Chapter 1

Survey About the Use of Bacterial Inoculants in Brazil: Effects on Silage Quality and Animal Performance

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Additional information is available at the end of the chapter

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Abstract

Our objective was to report the effect of bacterial inoculants on silage quality and animal responses in Brazil. A survey of bacterial inoculants utilization in Brazil was made based on a total of 178 published articles assessing a widely varied crops (alfalfa, cabbage, cassava, corn, grass, high-moisture corn (HMC), high-moisture sorghum, millet, oat, orange bagasse, peanut forage, sorghum, soybean, stylosantes Campo Grande, sugarcane, and sunflower). Sugarcane and grass silages comprised 58.1% of the total crops investigated. Homolactic inoculation reduced dry matter (DM) losses in alfalfa silages, but not in corn, grass, HMC, and sorghum silages. Heterolactic inoculation enhanced the aerobic stability of corn and HMC silages. The use of heterofermentative lactic acid-bacteria (LAB) was more effective to improve fermentation of sugarcane silages compared to homofermentative LAB. Inoculation impaired the DM intake in cattle fed corn, grass, and sugarcane silages, but DM intake increased in sheep due to inoculation. In some cases, silage digestibility was affected by inoculation. Positive responses to inoculation occurred most often when the compatibility between the bacterial inoculant and crop was better understood (e.g., homolactic inoculation for grass silage and heterolactic inoculation for sugarcane silage). The performance of animals consuming inoculated silages has been investigated in Brazil only a few times, but the data suggest a greater impact of bacterial inoculants on DM intake and weight gain in cattle and sheep than that indicated in temperate conditions.

Keywords: aerobic stability, digestibility, fermentation, growth performance, lactic acid bacteria

1. Introduction

Silage is the feedstuff produced by the fermentation of a crop, forage, or agricultural byproduct, usually at greater than 50% moisture content [1]. In Brazil, silage is the principal source of energy and fiber in the diets of dairy cattle [2] and is frequently used in feedlots for the production of beef cattle [3]. However, descriptions of silage production practices and utilization in Brazilian literature are poor [4]. Furthermore, there is a lack of extension programs in Brazil that disseminate and enhance the knowledge of farmers regarding silage management, which has contributed to the production of low-quality products in many cases. As a strategy to alter this scenario, several farmers have chosen to use bacterial inoculants in order to improve silage quality and reduce production costs by decreasing the loss of dry matter (DM). Nevertheless, in Brazil, there are few reviews and surveys concerning the impact of bacterial inoculants on ensiling practices. In addition, the most complete review of this topic (see [5]) indicated that the low number of studies conducted in Brazil at that time did not produce a definitive conclusion about the magnitude of the effect of additives on silage quality and animal performance.

Therefore, our objective was to conduct a survey on the use of bacterial inoculants in Brazil and understand how they affect ensiling processes and animal performance. Here, we highlight that the present survey had an exploratory focus and, because of this, we conducted only a descriptive analysis of the data found in the accessed studies throughout of this text.

2. Bacterial inoculants

Ensiling is the most common method used to preserve a great variety of forages for use during those seasons when the crop is unavailable and/or is decreasing in nutritive value. Ensiling is based on the conversion of simple plant sugars, such as glucose and fructose, to lactic acid by lactic acid bacteria (LAB) under anaerobic conditions [6, 7]. Epiphytic LAB are essential microflora for spontaneous silage fermentation; however, the number and genera of bacteria varies widely in forages [8]. Thus, bacterial inoculants (specifically homofermentative LAB-^{ho}LAB) have been used in order (1) to inhibit the growth of aerobic and undesirable anaerobic microorganisms, (2) promote a rapid decline in the pH of forage after ensiling in order to avoid greater activity of proteases and deaminases derived from its own plant tissues and/or microorganisms, and (3) increase DM recovery [9].

The international literature is rich with data describing the eventual benefits of inoculation. However, no conclusion has been reached about the effect of bacterial inoculants on silage quality and animal performance in Brazil (see [5]) considering previously summarized studies carried out from 1999 to 2009. After 2009, 85 new Brazilian studies (scientific articles published in national and international journals) evaluating the effect of bacterial inoculants for silage production were published (**Figure 1**). Thus, analyzing real life scenarios are important to understand how bacterial inoculants alter silage quality and how they affect the performance of animals consuming inoculated silages.

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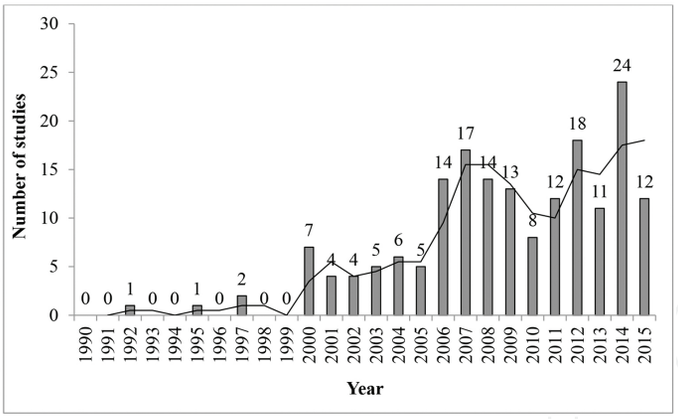

Figure 1. Number of Brazilian articles published concerning bacterial inoculant utilization from the last 26 years (total number of articles accessed = 178).

Initially, the small interest on the topic in the last century in Brazil likely reflected questions about the cost of those inoculants and their effectiveness as in other countries [10], although these are questions that are debated very often. The inconsistent results obtained from early studies carried out in Europe and North America due to low rates of inoculation and questionable viabilities of the bacteria [9], also likely contributed to the initial small interest. Conversely, advances in molecular biology associated with positive responses found across the world may have moved the crescent interest from Brazilian researchers to study bacterial inoculants for silage production. Moreover, the increasing number of techniques used to produce more viable and stable bacteria, and the additional tools developed to access the effects of silage inoculants, may also be part of the reason for the increased interest. Indeed, poor silage management has led to the production of silages of low nutritional value and undesirable sanitary aspects under tropical conditions. Surely, sugarcane and tropical grass silages are still the crops most susceptible to problems that occur during fermentation due to the action of undesirable microorganisms. Thus, these crops comprised 58.1% of all studies evaluated regarding the use of bacterial inoculants (**Figure 2**).

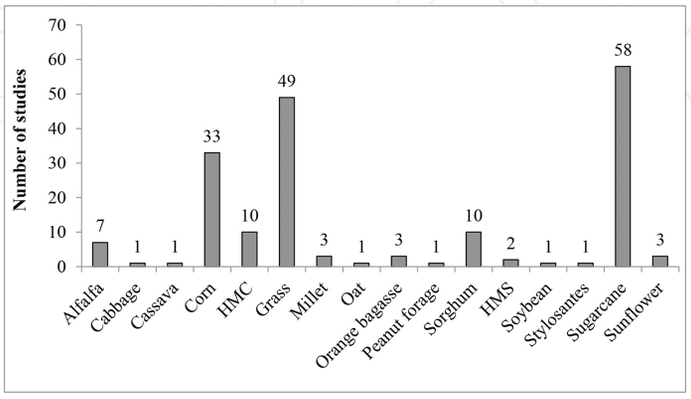

Figure 2. Number of Brazilian studies published regarding the utilization of silage inoculants by crop. *HMC, high-moisture corn; HMS, high-moisture sorghum.

Item	Alfalfa	Corn	Grass	HMC[1]	Sorghum	Sugarcane
One specie						
Bacillus subtilis	–	4	–	–	–	–
Lactobacillus brevis	–	–	–	–	–	27
Lactobacillus buchneri	–	16	8	8	–	62
Lactobacillus hilgardii	–	–	–	–	–	10
Lactobacillus kefiri	–	–	–	–	–	1
Lactobacillus paracasei	–	–	–	–	–	2
Lactobacillus plantarum	–	8	18	9	–	59
Leuconostoc mesenteroides	–	1	–	–	–	–
Streptococcus bovis	–	–	14	–	–	–
Streptococcus faecium	–	–	3	–	–	–
Two species						
L. buchneri + L. kefiri	–	–	–	–	–	1
L. buchneri + Propionibacterium acidipropionici	–	–	–	1	–	–
Lactobacillus casei + Streptococcus faecalis	–	–	–	2	–	–
L. plantarum + B. subtilis	–	1	–	–	–	–
L. plantarum + L. buchneri	–	4	–	–	–	1
L. plantarum + Pediococcus acidilactici	–	6	17	–	–	1
L. plantarum + Pediococcus pentosaceus	5	16	3	–	–	7
L. plantarum + P. acidipropionici	–	2	2	–	–	4
L. plantarum + S. faecium	6	14	12	–	26	3
Combo[2]	4	46	42	16	16	3

[1]HMC, high-moisture corn.
[2]Combination of three or more bacteria.

Table 1. Bacterial species applied in the six main crops used to produce silage in Brazil (number of treatments).

As mentioned earlier, the crescent development in molecular biology techniques has led to a wide range of microbial additives to aid in crop preservation. The LAB (genera *Lactobacillus*, *Pediococcus*, *Lactococcus*, *Enterococcus, Streptococcus*, and *Leuconostoc*) are the main group of bacteria used as silage inoculants, because they all produce lactic acid as a principal product from sugar fermentation [6]. Commonly, the LAB are classified into two groups based on the products of fermenting glucose, as follow: (1) homofermentative (first generation of silage inoculants) → produce two moles of lactic acid from one mole of glucose; and (2) heterofermentative (second generation of silage inoculants) → produce one mole of lactic acid, one mole of carbon dioxide (CO_2), and either one mole of ethanol or one mole of acetic acid from one mole of glucose [11]. However, actually three groups of LAB have been considered [12], as

follows: (1) obligate homofermentative → unable to ferment pentoses because the lack enzyme phosphoketolase; (2) facultative heterofermentative → ferment hexoses similarly to the obligate homofermentative but they are able to ferment pentoses; and (3) obligate heterofermentative → ferment hexoses to a range of products. Overall, under most silage conditions where substrate is not lacking, facultative heterofermentative LAB primarily make only lactic acid [9]. Thus, for the sake of simplicity, facultative heterofermentative LAB will be considered part of homofermentative LAB in this review for furthers comparison.

In Brazil, several homofermentative (*Lactobacillus plantarum*, *L. curvatus*, *L. acidophilus*, *L. paracasei*, *Lactococcus lactis*, *Leuconostoc mesenteroides*, *Pediococcus pentosaceus*, *P. acidilactici*, *Streptococcus faecium*, *S. faecalis*, and *S. bovis*) and heterofermentative LAB (*L. buchneri*, *L. hilgardii*, *L. kefiri*, *L. salivarius*, and *L. brevis*) have been used as silage inoculants, leading to different combinations for each crop (**Table 1**). Other microorganisms have also been tested, such as *Propionibacterium acidipropionici*, *Bacillus subtilis*, and *Saccharomyces* spp., but less frequently.

As described earlier, ^{ho}LAB and heterofermentative LAB (^{he}LAB) comprised first and second generation of silage inoculants, respectively. The ^{ho}LAB gained popularity in the late 1970s and early 1980s because it must quickly grow to dominate silage fermentation reducing DM and nutritive losses [9]. Conversely, homofermentative-inoculated silages often have lower stability during the feed-out phase, because of the greater concentration of lactic acid and residual water-soluble carbohydrates (WSC) [13]. Lactic acid and WSCs are utilized as substrates for the growth of aerobic microorganisms, notably yeasts [13]. Thus, *L. buchneri* was developed as a second generation inoculant to produce acetic acid and improve the aerobic stability of silage by inhibiting the growth of spoilage microorganisms [14]. Nowadays, some commercial silage inoculants contain multiple strains of ^{ho}LAB and often one strain of ^{he}LAB, because of the potential synergistic actions among bacterial strains. For example, previous studies showed that the association between *L. plantarum* and *L. buchneri* accelerated the initial rate of lactic acid fermentation, reducing the pH and causing lower protein degradation, in addition to enhancing the aerobic stability of corn and sorghum silages [13, 15].

In Brazil, ^{ho}LAB were primarily investigated and used as commercial silage inoculants to ensure suitable fermentation (**Figure 3**). Around the year 2000, Brazilian researchers turned their attention and curiosity to investigate the effects of ^{he}LAB on the ensiling of tropical crops, but articles on this topic only started to be published in 2006. Moreover, studies combining ^{ho}LAB and ^{he}LAB started at the same time that second generation silage inoculants were used, but articles evaluating ^{ho}LAB and ^{he}LAB combined started to be published earlier.

Despite the type of silage inoculant used for the six main crops used for ensiling in Brazil, ^{ho}LAB composed the only silage inoculant assessed for alfalfa and sorghum silages (**Figure 4**). Moreover, ^{ho}LAB still composed the majority (>69%) of the treatments for corn, HMC, and grass silages. Sugarcane was the only crop in which ^{he}LAB composed the majority (57%) of the treatments assessed. This scenario is not a surprise, since ^{ho}LAB were primarily investigated and used as commercial silage inoculants in the worldwide, and likely this reflected in a greater number of studies assessing ^{ho}LAB in Brazil. Alfalfa and grass silages often have low WSC content and high buffer capacity, and then pH declines more slowly after the crop is ensiled

[6]. Therefore, is comprehensive why only ^{ho}LAB were assessed for alfalfa and why ^{ho}LAB composed the majority of the treatments for grass. However, considering that corn and sorghum silages that are most susceptible to aerobic deterioration under tropical conditions [16] would be expected a greater number of studies concerning ^{he}LAB or combining ^{ho}LAB and ^{he}LAB to reduce this trouble.

