agronomic use depends on many factors. Because of this, each project have to include a specific analysis to determine available nutrient content (Nicoloso *et al.*, 2019).

Regarding the values of micronutrients (Zn, Fe, Cu and Mn) in Table 6, note that the highest mean Zn values are for the 1.00 gVS.L<sup>-1</sup>.day<sup>-1</sup> load. Fe values refer to the loads 1.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> and 0.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> for bovine and swine inoculum, respectively.

The highest values of Cu refer to the load 1.67 gVS.L<sup>-1</sup>.day<sup>-1</sup>, and those of Mn to the load 0.67 0.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> and 1.00 gVS.L<sup>-1</sup>.day<sup>-1</sup>, for bovine and swine inoculum respectively.

It is observed that the highest mean values of K refer to the loads 0.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> and 1.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> for bovine and swine inocula, respectively. There is a decrease in the concentration of this element with the use of bovine inoculum and increase with swine inoculum in relation to increased loads.

Inoculum	Feeding Loads	Micronutrients (mg.L <sup>-1</sup> )		Mean	SD	CV (%)	Macronutrients (mg.L <sup>-1</sup> )		Mean	SD	CV (%)
	L1 0.67			2.20	±0.84	38.21			534.60	$\pm 44.60$	8.51
Bovine	L2 1.00			2.41	±0.64	26.84			484.70	±42.70	8.81
	L3 1.67		Zn	1.95	$\pm 0.18$	9.56		К	478.60	$\pm 17.70$	3.69
	L1 0.67		_	2.35	±0.31	13.19		_	429.80	±27.80	6.46
Swine	L2 1.00			2.04	±0.16	7.88			486.24	$\pm 15.50$	3.19
	L3 1.67			3.36	±1.07	31.93			528.82	±10.86	2.05
	L1 0.67			4.63	±0.38	8.27			350.00	$\pm 24.60$	7.02
Bovine	L2 1.00			3.09	±1.12	36.23			359.33	±3.52	0.98
	L3 1.67		Fe	5.04	±0.16	3.27		Ν	344.87	±5.70	1.65
	L1 0.67			5.64	±0.33	5.92			356.80	$\pm 67.50$	18.91
Swine	L2 1.00			4.91	±1.59	32.50			333.43	$\pm 50.90$	5.01
	L3 1.67			3.94	±1.35	38.88			367.30	±18.37	13.87
	L1 0.67			0.31	±0.11	35.75			75.69	±5.65	7.46
Bovine	L2 1.00			0.27	$\pm 0.06$	23.27			54.95	±13.99	25.47
	L3 1.67		Cu	0.35	±0.07	19.50		Mg	50.84	±11.03	21.70
	L1 0.67			0.47	$\pm 0.11$	24.74			103.50	$\pm 12.70$	9.24
Swine	L2 1.00			0.42	$\pm 0.04$	10.93			62.88	±3.19	5.08
	L3 1.67			0.52	±0.08	15.37			63.78	±6.97	10.93
	L1 0.67			1.38	±0.21	15.65			316.60	$\pm 23.60$	7.45
Bovine	L2 1.00			1.33	±0.12	9.56			231.90	$\pm 25.60$	11.02
	L3 1.67		Mn	1.17	±0.17	14.57		Ne	139.42	±7.30	5.24
	L1 0.67			1.36	±0.04	3.31		INA	259.82	±17.23	6.63
Swine	L2 1.00			1.38	$\pm 1.05$	3.67			217.90	$\pm 21.70$	9.98
	L3 1.67			1.31	±0.09	7.00			157.75	±9.57	6.07
	L1 0.67			-	-	-			387.20	±40.60	10.49
Bovine	L2 1.00			-	-	-			366.92	$\pm 14.85$	4.05
	L3 1.67			-	-	-		Ca	305.10	$\pm 60.00$	19.66
	L1 0.67			-	-	-			291.80	$\pm 35.50$	12.16
Swine	L2 1.00			-	-	-			361.22	$\pm 15.21$	4.21
	L3 1.67			-	-	-			386.00	±35.70	9.24