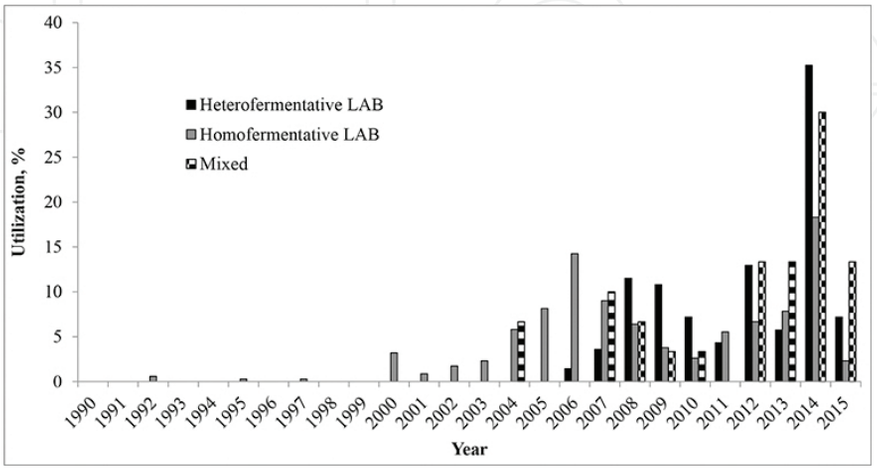

Figure 3. Evolution of the utilization of homofermentative and heterofermentative LAB, either alone or combined (mixed) in Brazil (% related to the number of treatments).

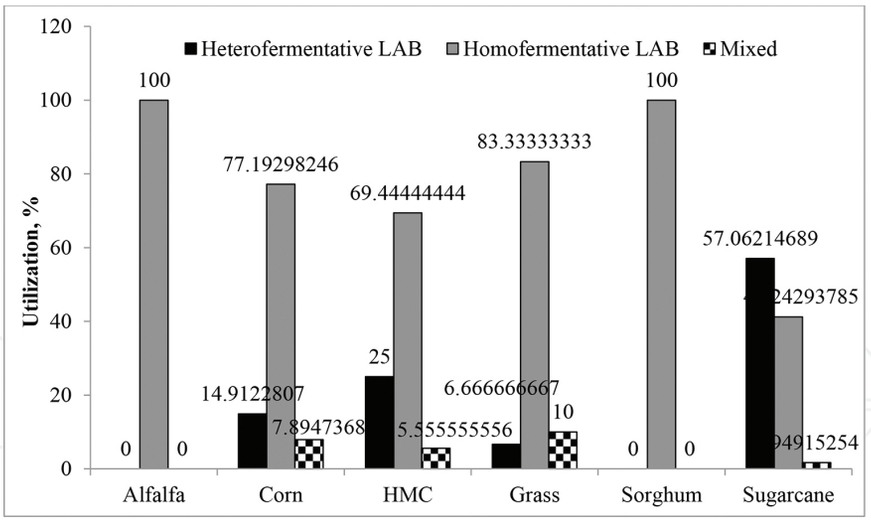

Figure 4. Assessment of homofermentative and heterofermentative LAB, either alone or combined (mixed) by crop in Brazil (% related to the number of treatments containing bacterial inoculants). *HMC, high-moisture corn.

The use of bacterial inoculants has also claimed to improve the nutritive value of silages by reflecting alterations in fermentation patterns, which may be important for tropical silages in particular. The use of tropical forages often results in silages with lower nutritive value than those produced under temperate conditions [16]. Unfavorable aspects of some crops (especially grasses), such as low WSC and DM content (both needed for proper fermentation) at the

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time of cutting when the highest nutritive value of the grass is achieved and at high buffering capacity, results in poor fermentation and low silage digestibility [17].

Epiphytic LAB utilize carbohydrates as energy and carbon sources for growth, and these microorganisms are only able to convert nonstructural carbohydrates (notably WSCs—mono- and disaccharides) into organic acids, because they lack the enzymatic complex required to metabolize complex polysaccharides [7]. Thus, enzyme-bacterial inoculants may become useful to improve the fermentation patterns and nutritive value mainly of ensiled crops having low WSC content. Bacterial inoculants ensure that LAB will dominate in silage fermentation, whereas the enzymes (i.e., fibrolytic enzymes) contained in those inoculants act on the cell wall, releasing a greater amount of fermentable sugars and increasing substrate availability, thereby improving silage digestibility [18]. Amylolytic and proteolytic enzymes are also commonly used in silage inoculants, and they are particularly useful for cereal silages, reducing the negative effect of the starch-protein matrix on starch digestion in ruminants [19, 20]. Therefore, it is easy to understand why enzyme-bacterial inoculants are used primarily in high-moisture corn (HMC) silages (>55%), followed by grass, corn, sorghum, alfalfa, and sugarcane silages (**Figure 5**). Obviously, the little interest in evaluating enzyme-bacterial inoculants for sugarcane silage is related to the great amount of WSC in this crop, particularly sucrose [21].

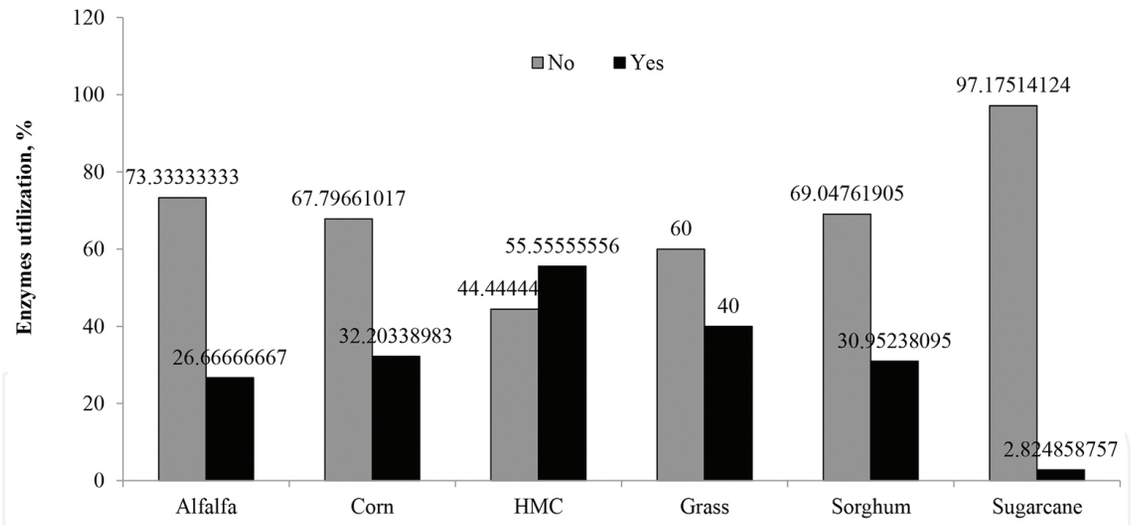

Figure 5. Enzyme utilization in silage inoculants by crop in Brazil (% related to the number of treatments containing bacterial inoculants). *HMC, high-moisture corn.

2.1. Fermentation patterns, nutritive value, and aerobic stability of silages

The use of bacterial inoculants as additives to improve silage fermentation has a long and diverse history. As described earlier, although silage inoculant utilization occurred later in Brazil than Europe and North America, many types and formulations of bacteria are currently sold commercially for this purpose. However, the compatibility between the plant and

microorganisms used will determine the success of the application of bacterial inoculants in silages [22]. When that compatibility is better understood, positive responses from inoculation occur more often.

Fermentation and microbiological profile	CCP	Chemical composition	CCP	Animal performance	CCP
pH	Decreasing	DM, % as fed	Increasing	DMI, kg/day	Increasing
Ammonia-N, % TN	Decreasing	Ash	Decreasing	DMI, % BW	Increasing
WSC	Increasing	EE	Increasing	OMI, kg/day	Increasing
Lactic acid	Increasing	CP	Increasing	NDFI, kg/day	Increasing
Acetic acid	Increasing (^{he}LAB) and decreasing (^{ho}LAB)	NDIN, % N	Decreasing	CPI, kg/day	Increasing
Propionic acid	Increasing	ADIN, % N	Decreasing	Digestible DMI, kg/day	Increasing
Butyric acid	Decreasing	NDF	Decreasing	Digestible OMI, kg/day	Increasing
Total acids[2]	Increasing	ADF	Decreasing	Digestible NDFI, kg/day	Increasing
Lactic:acetic acid	Increasing (^{ho}LAB) and decreasing (^{he}LAB)	Hemicellulose	Decreasing	Digestible CPI, kg/day	Increasing
Ethanol	Decreasing	Cellulose	Decreasing	DM digestibility	Increasing
Total acids:ethanol	Increasing	Lignin	Decreasing	OM digestibility	Increasing
Effluent, kg/t of fresh matter	Decreasing	IVDMD	Increasing	NDF digestibility	Increasing
Gas losses	Decreasing	IVOMD	Increasing	CP digestibility	Increasing
DM losses	Decreasing			Feed efficiency[3]	Decreasing
LAB, log cfu/g of fresh silage	Increasing			ADG, kg/day	Increasing
Yeasts, log cfu/g of fresh silage	Decreasing				
Molds, log cfu/g of fresh silage	Decreasing				
Aerobic stability, h	Increasing				
Maximum temperature, °C	Decreasing				

TN, total nitrogen; WSC, water-soluble carbohydrates; DM, dry matter; LAB, lactic acid bacteria; ^{he}LAB, heterofermentative LAB; ^{ho}LAB, homofermentative LAB; EE, ether extract; CP, crude protein; NDIN, neutral detergent insoluble N; ADIN, acid detergent insoluble N; NDF, neutral detergent fiber; ADF, acid detergent fiber; IVDMD, *in vitro* DM digestibility; IVOMD, *in vitro* organic matter digestibility; DMI, DM intake; BW, body weight; OMI, organic matter intake; NDFI, NDF intake; CPI, CP intake; ADG, average daily gain.
[1]Adapted from [5].
[2]Total acid content was calculated as the sum of lactic, acetic, and propionic acids.
[3]Feed efficiency was determined by dividing DMI by ADG.

Table 2. Criteria considered as positive (CCP) effect of inoculation for each variable (data are % of DM, unless otherwise stated)[1].

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In order to understand the extent to which each type of bacterial inoculant affects silage quality, we summarized data from corn, grass, sugarcane, alfalfa, sorghum, and HMC silages produced in Brazil. All comparisons in this survey were made from studies (at least two studies for each variable) that used a negative treatment (untreated forage—control) against one or more treatments containing bacterial inoculants. Some calculations were made when data were lacking from these publications as follows: hemicellulose content was calculated as neutral detergent fiber (NDF) minus acid detergent fiber (ADF), whereas cellulose content was calculated as ADF minus lignin; the proportion of hemicellulose, cellulose, and lignin were also calculated on a NDF basis; total acid production was calculated as the sum of lactic, acetic, and propionic acids; and the ratio of lactic:acetic acid and total acid:ethanol was also calculated. Butyric acid was not considered in the calculation of total acids because this acid has no beneficial effect on ensiling process [6]. Otherwise, lactic acid (acid more desired to reduce DM loss) and acetic and propionic acids (antifungal properties) have beneficial role during ensiling [6].

As described earlier, we performed only a descriptive analysis of data found in the studies investigated. For that, we did not consider a minimum or maximum time of fermentation to include the data from each study in the final dataset, because our objective was not to show the fermentation pattern regarding the length of ensiling. From the summarized data, the mean, median, standard deviation, and minimum and maximum values were calculated for all variables. Moreover, the frequency of positive responses from inoculation was also calculated, considering only the means declared statistically different in the studies that comprised the database. The difference between the means of untreated and inoculated silages, when there were positive responses, was also calculated. The criteria considered as positive for each variable are given in **Table 2**.

Enterobacteria count was not considered in this survey by lack of data, but it is important to state that enterobacteria are the principal competitors against LAB for sugars after the crop is ensiled, and acetic acid is the principal product of enterobacterial fermentation [8]. Conversely, enterobacteria population often declines after ensiling by influence of anaerobiosis and pH reduction due to the acids produced during fermentation [8].

2.1.1. Corn silage

Data were summarized from a total of 29 studies, of which 19, 7, and 7 investigated the effect of ^{ho}LAB, ^{he}LAB, and a combination between both (mixed), respectively. *Bacillus subtilis* was also investigated in two studies. Considering all treatments, the application rate of bacterial inoculants ranged from 5×10^4 to 1×10^9 colony forming units (cfu)/g of fresh forage.

The ranges of fermentation patterns, *in vitro* digestibility, and aerobic stability are given in **Table 3**. Considering the overall mean, lactic acid and silage pH were unaffected by ^{ho}LAB. The concentration of lactic acid was greater by 51.2% when both ^{ho}LAB and ^{he}LAB were applied than observed in untreated silage. The ^{ho}LAB increased by 12.8% the concentration of acid detergent insoluble N (ADIN), suggesting that the temperature of fermentation also increased following inoculation. In addition, ^{ho}LAB slightly reduced (−2.8%) the *in vitro* DM digestibility (IVDMD) of corn silages.

Item[1]	Untreated						Homofermentative LAB						Heterofermentative LAB						Mixed[4]					
	n[2]	Mean	Median	SD[3]	Min	Max	n	Mean	Median	SD	Min	Max	n	Mean	Median	SD	Min	Max	n	Mean	Median	SD	Min	Max
DMoven, % as fed	50	32.99	32.62	3.75	23.07	43.70	67	34.57	34.21	4.16	23.03	48.34	20	32.96	34.05	2.47	25.80	36.71	8	29.73	29.89	3.09	22.95	35.12
Ash	31	4.79	4.20	1.35	3.20	12.68	37	5.27	4.24	1.86	2.71	13.23	8	4.11	4.20	0.28	3.00	4.49	6	3.82	3.83	0.61	3.10	4.52
CP	47	7.22	7.22	0.83	5.29	9.70	64	7.33	7.18	0.75	3.90	10.20	17	7.97	7.50	0.98	6.59	9.43	8	7.43	7.75	0.91	5.50	8.90
NDIN, % N	2	12.64	12.64	12.34	0.30	24.97	2	1.84	1.84	1.37	0.47	3.20	1	-	-	-	-	-	1	-	-	-	-	-
ADIN, % N	4	18.84	20.78	4.61	9.62	24.17	7	21.24	19.55	3.18	17.33	27.00	1	-	-	-	-	-	2	12.89	12.89	3.47	9.42	16.35
NDF	45	53.29	53.38	4.71	38.80	64.93	60	54.48	54.17	5.12	40.00	66.47	17	49.88	49.20	6.08	38.15	62.00	8	51.19	50.11	4.45	39.30	59.50
ADF	25	30.32	29.48	3.75	22.57	39.13	24	31.56	30.20	4.58	23.20	42.52	9	26.60	25.80	2.40	21.80	33.99	8	29.99	30.00	2.99	22.60	36.87
Hemicellulose	25	22.78	22.95	2.78	16.20	31.80	24	24.26	23.86	2.69	17.96	31.37	9	18.63	18.50	1.53	16.30	21.24	8	21.43	19.82	3.21	16.70	32.10
Cellulose	14	25.01	25.30	2.54	17.30	28.93	15	25.23	25.27	2.50	18.50	29.59	4	23.11	22.98	1.74	19.90	26.58	6	24.26	24.98	1.98	19.20	27.04
Lignin	13	6.01	4.90	2.05	2.81	13.05	13	8.38	7.50	3.44	2.53	13.11	4	4.63	4.10	1.39	2.90	7.41	6	4.71	4.54	0.90	2.70	7.41
IVDMD	15	61.17	65.23	7.02	46.40	70.67	19	59.43	65.00	9.75	46.14	71.57	2	69.73	69.73	1.53	68.20	71.26	5	63.96	64.93	5.64	49.87	71.68
IVOMD	6	69.78	68.90	4.62	64.30	78.20	10	71.25	71.35	3.65	64.50	78.20	1	-	-	-	-	-	1	-	-	-	-	-
Effluent, kg/t	7	12.25	10.08	7.21	1.74	28.72	12	12.85	12.86	8.56	1.47	35.43	3	4.83	4.86	0.43	4.19	5.45	0	-	-	-	-	-
Gas losses	3	2.62	2.89	1.72	0.04	4.93	2	0.03	0.03	0.01	0.03	0.04	7	4.18	4.80	1.36	2.27	5.88	0	-	-	-	-	-
DM losses	12	4.74	5.60	2.74	0.55	9.21	13	4.80	5.20	1.27	1.37	6.21	12	6.21	5.53	2.06	2.50	14.01	3	5.25	3.92	3.26	1.70	10.14
1.2-propanediol	3	0.35	0.29	0.27	0.00	0.75	0	-	-	-	-	-	5	0.60	0.46	0.35	0.20	1.26	0	-	-	-	-	-
Lactic acid	13	4.93	4.30	1.69	1.47	7.97	12	4.69	5.08	1.66	1.00	8.01	20	4.90	4.78	1.16	2.17	6.88	5	7.46	7.89	1.64	4.46	9.30
Acetic acid	13	2.46	1.91	1.63	0.33	12.48	12	1.41	1.24	0.77	0.46	2.83	20	2.01	1.18	1.53	0.58	12.57	5	6.66	5.50	4.49	1.50	17.89
Propionic acid	8	0.26	0.05	0.30	0.01	1.46	12	0.23	0.02	0.25	0.01	0.62	13	0.36	0.20	0.31	0.01	1.33	1	-	-	-	-	-
Butyric acid	7	0.06	0.04	0.05	0.00	0.19	12	0.04	0.01	0.05	0.00	0.22	11	0.11	0.07	0.06	0.02	0.22	1	-	-	-	-	-
Total acids[5]	7	6.69	6.65	2.12	3.56	10.15	12	6.33	6.26	1.34	3.58	10.13	11	6.99	7.79	1.26	4.25	8.42	1	-	-	-	-	-
Ethanol	7	0.86	0.70	0.52	0.01	1.84	12	1.62	0.50	1.57	0.01	4.24	9	1.75	1.54	0.72	0.69	2.75	1	-	-	-	-	-
Lactic:acetic acid	13	4.45	3.33	2.97	0.34	14.89	12	5.77	4.26	3.83	0.39	10.90	20	4.07	3.84	1.65	0.34	8.15	5	1.90	1.69	0.79	0.36	2.97
Total acids:ethanol	5	149.73	9.50	227.36	1.93	718.13	11	122.12	12.17	177.89	1.81	674.85	7	4.96	2.16	3.34	1.84	10.81	0	-	-	-	-	-
pH	26	3.84	3.79	0.17	3.42	4.30	28	3.89	3.94	0.20	3.30	4.38	21	3.92	3.91	0.13	3.67	4.16	6	3.88	3.94	0.19	3.53	4.10
Ammonia-N, % TN	22	7.77	4.98	5.18	0.11	30.36	25	6.93	4.43	4.14	1.46	25.72	15	6.39	5.99	2.50	0.09	11.20	4	6.03	5.08	3.12	2.90	11.07
LAB, log cfu/g	9	7.57	8.43	1.33	5.45	9.14	12	8.07	8.41	0.66	6.68	9.28	5	6.85	6.64	0.61	5.88	7.75	0	-	-	-	-	-
Yeasts, log cfu/g	12	5.51	5.25	0.96	4.00	7.67	12	6.02	6.07	1.15	3.00	8.83	10	2.95	2.94	1.25	1.20	4.84	3	4.04	3.79	0.53	3.50	4.83
Molds, log cfu/g	7	3.32	3.70	1.40	1.07	5.69	0	-	-	-	-	-	14	2.95	2.80	1.08	0.00	5.14	2	4.64	4.64	0.44	4.20	5.07
Aerobic stability, h	10	36.49	32.90	21.77	0.30	92.20	3	31.08	33.00	7.89	19.25	41.00	12	109.87	89.95	51.58	26.91	228.00	5	27.65	12.08	23.76	0.20	60.50
Maximum T, ºC	7	29.97	27.75	3.51	26.00	42.00	13	29.32	27.75	2.54	25.25	38.00	0	-	-	-	-	-	1	-	-	-	-	-

[1]DM, dry matter; CP, crude protein; NDIN, neutral detergent insoluble N; ADIN, acid detergent insoluble N; NDF, neutral detergent fiber; ADF, acid detergent fiber; IVDMD, *in vitro* DM digestibility; IVOMD, *in vitro* organic matter digestibility; TN, total nitrogen; LAB, lactic acid bacteria.
[2]Number of means.
[3]Standard deviation.
[4]Silages inoculated with both heterofermentative and homofermentative bacteria.
[5]Total acid content was calculated as the sum of lactic, acetic, and propionic acids.

Table 3. Range of fermentation patterns, nutritive value, and aerobic stability of untreated and inoculated corn silages (data are given in % of DM, unless otherwise stated).

All silages were close to or inside the ideal range of the DM content (30–35% of DM) recommended for the production of corn silage [6]. Under these conditions, corn plants often exhibit a great amount of WSC and have a low buffer capacity since well managed. Thus, the lack of positive results from homolactic inoculation is likely related to the desired characteristics of corn plants used at ensiling, once all silages (including the untreated) produced a suitable quantity of lactic acid, with an ideal range between 4 and 7% of the DM [23].

Overall, although positive responses from ^{ho}LAB inoculation were not observed, ^{ho}LAB might be useful to increase lactic acid production and improve fermentation when silage is produced with corn plants harvested with moderately to high DM content (i.e., >37%), because a lack of moisture in dry forages restricts the overall fermentation process [6]. Furthermore, the quality of corn silage produced under tropical conditions is not properly a problem, even though its

quality often is lower than that produced under temperate conditions [16]. The main problem of corn silage produced under tropical conditions is related with aerobic deterioration [16] when the silos are opened. The elevated temperature occurring in tropical weather is favorable to yeasts' overgrowth [24], which initiates the spoilage of silages by using residual WSC and lactic acid as substrate to growth, with consequent reduction in the nutritive value of silages. In this regard, ^{he}LAB should be useful to reduce aerobic deterioration of corn silages, but in general, ^{ho}LAB composed 77.2% of all treatments concerning silage inoculants for corn silage. The greater ^{ho}LAB utilization likely still reflects the fact that homolactic inoculants were primarily developed as silage additives, and commercial products based on ^{ho}LAB are most available to be assessed compared with ^{he}LAB.

Despite heterolactic inoculation, acetic acid was unaffected, but the aerobic stability of silages was enhanced (+73.4 h), likely because of reductions in the number of yeasts. Nevertheless, ^{he}LAB increased ethanol production, gas, and DM losses during fermentation by 103.6, 59.7, and 31.2% compared with untreated silages, respectively. Extensive heterolactic fermentation unavoidably increases DM loss during the time the silo is closed, because additional products (i.e., acetic acid, ethanol, and CO_2) are formed besides lactic acid [11]. Furthermore, the concentration of 1,2-propanediol increased 116.7% in silages inoculated with ^{he}LAB. *L. buchneri* comprised the main ^{he}LAB evaluated in corn silage, and this bacterium is able to produce 1,2-propanediol, coupled with acetic acid, during anaerobic degradation of lactic acid [25]. The ammonia-N concentration of silages inoculated with ^{ho}LAB and ^{he}LAB, either alone or combined, is in agreement with well-fermented corn silages (range from 5 to 7%) [23].

Considering the overall means, ^{he}LAB reduced the NDF content of silages by 6.4%. In many cases, the reductions in NDF content have been attributed to the capacity of *L. buchneri* to produce ferulate esterase, an enzyme that acts on cell wall-releasing ferulic acid [27]. However, only some specific strains of *L. buchneri* have the capacity to produce ferulate esterase [26]. Moreover, a net hydrolysis of hemicellulose did not occur when the values were compared on an NDF basis (C.H.S. Rabelo and R.A. Reis). Thus, the reasons for reduced NDF content of corn silages inoculated with ^{he}LAB are still unclear; once DM loss increased, it did not provide better preservation of WSC, which could decrease NDF content by the concentration effect. Heterolactic inoculation also improved IVDMD by 14%, most probably due to a reduction in NDF content.

Although few studies combining ^{ho}LAB and ^{he}LAB were carried out in Brazil, overall means revealed increased lactic and acetic concentration when both inoculants were applied on corn silage compared to untreated silage. Combining ^{ho}LAB and ^{he}LAB may ensure a better fermentation process of corn silage with increased lactic and acetic acid concentration [13], as reported earlier. Consequently, a reduction in DM losses with an increased aerobic stability should be expected, but was not observed. Otherwise, silages treated with both ^{ho}LAB and ^{he}LAB slightly lowered NDF content and increased IVDMD. Even though the data of this survey about combining ^{ho}LAB and ^{he}LAB are not encouraged, most likely due to the low number of studies, further researches should consider the investigation of both ^{ho}LAB and ^{he}LAB for corn silage. The international literature has found a better fermentation process of

corn silage accompanied of a greater aerobic stability when [ho]LAB and [he]LAB were simultaneously used [13, 14, 15]. These responses may ensure most suitable nutritive value of silage and lead some beneficial on animal response.

The frequency and difference of positive responses found in corn silages from homolactic and heterolactic inoculations are given in **Table 4**. Considering only homolactic inoculation, the greatest frequency of positive responses occurred for aerobic stability, lactic acid, DM content, IVDMD, number of yeasts, and IVOMD. Furthermore, the greatest differences in response were observed for lactic acid, effluent production, and aerobic stability. The greater frequency of positive responses observed for aerobic stability is likely to be related to the low number of trials used to generate the data. According to **Table 3**, the average aerobic stability was greatest for [he]LAB among all treatments. Conversely, increases in the concentration of lactic acid, DM content, and IVDMD suggest better preservation of soluble sugars during ensiling. The [ho]LAB have been used to reduce variation in the ensiling process, usually by accelerating the post-ensiling decline in pH, while improving DM and nutrient retention [28].