#### **Table 6.** Characterization of digestate.



The highest average values of N refer to the loads of 1.00 gVS.L<sup>-1</sup>.day<sup>-1</sup> (359.33 mg.L<sup>-1</sup>) and 1.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> (367.30 mg.L<sup>-1</sup>) for bovine and swine inocula, respectively. In addition, the values of K and N are below the values found in effluent from poultry litter digestion by Singh *et al.* (2002): potassium (1632 mg.L<sup>-1</sup>) and nitrogen (1570 mg.L<sup>-1</sup>).

The highest average values of Mg and Na refer to the load 0.67 gVS.L<sup>-1</sup>.day<sup>-1</sup>, in addition to a decrease in the concentration of these elements with the increased loads. The Ca values refer to the loads of 0.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> and 1.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> for the use of bovine and swine inoculum, respectively. An increase in calcium (Ca) concentration was also observed with increasing charges for swine inoculum.

The results presented corroborate with Tessaro *et al.* (2015), which evaluated the digestion with poultry litter by varying the presence of digestate and water, and concluded that the digestate produced presented assimilable macro and micronutrients by vegetables such as nitrogen, phosphorus, potassium, calcium, magnesium, sodium, iron, boron, copper, zinc and manganese. However, it must be taken into consideration that nutrient loss and segregation processes may occur in the biodigester (Nicoloso *et al.*, 2019).

At the end of the experiment, biodigester sludge was quantified, but not presented differences in relation to the volume of sludge generated in the process (1.40 liters), being characterized in relation to nutrients, as presented in Table 7.

		0	υ					
Nutrients (mg.L <sup>-1</sup> )	Zn	Fe	Cu	Mn	K	Mg	Na	Ca
Sludge Bovine Sludge Swine	41.98 43.54	140.60 134.11	12.40 13.80	51.70 13.80	457.89 526.69	328.24 316.92	140.49 151.32	2815.58 3630.22

**Table 7.** Characterization of sludge the biodigester.

The bovine and swine sludge presented very close concentration in relation to the nutrients observed and have a higher concentration of Mn, K and Ca. The use of poultry litter as a digestate is economically desirable, since it represents an internal resource of the rural property. It is a waste that contains a high concentration of nutrients; however, it should be considered that fertilizers with higher proportions of organic nutrients in the organic medium and high lignin and fiber contents have lower decomposition rate in the soil and therefore lower release and availability of nutrients to the plants (Tessaro *et al.*, 2015; Nicoloso *et al.*, 2019).

## **4. CONCLUSIONS**

Comparing bovine and swine inocula, it was realized that both could be used in poultry litter digestion because of their stability, as shown by the ratios VA/TA and IA/PA, as well as by the pH values close to neutrality.

Relatively to the TS and VS removals efficiencies, the greatest mean values (59.46 and 78.21%, respectively) occurred with the swine inoculum, at the 0.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> load and with the bovine inoculum at the 1.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> load.

It was observed that the greatest mean values of  $COD_{total}$  removals referred to the 0.67 gVS.L<sup>-1</sup>.day<sup>-1</sup> load, with the swine inoculum and 1.00 gVS.L<sup>-1</sup>.day<sup>-1</sup>, with the bovine inoculum.

Regarding  $COD_{soluble}$  removal, the greatest mean values were reached at the 1.00 gVS.L<sup>-1</sup>.day<sup>-1</sup> load, being 61.31% and 53.29% for bovine and swine inoculum, respectively.

It is worth mentioning that the choice inoculum depends on the main purpose process (biogas or biofertilizer production). Therefore, in this case, where the goal was biofertilizer production, the use of swine inoculum is recommended, because it resulted in the best TS and COD removals and higher potassium (K) and calcium (Ca) levels in the generated sludge.

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