Item[1]	Homofermentative LAB					Heterofermentative LAB				
	Number of treatments		Mean		Difference, %	Number of treatments		Mean		Difference, %
	Total	Positive responses, %	Untreated	Inoculated		Total	Positive responses, %	Untreated	Inoculated	
DMoven, % as fed	67	38.8	36.54	38.05	+ 4.1	20	5.0	31.70	33.40	+ 5.4
Ash	37	10.8	4.65	3.96	- 14.8	8	0.0	-	-	-
CP	64	17.2	6.58	7.35	+ 11.7	17	5.9	8.30	9.20	+ 10.8
NDIN, % N	2	0.0	-	-	-	1	0.0	-	-	-
ADIN, % N	7	0.0	-	-	-	1	0.0	-	-	-
NDF	60	15.0	57.02	50.68	- 11.1	17	5.9	49.90	43.50	- 12.8
ADF	24	8.3	38.56	35.36	- 8.3	9	0.0	-	-	-
Hemicellulose	24	0.0	-	-	-	9	11.1	23.80	18.80	- 21.0
Cellulose	15	13.3	27.45	24.16	- 12.0	4	0.0	-	-	-
Lignin	13	0.0	-	-	-	4	25.0	4.10	2.90	- 29.3
IVDMD	19	36.8	62.31	66.97	+ 7.5	2	50.0	56.00	68.20	+ 21.8
IVOMD	10	30.0	64.80	76.60	+ 18.2	1	100.0	65.90	71.30	+ 8.2
Effluent, kg/t	12	8.3	28.72	14.25	- 50.4	3	0.0	-	-	-
Gas losses	2	0.0	-	-	-	7	0.0	-	-	-
DM losses	13	30.8	7.95	6.19	- 22.2	12	0.0	-	-	-
WSC	1	0.0	-	-	-	0	0.0	-	-	-
1.2-propanediol	0	0.0	-	-	-	5	60.0	0.15	0.84	+ 479.8
Lactic acid	12	41.7	2.97	5.72	+ 92.3	21	14.3	2.97	4.42	+ 48.6
Acetic acid	12	0.0	-	-	-	21	47.6	1.02	1.40	+ 37.4
Propionic acid	12	0.0	-	-	-	13	0.0	-	-	-
Butyric acid	12	0.0	-	-	-	12	0.0	-	-	-
Ethanol	12	0.0	-	-	-	9	0.0	-	-	-
Lactic: acetic acid	12	0.0	-	-	-	21	4.8	5.00	3.00	- 40.0
pH	28	10.7	3.63	3.44	- 5.1	21	0.0	-	-	-
Ammonia-N, % TN	25	16.0	16.21	13.44	- 17.1	16	0.0	-	-	-
LAB, log cfu/g	12	8.3	6.80	7.41	+ 9.0	5	60.0	5.74	7.25	+ 26.4
Yeasts, log cfu/g	12	33.3	6.10	5.04	- 17.4	10	50.0	4.35	1.70	- 60.9
Molds, log cfu/g	0	0.0	-	-	-	14	7.1	1.07	0.00	- 100.0
Aerobic stability, h	3	66.7	25.00	37.00	+ 48.0	12	75.0	49.86	130.66	+ 162.0

[1]DM, dry matter; CP, crude protein; NDIN, neutral detergent insoluble N; ADIN, acid detergent insoluble N; NDF, neutral detergent fiber; ADF, acid detergent fiber; IVDMD, *in vitro* DM digestibility; IVOMD, *in vitro* organic matter digestibility; TN, total nitrogen; LAB, lactic-acid bacteria.

Table 4. Summary of positive responses of silage inoculants on the fermentation patterns, nutritive value, and aerobic stability of corn silages (data are given in % of DM, unless otherwise stated).

Despite heterolactic inoculation, greater frequencies of positive responses were observed for IVOMD, aerobic stability, number of LAB and yeasts, IVDMD, and acetic acid. In addition, the greatest magnitudes of response were observed for the concentration of 1,2-propanediol, aerobic stability, and molds.

The low number of means for some variables contributed to large values for the frequency of positive responses, as well as the difference between untreated and inoculated silages. However, the data clearly showed that ^{he}LAB in corn silage, composed mainly of *L. buchneri*, were biologically effective. *L. buchneri* has been shown to enhance the aerobic stability of silages by increasing the production of acetic acid, which decreases the growth of spoilage microorganisms [29]. Acetic acid has antifungal characteristics [30], and heterolactic inoculation may be particularly important in silages produced under tropical conditions, as elevated temperatures are favorable for yeast growth [24].

2.1.2. Tropical grass silage

Data were summarized from a total of 45 studies, of which 40, 4, and 6 investigated the effect of ^{ho}LAB, ^{he}LAB, and a combination of both (mixed), respectively. In these studies, several tropical grasses were investigated: 18 studies with *Pennisetum purpureum* (Elephant grass cv. Napier and Cameroon), 12 studies with *Panicum maximum* (Guinea grass cv. Mombasa and Tanzania), 11 studies with *Brachiaria brizantha* (Palisadegrass cv. Marandu, Xaraes, and Piata), 3 studies with *Cynodon dactylon* (Bermudagrass), 2 studies with *Cynodon nlemfuensis* (Stargrass), and 1 study with *Brachiaria decumbens*. Considering all treatments, the application rate of silage inoculant ranged from 5×10^4 to 8×10^{10} cfu/g of fresh forage.

The range of fermentation patterns, *in vitro* digestibility, and aerobic stability are given in **Table 5**. Considering the overall mean, homolactic inoculation increased the concentration of lactic acid by 29.4%, leading to a pH drop from 4.75 (untreated silage) to 4.47. The main purpose to use ^{ho}LAB is ensuring a rapid pH decline in earlier times of fermentation (often the first 2 days of ensiling) because the greater production of lactic acid [6]. Indeed, pediococci, streptococci, and lactobacilli comprised the majority commercial homolactic inoculants investigated in Brazilian studies, and they lead to the rapid production of lactic acid and great sugar-to-lactic acid conversion efficiency [6]. Otherwise, after the stable phase of fermentation is reached, similar pH can be reported between untreated and inoculated silage with ^{ho}LAB [6]. The DM losses and ammonia-N concentration decreased 11.4 and 11.7%, respectively, due to the use of ^{ho}LAB. The reduction observed for ammonia-N is likely due to a rapid drop in pH, avoiding proteolysis by the plant, and the action of undesirable microorganisms, such as clostridia. Furthermore, the ADIN content decreased 15.1% due to homolactic inoculation. Results from the present survey agree with the international literature, wherein inoculation with ^{ho}LAB generally results in a faster rate of fermentation, less proteolysis, more lactic acid, less acetic and butyric acids, less ethanol, and a greater recovery of energy and organic matter(OM) [9]. Moreover, the data from this survey suggest that homolactic inoculation is most effective in tropical grass silages, compared to other crops. Homolactic inoculation was also most effective in improving the fermentation process of grass silages, compared with corn and sorghum silages in temperate climates [31]. The reasons for that are because the reduced

WSC concentration and epiphytic bacteria populations found prior to ensiling in those crop, which commits the ensiling process [31]. In our survey, although homolactic inoculation consistently improved the fermentation parameters of tropical grass silages, a small effect was observed on the nutritive characteristics, and IVDMD was only slightly improved (+1.5%).

In some cases, adding homolactic inoculants reduced the aerobic stability of silages, because the lactic acid they produce is used as a growth substrate by yeasts that initiate spoilage [32]. However, unexpectedly the aerobic stability of tropical grass silages increased from 59.5 to 114 h when ^{ho}LAB were applied at ensiling, which is likely to be due to the greater production of acids and a lower pH, inhibiting the growth of aerobic microorganisms. But this is only a hypothesis and perhaps factors other than fermentation end products likely contributed to increase the aerobic stability of grass silages treated with ^{ho}LAB.

Item[1]	Untreated						Homofermentative LAB						Heterofermentative LAB						Mixed[4]					
	n[2]	Mean	Median	SD[3]	Min	Max	n	Mean	Median	SD	Min	Max	n	Mean	Median	SD	Min	Max	n	Mean	Median	SD	Min	Max
DMoven, % as fed	49	23.91	21.50	5.73	14.02	54.90	78	24.19	22.20	5.97	14.13	50.00	3	24.33	27.45	4.22	18.00	27.54	10	23.94	23.17	2.78	18.20	29.70
Ash	17	10.82	11.00	1.80	6.99	16.20	24	10.89	10.75	1.59	6.92	20.60	0	-	-	-	-	-	6	9.53	10.07	0.97	7.10	10.48
CP	55	8.27	7.07	2.78	2.40	43.55	85	8.49	7.74	2.50	2.20	45.85	5	6.16	6.20	1.49	3.90	8.10	10	9.99	11.02	2.81	5.60	16.35
NDIN, % N	4	23.77	24.19	11.27	6.51	40.20	3	21.03	16.01	13.18	6.27	40.80	0	-	-	-	-	-	1	-	-	-	-	-
ADIN, % N	14	17.92	12.71	8.33	5.58	52.49	15	15.21	14.73	4.15	5.27	27.10	0	-	-	-	-	-	1	-	-	-	-	-
NDF	49	69.53	70.20	5.69	47.98	81.97	72	69.22	69.44	4.97	48.59	81.00	5	72.74	73.10	3.51	67.40	79.90	10	77.09	78.96	4.52	64.98	83.20
ADF	44	41.45	44.33	6.60	20.93	52.37	65	41.04	42.48	6.43	21.47	54.40	1	-	-	-	-	-	10	44.41	43.50	3.70	39.64	50.91
Hemicellulose	41	28.62	28.81	4.06	20.97	40.92	63	27.98	29.44	4.79	-0.25	38.88	1	-	-	-	-	-	10	32.68	32.19	3.53	25.34	38.18
Cellulose	21	38.47	38.20	3.80	26.50	45.80	29	38.59	37.65	2.89	32.76	45.70	0	-	-	-	-	-	8	33.03	30.84	5.38	27.10	43.79
Lignin	21	7.24	6.83	1.95	4.42	12.47	29	7.33	7.30	1.71	4.06	11.82	0	-	-	-	-	-	8	10.55	11.38	3.18	4.25	14.85
IVDMD	18	56.23	56.27	6.47	38.30	74.54	27	57.08	60.69	7.63	32.52	67.80	1	-	-	-	-	-	6	69.49	70.98	4.98	56.60	75.54
IVOMD	3	57.87	58.40	1.18	56.10	59.10	3	59.77	58.30	1.96	58.30	62.70	0	-	-	-	-	-	0	-	-	-	-	-
Effluent, kg/t	16	31.74	25.70	18.25	4.90	68.50	20	31.55	24.85	16.75	3.50	61.00	1	-	-	-	-	-	6	40.23	30.60	17.26	26.90	92.00
Gas losses	17	6.06	6.70	3.65	0.28	16.20	16	5.31	4.15	3.18	0.53	14.70	1	-	-	-	-	-	6	0.85	0.54	0.68	0.26	2.90
DM losses	22	12.45	10.10	5.42	2.90	25.38	31	11.03	8.31	5.18	2.10	24.60	1	-	-	-	-	-	2	14.09	14.09	6.09	8.00	20.18
WSC	3	1.37	1.80	0.58	0.50	1.82	3	1.43	1.20	0.77	0.50	2.58	0	-	-	-	-	-	0	-	-	-	-	-
Lactic acid	23	3.87	3.49	1.67	0.05	8.97	35	5.01	4.43	1.70	0.12	10.40	0	-	-	-	-	-	1	-	-	-	-	-
Acetic acid	16	1.36	1.09	0.76	0.30	4.53	28	0.98	0.73	0.59	0.05	3.44	0	-	-	-	-	-	1	-	-	-	-	-
Propionic acid	9	0.38	0.29	0.31	0.00	1.34	17	0.29	0.23	0.20	0.00	1.14	0	-	-	-	-	-	0	-	-	-	-	-
Butyric acid	16	0.06	0.05	0.03	0.00	0.21	24	0.05	0.03	0.03	0.00	0.18	0	-	-	-	-	-	1	-	-	-	-	-
Total acids[5]	7	6.04	6.09	2.19	0.50	10.45	15	7.30	7.25	1.43	0.63	10.49	0	-	-	-	-	-	0	-	-	-	-	-
Ethanol	2	1.17	1.17	0.14	1.04	1.31	4	1.20	0.94	0.41	0.89	2.02	0	-	-	-	-	-	0	-	-	-	-	-
Lactic:acetic acid	16	4.54	3.47	2.90	0.11	10.52	28	15.92	6.63	15.16	0.24	208.00	0	-	-	-	-	-	1	-	-	-	-	-
Total acids:ethanol	2	5.89	5.89	2.63	3.26	8.51	4	8.52	10.10	2.96	2.61	11.26	0	-	-	-	-	-	0	-	-	-	-	-
pH	59	4.75	4.70	0.55	3.36	6.80	80	4.47	4.26	0.54	3.15	6.50	2	4.85	4.85	0.05	4.80	4.90	12	4.51	4.39	0.33	4.00	5.35
Ammonia-N, % TN	52	9.70	8.86	4.28	1.23	39.95	73	8.56	8.50	2.90	0.85	27.78	4	24.59	21.94	15.54	3.20	51.28	9	4.20	3.65	0.98	3.20	8.60
LAB, log cfu/g	5	7.17	7.38	1.04	4.58	8.45	9	8.47	8.57	0.87	7.25	10.03	0	-	-	-	-	-	1	-	-	-	-	-
Yeasts, log cfu/g	4	4.69	4.54	1.68	2.76	6.90	9	3.81	3.70	1.24	2.06	6.70	0	-	-	-	-	-	0	-	-	-	-	-
Aerobic stability, h	2	59.45	59.45	36.55	22.90	96.00	3	114.00	114.00	4.00	108.00	120.00	1	-	-	-	-	-	1	-	-	-	-	-
Maximum T, ºC	10	27.53	25.85	3.82	23.00	35.30	10	28.65	29.80	3.86	23.00	40.00	0	-	-	-	-	-	0	-	-	-	-	-

[1]DM, dry matter; CP, crude protein; NDIN, neutral detergent insoluble N; ADIN, acid detergent insoluble N; NDF, neutral detergent fiber; ADF, acid detergent fiber; IVDMD, *in vitro* DM digestibility; IVOMD, *in vitro* organic matter digestibility; WSC, water-soluble carbohydrates; TN, total nitrogen; LAB, lactic-acid bacteria.
[2]Number of means.
[3]Standard deviation.
[4]Silages inoculated with both heterofermentative and homofermentative bacteria.
[5]Total acid content was calculated as the sum of lactic, acetic, and propionic acids.

Table 5. Range of fermentation patterns, nutritive value, and aerobic stability of untreated and inoculated grass silages (data are given in % of DM, unless otherwise stated).

Despite heterolactic inoculation, *L. buchneri* was the only ^{he}LAB evaluated in the studies that impaired silage quality by increasing pH, ammonia-N, and NDF and reducing crude protein (CP). The responses to inoculation with *L. buchneri* may be crop specific, as evidenced by a meta-analytical study that showed higher effectiveness when applied in corn silages, compared with grass and small-grain silages [29].

Overall, there were not consistent results by combining ^{ho}LAB and ^{he}LAB for grass silage. Utilization of both ^{ho}LAB and ^{he}LAB reduced the pH and ammonia-N concentration in silage; however, DM losses increased by 13.2%. The CP content also increased (+20.8%) following inoculation with both ^{ho}LAB and ^{he}LAB. Although NDF content increased 10.9% due to inoculation, IVDMD also improved by 23.6%. The number of studies assessing both ^{ho}LAB and ^{he}LAB as silage inoculants for grass is still very low, but the results reported in this survey suggest a suitable strategy to improve fermentation process along with enhanced silage digestibility.

The ash content of grass silages had an elevated value in all treatments (>9.5%) suggesting contamination, probably by soil, during the ensiling process. Tractors are utilized to transport the harvested forage, fill the silo, and compact the forage mass. Normally, soil in the tractor's tire might be deposited in the forage mass. Moreover, soil contamination is often responsible for the increased number of Clostridia and Bacilli in the ensiling forage [33, 34].

There was no comparison regarding positive responses and differences for heLAB and control silages (**Table 6**), because only a few studies used this group of bacteria (**Table 5**). Homolactic inoculation had the greatest frequency of positive responses for IVOMD, gas losses, acetic acid, lactic acid, and lactic:acetic acid. Furthermore, the greatest differences of response were observed for lactic:acetic acid, yeasts, WSC, and lactic and propionic acids. The increased production of lactic acid allowed by homolactic inoculation reduced gas and DM losses, after CO_2 production ceases and, consequently, preserved a greater amount of soluble sugars, increasing silage digestibility [6].

Regarding association of both ^{ho}LAB and ^{he}LAB, the greatest frequency of positive responses was observed for butyric acid and DM losses. In addition, the greatest differences in the response observed for the concentration of butyric acid, effluent production, and DM losses is likely to be related to the low number of studies evaluated.

The data from this survey suggest that ^{ho}LAB should be the only group used for the ensiling of grass, because this group had the greatest frequency of positive responses compared to ^{he}LAB and to utilization of ^{ho}LAB and ^{he}LAB combined.

2.1.3. Sugarcane silage

Data were summarized from a total of 50 studies, of which 21, 40, and 7 investigated the effect of ^{ho}LAB, ^{he}LAB, and a combination of both (mixed), respectively. Considering all treatments, the application rate of silage inoculants ranged from 2.5×10^4 to 2.5×10^{10} cfu/g of fresh forage.

The range of fermentation parameters, *in vitro* digestibility, and aerobic stability are given in **Table 7**.

Item[1]	Homofermentative LAB					Mixed[2]				
	Number of treatments		Mean		Difference, %	Number of treatments		Mean		Difference, %
	Total	Positive responses, %	Untreated	Inoculated		Total	Positive responses, %	Untreated	Inoculated	
DMoven, % as fed	78	26.9	22.32	24.55	+10.0	10	20.0	20.90	21.94	+5.0
Ash	24	0.0	–	–	–	6	0.0	–	–	–
CP	85	27.1	7.82	9.36	+19.7	10	10.0	6.16	6.20	+0.7
NDIN, % N	3	0.0	–	–	–	1	0.0	–	–	–
ADIN, % N	15	0.0	–	–	–	1	0.0	–	–	–
NDF	72	9.7	69.99	66.15	−5.5	10	10.0	77.08	64.98	−15.7
ADF	65	9.2	46.23	41.46	−10.3	10	20.0	52.37	50.87	−2.9
Hemicellulose	62	4.8	37.34	35.18	−5.8	10	0.0	–	–	–
Cellulose	29	13.8	38.81	35.40	−8.8	8	0.0	–	–	–
Lignin	29	13.8	5.90	5.20	−11.9	8	0.0	–	–	–
IVDMD	27	18.5	58.17	64.08	+10.2	6	0.0	–	–	–
IVOMD	3	66.7	57.25	60.50	+5.7	0	0.0	–	–	–
Effluent, kg/t	20	0.0	–	–	–	6	16.7	68.50	48.20	−29.6
Gas losses	19	57.9	5.56	3.49	−37.3	6	0.0	–	–	–
DM losses	31	25.8	14.60	9.52	−34.8	2	50.0	10.90	8.00	−26.6
WSC	3	33.3	1.82	2.58	+41.8	0	0.0	–	–	–
Lactic acid	35	48.6	3.59	5.08	+41.7	1	0.0	–	–	–
Acetic acid	28	53.6	1.16	0.74	−36.5	1	0.0	–	–	–
Propionic acid	17	5.9	0.77	1.09	+41.6	0	0.0	–	–	–
Butyric acid	24	37.5	0.05	0.03	−31.2	1	100.0	0.082	0.004	−95.1
Ethanol	4	0.0	–	–	–	0	0.0	–	–	–
Lactic:acetic acid	28	46.4	5.30	10.97	+106.9	1	0.0	–	–	–
pH	80	36.3	4.56	4.20	−8.0	12	0.0	–	–	–
Ammonia-N, % TN	73	30.1	9.97	7.65	−23.3	9	0.0	–	–	–
LAB, log cfu/g	9	44.4	8.36	9.32	+11.4	1	0.0	–	–	–
Yeasts, log cfu/g	9	11.1	5.83	2.06	−64.7	0	0.0	–	–	–
Molds, log cfu/g	4	0.0	–	–	–	0	0.0	–	–	–
Aerobic stability, h	3	33.3	96.00	120.00	+25.0	1	0.0	–	–	–

[1]DM, dry matter; CP, crude protein; NDIN, neutral detergent insoluble N; ADIN, acid detergent insoluble N; NDF, neutral detergent fiber; ADF, acid detergent fiber; IVDMD, *in vitro* DM digestibility; IVOMD, *in vitro* organic matter digestibility; WSC, water-soluble carbohydrates; TN, total nitrogen; LAB, lactic-acid bacteria.
[2]Silages inoculated with both heterofermentative and homofermentative bacteria.

Table 6. Summary of positive responses of silage inoculants on the fermentation patterns, nutritive value, and aerobic stability of grass silages (data are given in % of DM, unless otherwise stated).

Heterolactic inoculants have been used to increase the production of acetic acid in order to reduce aerobic deterioration [28]. For sugarcane silages, the use of [he]LAB was proposed to avoid

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yeast overgrowth and associated ethanol production, with reduced DM losses [35]. Moreover, the reduced DM losses involve a better preservation of WSC [35], which may lead an increased IVDMD of sugarcane silages.

Item[1]	Untreated						Homofermentative LAB						Heterofermentative LAB					
	n[2]	Mean	Median	SD[3]	Min	Max	n	Mean	Median	SD	Min	Max	n	Mean	Median	SD	Min	Max
DMoven, % as fed	52	24.03	23.23	2.96	16.40	33.70	51	23.75	23.96	2.47	15.70	32.60	64	26.10	26.41	2.91	19.00	34.90
Ash	20	4.64	4.74	1.44	1.75	7.36	8	5.24	5.60	1.18	2.80	7.14	20	4.54	4.86	1.46	2.00	7.24
CP	46	3.59	3.55	0.85	1.50	6.70	27	3.83	3.90	0.98	1.74	6.23	43	3.71	3.60	0.72	1.70	7.16
NDIN, % N	2	18.50	18.50	17.01	1.49	35.50	2	11.43	11.43	10.07	1.36	21.50	1	-	-	-	-	-
ADIN, % N	6	10.09	1.57	11.46	1.42	35.97	6	10.93	1.61	12.62	1.32	37.31	5	8.62	1.43	11.61	1.19	37.64
NDF	48	65.63	66.15	5.46	50.19	84.54	39	65.13	64.85	3.06	55.40	75.30	56	62.70	61.95	4.89	48.89	75.90
ADF	41	44.73	43.80	5.49	30.35	58.40	25	41.19	40.36	3.43	34.81	50.38	37	43.00	43.30	5.77	29.21	63.80
Hemicellulose	38	21.02	21.75	3.95	9.80	30.06	22	23.50	23.66	4.00	10.60	30.49	34	20.59	21.35	3.30	5.90	35.40
Cellulose	20	36.11	36.42	4.89	23.93	44.21	13	32.65	31.50	3.31	26.90	39.51	14	35.59	36.85	5.37	22.71	47.40
Lignin	20	8.95	8.05	2.60	4.93	15.20	13	6.87	7.80	1.85	3.21	9.70	14	10.16	8.35	3.46	6.40	16.40
IVDMD	32	48.09	47.30	6.68	32.60	65.40	19	52.24	51.16	4.67	41.10	63.90	30	50.99	48.73	6.65	36.80	69.00
Effluent, kg/t	20	50.86	45.60	27.36	5.40	107.31	3	28.10	29.90	18.34	0.59	53.80	21	48.37	46.10	22.86	2.30	92.18
Gas losses	20	21.14	19.14	6.44	9.43	36.00	2	15.30	15.30	0.50	14.80	15.80	23	18.34	15.91	6.95	8.50	36.20
DM losses	28	24.63	21.91	9.16	6.08	66.00	30	23.73	25.21	5.80	7.69	37.89	35	20.27	18.10	7.27	5.19	66.60
WSC	21	6.65	4.50	4.82	0.74	25.30	47	7.06	3.16	6.34	0.94	33.40	53	7.09	3.19	6.53	1.20	30.80
Lactic acid	27	3.41	3.63	1.43	0.02	6.07	41	4.37	4.50	0.85	0.34	6.63	55	3.09	3.14	0.76	0.07	8.00
Acetic acid	30	2.81	2.39	1.42	0.28	6.75	52	1.53	0.92	1.17	0.20	6.97	57	2.73	2.25	1.47	0.50	8.51
Propionic acid	26	0.46	0.29	0.39	0.00	2.47	48	0.49	0.34	0.30	0.01	1.90	55	0.52	0.40	0.39	0.00	3.91
Butyric acid	20	0.05	0.03	0.05	0.00	0.28	39	0.26	0.19	0.20	0.00	0.99	47	0.22	0.13	0.17	0.00	1.21
Total acids[4]	21	6.34	5.94	2.41	0.62	12.75	37	5.77	5.57	1.15	0.63	11.70	53	6.17	5.24	1.66	3.42	15.57
Ethanol	31	8.94	8.27	4.76	0.44	26.52	53	13.91	15.08	5.08	0.86	21.80	58	6.36	6.29	3.36	0.29	20.62
Lactic:acetic acid	26	1.92	1.25	1.38	0.01	9.00	41	8.00	6.67	5.05	0.36	20.70	54	1.83	1.43	1.07	0.02	5.08
Total acids:ethanol	18	0.84	0.49	0.54	0.23	2.90	35	0.80	0.34	0.76	0.27	6.15	51	1.70	0.78	1.36	0.42	13.76
pH	53	3.57	3.58	0.18	2.94	4.30	65	3.59	3.61	0.15	3.05	4.10	77	3.58	3.61	0.14	2.85	3.90
Ammonia-N, % TN	19	6.55	4.75	3.81	0.65	14.81	24	7.09	6.65	2.61	1.75	14.68	12	6.41	5.90	3.77	0.94	11.95
LAB, log cfu/g	16	7.00	8.01	1.62	2.05	8.90	31	6.75	6.82	0.55	5.65	8.16	49	7.25	7.03	1.37	2.09	9.95
Yeasts, log cfu/g	19	5.53	5.76	0.86	2.47	7.20	34	5.85	6.10	0.48	4.66	6.71	51	4.61	4.81	1.00	2.00	7.20
Aerobic stability, h	12	40.60	33.35	20.51	5.63	97.60	18	29.24	21.43	12.82	16.00	107.00	24	39.74	23.00	24.73	7.63	211.00
Maximum T, ºC	9	38.74	39.30	5.06	29.80	47.00	16	42.87	43.50	2.22	31.70	47.20	20	41.53	43.65	3.97	22.50	45.70

[1]DM, dry matter; CP, crude protein; NDIN, neutral detergent insoluble N; ADIN, acid detergent insoluble N; NDF, neutral detergent fiber; ADF, acid detergent fiber; IVDMD, *in vitro* DM digestibility; WSC, water-soluble carbohydrates; TN, total nitrogen; LAB, lactic-acid bacteria.
[2]Number of means.
[3]Standard deviation.
[4]Total acid content was calculated as the sum of lactic, acetic, and propionic acids.

Table 7. Range of fermentation patterns, nutritive value, and aerobic stability of untreated and inoculated sugarcane silages (data are given in % of DM, unless otherwise stated).

In this regard, considering the overall mean, acetic acid was unaffected, but [he]LAB reduced the ethanol concentration by 28.9%, because the number of yeasts was reduced. Reductions in yeast growth were probably due to the slight drop in the lactic:acetic acid ratio, in addition to a 12.8% increase in the production of propionic acid, which also has antifungal properties [30]. *L. brevis*, *L. buchneri*, and *L. hilgardii* are the most common [he]LAB used in sugarcane silage by Brazilian studies, and they are capable in producing 1,2-propanediol anaerobically [36]. Thus, the greatest production of propionic acid is likely to be related to the conversion of 1,2-propanediol to equimolar portions of 1-propanol and propionic acid, a process driven by

Lactobacillus diolivorans, assuming that this bacterium was present in ensiled forage [37, 38]. Moreover, heterolactic inoculation reduced gas and DM losses by 13.2% and 17.7%, respectively. Fermentative losses decreased because of the control of yeast growth. For each mole of glucose consumed, yeasts produce two moles of ethanol and CO_2, leading to 49% of DM losses in the ethanolic pathway [6]. In addition, *L. buchneri* was the main bacterium used in sugarcane silage, and this bacterium is known for its lack of acetaldehyde dehydrogenase [39], which reduces ethanol production. Conversely, the enhanced aerobic stability caused by heterolactic inoculation did not occur based on the overall mean.

The ADIN content decreased 14.6% due to heterolactic inoculation, suggesting that the control of yeast activity reduced the temperature of the ensiled mass during fermentation. Despite the effects on the fiber fraction, ^{he}LAB reduced the NDF content by 4.5%, likely due to increased hydrolysis of hemicellulose during fermentation [6]. Indeed, a net disappearance of hemicellulose was observed in ^{he}LAB-treated sugarcane silages (**Figure 6**), and as a consequence, the IVDMD increased by 6% on average.

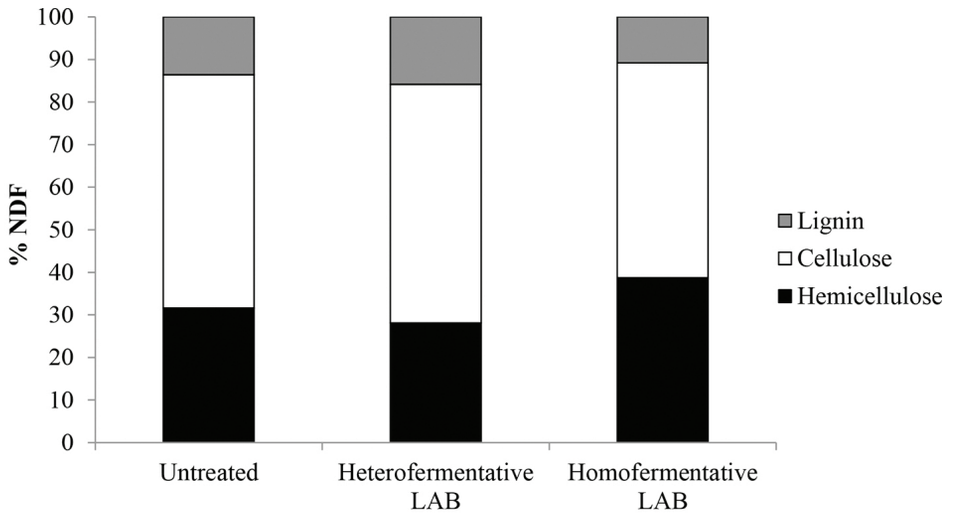

Figure 6. Proportion of hemicellulose, cellulose, and lignin in sugarcane silages untreated or inoculated with homofermentative and heterofermentative LAB (as-is a NDF basis).

The main action of homolactic inoculation is related to the increased preservation of nutrients during fermentation via the production of lactic acid [6]. In this regard, lactic acid increased by 28.2% in sugarcane silages inoculated with ^{ho}LAB. In addition, there was greater preservation of residual WSC (+6.2%), a reduction in the concentration of acetic acid (−45.7%), and a decrease in DM losses (−3.6%). As a consequence, IVDMD improved by 8.6%. However, homolactic inoculation increased ethanol production by 55.5% once yeasts are able to use WSC to grow in anaerobic conditions [8]. Furthermore, homolactic inoculation reduced the aerobic stability of silages by 11.4 h.

The frequency and magnitude of positive responses found in sugarcane silages from homolactic and heterolactic inoculations are given in **Table 8**.

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Item[1]	Homofermentative LAB					Heterofermentative LAB				
	Number of treatments		Mean		Difference, %	Number of treatments		Mean		Difference, %
	Total	Positive responses, %	Untreated	Inoculated		Total	Positive responses, %	Untreated	Inoculated	
DMoven, % as fed	51	7.8	24.07	27.50	+ 14.3	63	42.9	24.43	27.25	+ 11.5
Ash	8	12.5	5.24	2.98	- 43.1	20	5.0	6.97	5.91	- 15.2
CP	27	3.7	1.50	2.50	+ 66.7	42	7.1	2.96	3.52	+ 18.9
ADIN, % N	6	0.0	-	-	-	5	0.0	-	-	-
NDF	39	5.1	67.15	60.20	- 10.4	55	12.7	68.50	63.42	- 7.4
ADF	25	8.0	43.56	37.59	- 13.7	36	11.1	45.28	41.25	- 8.9
Hemicellulose	22	0.0	-	-	-	33	0.0	-	-	-
Cellulose	13	7.7	35.87	30.51	- 14.9	14	0.0	-	-	-
Lignin	13	0.0	-	-	-	14	0.0	-	-	-
IVDMD	19	10.5	43.26	52.63	+ 21.7	29	6.9	44.94	54.80	+ 21.9
IVOMD	2	0.0	-	-	-	0	0.0	-	-	-
Effluent, kg/t	3	33.3	6.98	0.59	- 91.5	21	9.5	98.91	78.38	- 20.8
Gas losses	2	0.0	-	-	-	23	30.4	17.31	11.28	- 34.8
DM losses	30	10.0	25.87	25.34	- 2.1	39	48.7	25.66	17.08	- 33.4
WSC	48	8.3	1.31	2.74	+ 109.6	55	25.5	4.28	5.87	+ 37.1
Lactic acid	41	19.5	2.13	3.71	+ 74.5	54	7.4	2.13	2.84	+ 33.6
Acetic acid	52	11.5	1.68	0.59	- 64.7	56	51.8	2.32	3.02	+ 30.1
Propionic acid	48	31.3	0.35	0.55	+ 58.3	54	37.0	0.33	0.65	+ 94.6
Butyric acid	39	2.6	0.07	0.03	- 61.5	46	8.7	0.12	0.03	- 75.5
Ethanol	53	9.4	9.09	2.43	- 73.3	57	56.1	10.39	5.72	- 45.0
Lactic:acetic acid	41	2.4	1.19	1.76	+ 48.4	53	1.9	1.19	0.72	- 39.0
pH	65	9.2	3.53	3.37	- 4.4	76	10.5	3.63	3.43	- 5.7
Ammonia-N, % TN	24	8.3	7.28	7.25	- 0.4	11	27.3	7.28	5.76	- 20.9
LAB, log cfu/g	31	0.0	-	-	-	49	14.3	7.83	9.39	+ 20.0
Yeasts, log cfu/g	34	5.9	6.21	4.23	- 31.9	51	19.6	5.87	3.34	- 43.1
Aerobic stability, h	18	0.0	-	-	-	23	8.7	76.80	151.50	+ 97.3

[1]DM, dry matter; CP, crude protein; NDIN, neutral detergent insoluble N; ADIN, acid detergent insoluble N; NDF, neutral detergent fiber; ADF, acid detergent fiber; IVDMD, *in vitro* DM digestibility; WSC, water-soluble carbohydrates; TN, total nitrogen; LAB, lactic-acid bacteria.

Table 8. Summary of positive responses of silage inoculants on the fermentation patterns, nutritive value, and aerobic stability of sugarcane silages (data are given in % of DM, unless otherwise stated).

Homolactic inoculation had the greatest frequency of positive responses for effluent production and propionic acid, but there is no clear explanation for these results. Furthermore, the greatest difference of responses was observed for WSC, effluent production, and lactic acid. Commercial homolactic inoculants investigated in Brazilian studies were often composed of pediococci, streptococci, and lactobacilli. Thus, the inoculation of silages with pediococci and streptococci leads to the rapid production of lactic acid and great sugar-to-lactic acid conversion efficiency [6, 40]. Afterward, the more acid-tolerant lactobacilli continue producing lactic acid until stable fermentation is achieved [6]. Therefore, the greater production of lactic acid and preservation of WSC from homolactic inoculation in sugarcane silages is expected.

Regarding heterolactic inoculation, the greatest frequency of positive responses was observed for ethanol, acetic acid, and DM losses. In addition, the greatest differences in responses were observed for aerobic stability and propionic acid. Second generation bacterial inoculants are expected to improve the aerobic stability of silages. As described earlier, the bacteria that composed the ^{he}LAB group used for sugarcane ensiling are able to convert lactic acid into acetic acid and 1,2-propanediol [25, 36, 41] when the primary fermentation is ended up. In turn, acetic

acid has an antagonistic effect on the growth of yeasts [30], and reductions in ethanol production are expected.

Item[1]	Untreated						Homofermentative LAB					
	n[2]	Mean	Median	SD[3]	Min	Max	n	Mean	Median	SD	Min	Max
DMoven, % as fed	7	41.26	51.29	14.46	14.64	56.20	11	33.39	23.49	15.60	14.81	62.64
Ash	4	11.51	12.47	1.99	7.60	13.49	4	10.54	11.64	1.76	7.02	11.85
CP	7	19.75	19.51	2.00	16.38	24.33	11	20.14	20.49	1.83	15.90	23.44
NDIN, % N	2	13.03	13.03	1.71	11.32	14.73	3	13.47	12.28	1.82	11.93	16.21
ADIN, % N	3	15.04	15.92	2.31	11.57	17.63	7	16.68	17.17	1.55	11.24	19.08
NDF	8	45.06	45.82	3.19	40.18	52.04	13	44.26	43.43	4.18	37.86	54.28
ADF	7	38.29	39.76	2.26	33.99	40.39	9	38.00	39.94	3.00	33.22	42.50
Hemicellulose	6	7.59	6.88	1.99	5.43	13.57	7	8.06	7.25	2.50	4.14	11.78
Cellulose	3	26.42	25.41	1.47	25.22	28.63	5	26.44	25.60	1.53	24.38	29.72
Lignin	4	12.20	11.51	2.16	9.25	16.52	9	13.29	12.71	3.12	8.84	18.87
IVDMD	3	68.92	66.50	5.92	62.46	77.81	7	67.98	65.13	6.22	60.21	75.57
DM losses	2	10.58	10.58	1.09	9.49	11.67	6	5.17	4.95	3.13	1.33	9.55
WSC	3	2.78	2.44	1.00	1.62	4.27	7	3.17	2.97	1.32	1.57	4.84
Lactic acid	3	4.92	4.45	2.82	1.16	9.15	7	7.17	5.62	4.00	0.95	13.83
Acetic acid	3	5.03	3.90	3.51	0.89	10.29	7	5.24	3.93	2.05	2.35	8.36
Propionic acid	3	0.14	0.14	0.10	0.00	0.29	7	0.20	0.10	0.15	0.00	0.41
Butyric acid	3	0.33	0.01	0.43	0.00	0.99	7	1.00	0.02	1.13	0.00	2.85
Total acids[4]	3	10.09	11.74	3.16	5.34	13.2	7	12.61	13.23	2.86	7.97	17.84
Ethanol	3	0.37	0.46	0.23	0.02	0.61	7	1.44	0.51	1.26	0.02	3.08
Lactic:acetic acid	3	2.46	2.40	1.61	0.11	4.87	7	2.26	3.02	1.59	0.12	4.57
Total acids:ethanol	3	89.78	25.29	88.5	21.5	223	7	79.97	32.91	103.67	3.20	442.83
pH	6	4.83	4.66	0.39	4.25	5.50	10	4.98	4.78	0.60	4.22	6.11
Ammonia-N, % TN	6	13.85	8.21	9.50	5.21	29.48	10	22.85	28.61	10.99	5.30	37.27
Maximum T, °C	3	26.85	27.00	1.70	24.30	29.25	7	27.21	27.33	1.18	23.78	28.63

[1]DM, dry matter; CP, crude protein; NDIN, neutral detergent insoluble N; ADIN, acid detergent insoluble N; NDF, neutral detergent fiber; ADF, acid detergent fiber; IVDMD, *in vitro* DM digestibility; WSC, water-soluble carbohydrates; TN, total nitrogen.
[2]Number of means.
[3]Standard deviation.
[4]Total acid content was calculated as the sum of lactic, acetic, and propionic acids.

Table 9. Range of fermentation patterns, nutritive value, and aerobic stability of untreated and inoculated alfalfa silages (data are given in % of DM, unless otherwise stated).

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2.1.4. Alfalfa, sorghum, and high-moisture corn silages

Data on alfalfa, sorghum, and HMC silages were summarized from 7, 10, and 10 studies, respectively. All studies comprising alfalfa and sorghum evaluated ^{ho}LAB only. For HMC silages, ^{ho}LAB, ^{he}LAB, and a combination between both (mixed) were investigated in six, three, and one study, respectively. Considering all treatments, the application rate of silage inoculant for alfalfa, sorghum, and HMC ranged from 1×10^5 to 9.9×10^5 cfu/g, 9.99×10^4 to 8×10^5 cfu/g, and 5×10^4 to 1×10^6 cfu/g of fresh forage, respectively.

Item[1]	Untreated						Homofermentative LAB					
	n[2]	Mean	Median	SD[3]	Min	Max	n	Mean	Median	SD	Min	Max
DMoven, % as fed	25	30.58	30.89	4.48	19.80	42.33	35	28.44	26.31	4.17	21.70	42.29
Ash	5	5.25	4.20	1.55	3.79	8.77	5	4.88	3.76	1.60	3.27	8.53
CP	18	7.03	7.04	1.40	5.15	13.28	22	7.80	7.55	1.85	5.32	14.08
NDF	23	52.97	53.13	8.87	36.67	73.89	31	56.13	58.67	7.37	35.36	71.42
ADF	19	28.20	23.70	6.78	18.99	44.95	23	31.33	28.77	7.49	19.60	45.78
Hemicellulose	19	22.84	23.20	3.13	14.76	31.65	23	22.90	22.61	2.56	11.68	27.70
Cellulose	14	23.25	21.22	4.51	17.03	39.61	16	24.63	23.37	3.80	17.28	39.99
Lignin	14	3.93	3.42	1.47	1.96	8.34	16	4.58	3.84	2.03	1.99	9.32
IVDMD	16	58.39	59.02	2.54	46.38	62.88	22	59.46	59.77	1.51	55.00	61.75
DM losses	11	1.88	1.69	0.77	0.00	5.12	13	4.18	2.48	2.92	0.31	14.14
WSC	12	1.12	0.32	1.28	0.12	7.34	14	1.49	0.23	2.02	0.14	6.62
Lactic acid	8	5.69	5.20	1.12	3.95	8.54	10	5.80	6.06	1.32	3.90	7.65
Acetic acid	5	1.55	1.52	0.42	0.86	2.42	7	1.53	1.21	0.66	0.82	2.93
Lactic:acetic acid	5	4.38	3.82	1.17	2.89	7.14	7	5.35	3.79	2.50	2.50	8.33
pH	16	3.94	3.86	0.18	3.74	4.94	20	3.94	3.87	0.16	3.66	4.88
Ammonia-N, % TN	15	6.01	4.62	3.76	0.26	16.87	17	5.48	4.05	3.18	0.38	16.79

[1]DM, dry matter; CP, crude protein; NDF, neutral detergent fiber; ADF, acid detergent fiber; IVDMD, *in vitro* DM digestibility; WSC, water-soluble carbohydrates; TN, total nitrogen.
[2]Number of means.
[3]Standard deviation.

Table 10. Range of fermentation patterns, nutritive value, and aerobic stability of untreated and inoculated sorghum silages (data are given in % of DM, unless otherwise stated).

The range of fermentation parameters, *in vitro* digestibility, and aerobic stability in alfalfa, sorghum, and HMC silages are given in **Tables 9**, **10**, and **11**, respectively. Considering the

overall mean, there was a large difference in the DM content of alfalfa silages, with 33.4% in inoculated silage and 41.3% in untreated silage. Homolactic inoculation increased the concentration of lactic acid by 45.8% in alfalfa silage; however, the pH of silage did not decline, compared with untreated silage; this point may be a consequence of the greater moisture content found in ^{ho}LAB-inoculated silages.

The ^{ho}LAB reduced DM losses by 51.1% in alfalfa silages. Conversely, homolactic inoculation increased the concentration of ammonia-N by 65%, and an increase from 0.37 to 1.44% in the ethanol concentration was also observed. The greater concentration of ammonia-N was unexpected, since lactic acid produced by ^{ho}LAB should be able to decrease proteolytic bacterial populations within the ensiled mass.

Considering the frequency of positive responses of inoculation, only the acetic acid concentration was affected, which was reduced by ^{ho}LAB in 14.3% (−35.95%) of the treatments. Usually, improvements on quality of alfalfa silages have been reported due to the homolactic inoculation [42, 43] most likely due to increases on the numbers of LAB, which is quite low in alfalfa [44]. Although the present survey does not contain data regarding number of LAB in alfalfa silages, homolactic inoculation improved the preservation of this crop.

Item[1]	Untreated						Homofermentative LAB						Heterofermentative LAB					
	n[2]	Mean	Median	SD[3]	Min	Max	n	Mean	Median	SD	Min	Max	n	Mean	Median	SD	Min	Max
DMoven, % as fed	17	64.36	64.43	2.55	52.47	69.22	17	64.86	64.23	3.95	51.52	72.51	8	65.94	65.98	0.50	64.92	66.66
Ash	14	1.42	1.33	0.17	1.27	2.43	13	1.32	1.32	0.06	1.23	1.42	1	-	-	-	-	-
CP	23	9.12	9.23	1.88	6.39	17.32	23	9.02	9.35	1.68	6.62	11.71	5	12.03	10.69	2.51	10.17	18.32
EE	12	3.76	3.54	0.52	2.93	5.21	15	3.94	3.79	0.74	2.71	5.68	0	-	-	-	-	-
ADIN, % N	8	0.00	0.00	0.00	0.00	0.01	8	0.01	0.01	0.00	0.00	0.02	0	-	-	-	-	-
NDF	13	15.84	14.71	6.20	5.53	39.50	15	16.00	10.84	7.55	7.38	32.67	5	6.96	6.24	1.38	5.79	10.42
ADF	17	4.38	4.44	1.69	1.19	7.60	16	4.76	4.85	1.85	0.54	8.32	5	2.19	1.43	1.33	1.04	5.51
Hemicellulose	8	15.54	13.50	5.66	4.34	36.12	8	17.29	17.98	6.81	6.20	29.10	4	4.74	4.77	0.21	4.45	4.97
Gas losses	2	1.93	1.93	0.08	1.85	2.01	0	-	-	-	-	-	8	2.15	2.30	0.70	1.21	2.99
DM losses	9	1.78	1.50	0.87	0.46	3.69	6	1.41	1.02	1.12	0.11	3.29	4	1.54	1.57	0.12	1.33	1.71
Lactic acid	7	1.73	1.36	0.62	1.22	3.90	6	1.27	1.25	0.11	1.13	1.49	4	3.58	3.66	0.21	3.16	3.84
Acetic acid	7	0.19	0.16	0.05	0.15	0.36	6	0.19	0.19	0.02	0.15	0.23	4	0.37	0.36	0.03	0.34	0.42
Propionic acid	7	0.10	0.10	0.02	0.01	0.14	6	0.13	0.13	0.02	0.10	0.15	4	0.02	0.02	0.01	0.01	0.03
Total acids[4]	6	1.65	1.65	0.08	1.47	1.77	6	1.58	1.56	0.09	1.44	1.84	0	-	-	-	-	-
Lactic: acetic acid	6	8.50	8.55	0.91	6.25	9.86	6	6.95	6.85	1.00	5.22	8.94	0	-	-	-	-	-
pH	12	4.02	3.98	0.11	3.83	4.40	8	4.12	4.08	0.19	3.81	4.42	9	4.03	4.02	0.10	3.90	4.20
Ammonia-N, % TN	3	0.56	0.72	0.21	0.24	0.72	0	-	-	-	-	-	4	0.28	0.28	0.02	0.26	0.32
LAB, log cfu/g	2	5.53	5.53	0.62	4.91	6.14	4	6.09	6.44	0.77	4.56	6.92	0	-	-	-	-	-
Yeasts, log cfu/g	4	4.63	4.32	1.25	3.20	6.70	4	5.27	5.76	0.97	3.33	6.23	8	4.30	5.10	1.80	1.34	6.39
Molds, log cfu/g	2	3.67	3.67	0.02	3.65	3.68	0	-	-	-	-	-	8	3.06	2.99	0.67	1.71	4.42
Aerobic stability, h	8	51.91	54.75	10.06	36.00	68.00	6	45.00	45.00	12.50	25.50	60.00	8	154.39	111.15	70.41	86.70	265.00
Maximum T, ºC	6	32.80	34.00	3.23	25.20	37.20	6	34.57	37.35	5.64	25.00	40.50	0	-	-	-	-	-

[1]DM, dry matter; CP, crude protein; EE, ether extract; ADIN, acid detergent insoluble N; NDF, neutral detergent fiber; ADF, acid detergent fiber; TN, total nitrogen; LAB, lactic-acid bacteria.
[2]Number of means.
[3]Standard deviation.
[4]Total acid content was calculated as the sum of lactic, acetic, and propionic acids.

Table 11. Range of fermentation patterns, nutritive value, and aerobic stability of untreated and inoculated high-moisture corn silages (data are given in % of DM, unless otherwise stated).

Sorghum silages had few alterations on fermentation parameters due to homolactic inoculation. However, DM losses increased from 1.88 to 4.18% when silages were inoculated, when compared with losses in untreated silage.

The inoculation of sorghum silages also increased the NDF content by 6%, but the ammonia-N concentration decreased by 8.8%. Positive responses from inoculation in sorghum silages occurred only for DM (+14.5%), CP (+15.2%), NDF (−8.5%), and IVDMD (+20.8%) at frequencies of 8.6, 4.6, 6.5, and 9.1%, respectively. Overall, the lack of positive results from inoculation is likely related to the suitable characteristics of sorghum for the ensiling process [45]. Similar to corn, sorghum plants also have good fermentation capability, considerable WSC and DM contents, and low buffer capacity. However, sorghum silages often have low aerobic stability because the suitable characteristics described earlier [13, 15]. Although aerobic deterioration can become a great problem under tropical conditions, there is not any study that assessed ^{he}LAB for sorghum silage in Brazil.

Item[1]	Homofermentative LAB					Heterofermentative LAB				
	Number of treatments		Mean		Difference, %	Number of treatments		Mean		Difference, %
	Total	Positive responses, %	Untreated	Inoculated		Total	Positive responses, %	Untreated	Inoculated	
DMoven, % as fed	17	11.8	69.07	71.71	+ 3.8	8	0.0	-	-	-
Ash	13	15.4	1.33	1.24	- 6.4	1	100.0	2.43	2.31	- 4.9
CP	23	8.7	6.50	7.64	+ 17.6	5	0.0	-	-	-
EE	15	6.7	3.16	3.84	+ 21.5	0	0.0	-	-	-
ADIN	8	12.5	0.008	0.005	- 37.5	0	0.0	-	-	-
NDF	15	33.3	21.89	19.70	- 10.0	5	0.0	-	-	-
ADF	16	18.8	2.42	1.23	- 49.2	5	0.0	-	-	-
Hemicellulose	8	37.5	13.41	8.25	- 38.5	4	0.0	-	-	-
Effluent, kg/t	0	0.0	-	-	-	4	0.0	-	-	-
Gas losses	0	0.0	-	-	-	8	0.0	-	-	-
DM losses	6	50.0	2.74	0.31	- 88.6	4	0.0	-	-	-
Lactic acid	6	0.0	-	-	-	4	0.0	-	-	-
Acetic acid	6	0.0	-	-	-	4	0.0	-	-	-
Propionic acid	6	83.3	0.11	0.13	+ 18.3	4	0.0	-	-	-
Lactic: acetic acid	6	16.7	6.25	8.94	+ 43.0	0	0.0	-	-	-
pH	8	0.0	-	-	-	9	33.3	3.98	3.90	- 2.0
Ammonia-N, % TN	0	0.0	-	-	-	4	0.0	-	-	-
LAB, cfu/g	4	50.0	5.53	6.83	+ 23.5	0	0.0	-	-	-
Yeasts, cfu/g	4	0.0	-	-	-	8	50.0	6.70	2.51	- 62.5
Molds, cfu/g	0	0.0	-	-	-	8	0.0	-	-	-
Aerobic stability, h	6	16.7	42.00	54.00	+ 28.6	8	87.5	65.90	158.37	+ 140.3

[1]DM, dry matter; CP, crude protein; EE, ether extract; ADIN, acid detergent insoluble N; NDF, neutral detergent fiber; ADF, acid detergent fiber; TN, total nitrogen; LAB, lactic-acid bacteria.

Table 12. Summary of positive responses of silage inoculants on the fermentation patterns, nutritive value, and aerobic stability of high-moisture corn silages (data are given in % of DM, unless otherwise stated).

It was not observed significant differences for lactic acid production and final pH by homolactic inoculation in HMC silages. As described earlier, ^{ho}LAB are used with the goal to increase lactic acid production and quickly reduce pH of the ensiled crop [6, 8]. In addition, there is an expected inhibition on the growth of undesirable microorganisms such as enterobacteria and clostridia [6, 8]. These effects likely help us to understand why DM losses decreased by 20.4% due to homolactic inoculation. Considering the overall mean, homolactic inoculation reduced

the aerobic stability by 6.9 h, compared with untreated silage. Homolactic inoculation can impair the aerobic stability of silages in some cases [32], because the lactic acid produced and the increased preservation of the forage crop can lead to an increase in the number of spoilage microorganisms, mainly yeasts.

Considering the overall mean, heterolactic inoculation of HMC silages increased the concentration of lactic and acetic acids by 106.5 and 92.7%, respectively. Due to the antifungal properties of acetic acid [30], the aerobic stability of HMC silages inoculated with ^{he}LAB increased by 102.5 h compared to untreated silage. Furthermore, heterolactic inoculation reduced the NDF content (−56%) and increased the CP content (+32%).

The frequency and difference of the positive responses found in HMC silages from homolactic and heterolactic inoculations are given in **Table 12**. Homolactic inoculation had the greatest frequency of positive responses for DM losses and LAB count. Furthermore, the greatest difference of responses was observed for DM losses and ADF content. Despite heterolactic inoculation, the greatest frequency of positive responses and the greatest magnitude of responses were observed for aerobic stability.

The fermentation of HMC silages is often restricted due to low moisture and fermentable sugar content, and the quantity of total acids produced is quite low [46]. Indeed, the data from this survey showed an increase in fermentation products in HMC silages treated with bacterial inoculants, and ^{he}LAB had the greatest impact on fermentation end products and aerobic stability.

Even without statistical analysis, the mean and median values for most variables were very similar, indicating that the data were normally distributed. Although the results of the current survey for all crops investigated are encouraging, some caution should be used when interpreting the data, because the inoculants, application rate, strains, and crops were not the same in each study and the conditions were highly variable. Moreover, the goal of this chapter was to conduct a survey that provides an exploratory picture of the silage trials carried out in Brazil, more than a proper comparison among treatments, which require analyses more specific.

2.2. Animal performance

Considerable efforts have been devoted to understand how silage inoculants affect animal performance, since such improvements are, in many cases, the principal economic justification for their use, in addition to improved nutrient recovery and enhanced aerobic stability already presented above.

Significant improvements on the performance of animals fed inoculated silages have been found in studies carried out in Europe and North America, although less frequently than studies regarding changes in fermentation caused by inoculation [47]. In a previous review concerning bacterial inoculants in Brazil (see [5]), there was not a definitive conclusion regarding the effect of inoculation on animal performance due to the low number of studies, but the authors suggested that the difference and frequency of responses should be similar to those observed in other countries (see [48]).

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In our survey, we found 42 studies that included feeding inoculated silages to animals in Brazil. In these studies, feed intake, digestibility, and/or growth performance were measured. Twenty of the 42 studies were conducted in cattle, 19 in sheep, 2 in pigs, and 1 in poultry. In this survey, we summarized data into two groups of silages: (1) untreated and (2) inoculated (regardless of the type of bacterial inoculant used). Only the performance of cattle and sheep fed corn, grass, and sugarcane silages were reported in this chapter, because there were a greater number of trials in these crops than others. Nevertheless, the number of studies is much lower than those reported in the international literature.

Item[1]	Cattle												Sheep											
	Untreated						Inoculated						Untreated						Inoculated					
	n[2]	Mean	Median	SD[3]	Min	Max	n	Mean	Median	SD	Min	Max	n	Mean	Median	SD	Min	Max	n	Mean	Median	SD	Min	Max
Intake, kg/day																								
DM	4	8.37	8.08	1.53	6.63	10.70	5	7.93	7.14	1.34	6.63	10.20	4	1.06	1.07	0.20	0.73	1.37	7	1.14	1.11	0.18	0.70	1.37
DM, % BW	2	2.19	2.19	0.21	1.98	2.40	2	2.22	2.22	0.18	2.04	2.40	2	2.55	2.55	0.27	2.28	2.81	3	2.42	2.39	0.22	2.13	2.75
OM	3	7.74	6.81	1.71	6.10	10.30	4	7.22	6.61	1.29	5.87	9.80	2	1.14	1.14	0.18	0.96	1.31	4	1.09	1.04	0.08	1.02	1.25
NDF	3	2.93	2.92	0.32	2.46	3.40	4	2.84	2.84	0.26	2.40	3.30	2	0.47	0.47	0.06	0.41	0.53	4	0.48	0.47	0.03	0.44	0.51
CP	3	0.91	0.80	0.19	0.74	1.20	4	0.87	0.78	0.17	0.71	1.20	2	0.14	0.14	0.01	0.13	0.15	4	0.14	0.14	0.01	0.13	0.15
Intake of digestible nutrients, kg/day																								
DM	2	4.33	4.33	0.11	4.22	4.44	3	4.49	4.52	0.35	3.97	4.98	3	0.67	0.72	0.14	0.47	0.83	5	0.66	0.71	0.10	0.45	0.80
OM	2	4.41	4.41	0.23	4.18	4.64	3	4.44	4.56	0.34	3.92	4.83	2	0.76	0.76	0.06	0.69	0.82	4	0.70	0.70	0.05	0.60	0.79
NDF	2	1.37	1.37	0.03	1.34	1.40	3	1.36	1.35	0.13	1.17	1.55	2	0.20	0.20	0.02	0.19	0.22	4	0.21	0.21	0.02	0.19	0.24
CP	2	0.47	0.47	0.05	0.42	0.52	3	0.46	0.46	0.03	0.42	0.50	2	0.09	0.09	0.00	0.09	0.10	4	0.09	0.09	0.01	0.07	0.10
Digestibility, %																								
DM	3	64.45	63.67	1.58	62.86	66.83	4	66.49	68.18	3.33	59.83	69.77	5	62.08	60.40	4.92	55.46	71.90	7	62.23	61.20	3.79	54.80	68.30
OM	2	68.31	68.31	0.14	68.17	68.45	3	69.66	70.70	1.90	66.81	71.47	2	67.35	67.35	5.15	62.20	72.50	4	64.48	65.50	4.58	56.90	70.00
NDF	2	51.46	51.46	5.46	46.00	56.92	3	50.38	48.85	2.08	48.80	53.50	5	45.43	50.63	8.29	34.95	54.20	7	46.90	48.95	5.50	37.21	54.90
CP	2	60.93	60.93	4.40	56.53	65.33	3	60.84	60.01	1.73	59.08	63.44	3	62.37	60.50	6.55	54.41	72.20	5	62.90	66.00	4.24	56.02	66.80
Performance																								
Feed efficiency	4	6.22	6.56	1.15	4.34	7.40	4	7.12	7.14	0.65	5.90	8.28	1	-	-	-	-	-	2	5.23	5.23	0.09	5.14	5.31
ADG, kg/day	4	1.41	1.29	0.25	1.15	1.90	4	1.37	1.32	0.22	1.03	1.80	1	-	-	-	-	-	2	0.20	0.20	0.00	0.20	0.21

[1]DM, dry matter; BW, body weight; OM, organic matter; NDF, neutral detergent fiber; CP, crude protein; ADG, average daily gain.
[2]Number of means.
[3]Standard deviation.

Table 13. Range of feed intake, digestibility, and growth performance of cattle and sheep fed untreated and inoculated corn silages.

The inoculation of corn silage slightly depressed DM intake, feed efficiency, and average daily gain (ADG) of cattle (**Table 13**). Conversely, cattle fed inoculated corn silages had small increases in DM and OM digestibility, resulting in a higher intake of digestible DM (+0.16 kg/day). Regarding the performance of sheep, the inoculation of corn silage increased DM intake by 7.2%, but the digestibility and intake of digestible nutrients were unaffected, in general. Data regarding ADG were not considered, because only one study measured this parameter and, as a prerequisite of this survey, all comparisons between treatments were made considering a minimum of two studies.

The inoculation of tropical grass silages reduced the DM intake (−0.14 kg/day) in cattle (**Table 14**).

However, cattle fed inoculated grass silages exhibited better feed efficiency than cattle fed untreated silage, whereas ADG was similar between treatments (**Table 14**). Digestibility of DM, OM, NDF, and CP was slightly affected by inoculation. Furthermore, sheep fed inoculated silages exhibited higher DM intake (+11.7%), whereas bacterial inoculants had little effect on silage digestibility.

The inoculation of sugarcane silages negatively impacted DM intake in cattle (−0.56 kg/day), as well as the intake of digestible nutrients (**Table 15**). As consequence, the ADG of cattle fed inoculated silages was lower than cattle fed untreated silages (1.17 vs. 1.21 kg/day, respectively). Few measurements were made in sheep fed sugarcane silages, but positive responses from inoculation were observed on DM and NDF intake, which increased by 4.6 and 11.3%, respectively; however, inoculation reduced DM digestibility by 16.6%.

Item[1]	Cattle												Sheep											
	Untreated						Inoculated						Untreated						Inoculated					
	n[2]	Mean	Median	SD[3]	Min	Max	n	Mean	Median	SD	Min	Max	n	Mean	Median	SD	Min	Max	n	Mean	Median	SD	Min	Max
Intake, kg/day																								
DM	3	8.10	9.70	2.26	4.71	9.90	3	7.96	9.10	2.19	4.67	10.10	2	0.77	0.77	0.33	0.44	1.10	3	0.86	1.02	0.29	0.42	1.13
DM, % BW	4	2.39	2.35	0.07	2.31	2.53	4	2.36	2.38	0.05	2.25	2.42	3	1.74	1.63	0.28	1.43	2.16	6	1.90	1.89	0.23	1.36	2.29
Intake of digestible nutrients, kg/day																								
DM	3	4.84	5.31	1.16	3.10	6.13	3	4.77	5.09	1.11	3.10	6.11	2	0.50	0.50	0.29	0.21	0.78	3	0.55	0.65	0.22	0.21	0.79
Digestibility, %																								
DM	3	60.78	61.90	4.05	54.70	65.74	3	60.92	60.50	3.62	55.90	66.35	6	58.53	60.45	9.08	42.79	71.10	11	59.44	60.88	6.09	49.47	69.50
OM	2	60.10	60.10	3.40	56.70	63.50	2	60.10	60.10	2.30	57.80	62.40	1	-	-	-	-	-	2	65.25	65.25	0.20	65.04	65.45
NDF	3	50.09	44.70	8.46	42.80	62.78	3	48.80	43.50	8.40	41.50	61.39	5	55.80	54.71	8.07	44.52	69.20	9	55.99	60.67	8.77	36.10	69.48
CP	3	58.83	55.90	4.97	54.30	66.28	3	56.60	52.30	6.74	50.80	66.71	6	62.17	65.72	9.24	43.55	73.96	11	64.12	64.61	5.38	45.85	75.80
Performance																								
Feed efficiency	3	8.21	8.21	0.40	7.62	8.82	3	7.85	8.12	0.56	7.00	8.42	0	-	-	-	-	-	0	-	-	-	-	-
ADG, kg/day	3	1.15	1.10	0.10	1.06	1.30	3	1.17	1.20	0.10	1.02	1.30	0	-	-	-	-	-	0	-	-	-	-	-

[1]DM, dry matter; BW, body weight; OM, organic matter; NDF, neutral detergent fiber; CP, crude protein; ADG, average daily gain.
[2]Number of means.
[3]Standard deviation.

Table 14. Range of feed intake, digestibility, and growth performance of cattle and sheep fed untreated and inoculated grass silages.

Overall means of this survey consistently appointed for a reduction in DM intake when cattle were fed inoculated corn, grass, and sugarcane silages. However, effects of silage inoculants on feed intake and growth performance are widely varied and likely are microorganisms and strains specific along with dose dependent.

We also calculated the frequency and difference of positive responses, in addition to the impact of bacterial inoculation in experiments with cattle and sheep (**Tables 16** and **17**). There was great frequency of positive responses of inoculation concerning DM and OM digestibility in cattle fed corn silage. Similarly, inoculation had a great impact on the performance of cattle fed sugarcane silage, with feed efficiency and ADG improving by 80%. The greater ADG observed in cattle fed sugarcane silage likely arises from a better preservation of WSC during fermentation leading to the improved nutritive value of inoculated silages. In this regard, improve-

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ments in nutritive value of silages from bacterial inoculation may be strongly correlated with enhanced animal performance [47, 48]. However, the great frequency and difference of the responses might be associated with the low number of studies carried out that evaluated animal performance in Brazil.

The frequency of positive responses observed in sheep consuming inoculated silages was greater than those found in cattle. Sheep fed corn silage had a great frequency of positive responses for inoculation concerning DM, OM, NDF, and CP intake (≥50%). The ADG also improved in 50% of treatments, an overall increase of 4%. For grass silage, the greater frequency of positive responses from inoculation was observed for digestibility (DM, NDF, and CP). Conversely, only the intake of digestible NDF and NDF digestibility had positive responses by inoculation in sugarcane silages.

Item[1]	Cattle												Sheep											
	Untreated						Inoculated						Untreated						Inoculated					
	n[2]	Mean	Median	SD[3]	Min	Max	n	Mean	Median	SD	Min	Max	n	Mean	Median	SD	Min	Max	n	Mean	Median	SD	Min	Max
Intake, kg/day																								
DM	8	10.36	11.15	1.94	6.90	12.71	11	9.80	9.61	1.64	6.89	12.80	2	1.42	1.42	0.03	1.39	1.45	2	1.49	1.49	0.16	1.33	1.64
DM, % BW	4	2.16	2.33	0.44	1.29	2.70	5	2.02	2.35	0.60	1.26	2.80	1	-	-	-	-	-	1	-	-	-	-	-
OM	2	11.60	11.60	0.10	11.50	11.70	2	11.65	11.65	0.75	10.90	12.40	1	-	-	-	-	-	1	-	-	-	-	-
NDF	5	5.74	6.11	0.81	3.72	6.40	6	5.10	5.51	0.88	3.78	6.03	2	0.67	0.67	0.06	0.61	0.72	2	0.74	0.74	0.00	0.74	0.74
CP	3	1.70	1.80	0.20	1.41	1.90	3	1.65	1.80	0.27	1.25	1.90	0	-	-	-	-	-	0	-	-	-	-	-
Intake of digestible nutrients, kg/day																								
DM	3	5.62	5.45	0.94	4.37	7.02	4	4.99	4.93	0.55	4.39	5.70	1	-	-	-	-	-	1	-	-	-	-	-
OM	2	6.12	6.12	0.41	5.72	6.53	2	5.78	5.78	0.09	5.69	5.88	1	-	-	-	-	-	1	-	-	-	-	-
NDF	3	1.77	1.78	0.13	1.57	1.96	4	1.54	1.60	0.46	0.89	2.09	1	-	-	-	-	-	1	-	-	-	-	-
CP	2	1.23	1.23	0.06	1.17	1.29	2	1.13	1.13	0.02	1.11	1.15	0	-	-	-	-	-	0	-	-	-	-	-
Digestibility, %																								
DM	3	54.97	55.30	5.84	46.20	63.40	4	55.10	55.50	8.60	44.50	64.90	2	61.32	61.32	7.68	53.64	69.00	3	51.12	46.67	13.99	34.58	72.10
OM	3	57.07	55.80	5.76	49.70	65.70	4	57.98	58.85	8.18	47.40	66.80	1	-	-	-	-	-	1	-	-	-	-	-
NDF	3	35.47	29.20	11.49	24.50	52.70	4	35.95	36.90	17.00	14.80	55.20	1	-	-	-	-	-	1	-	-	-	-	-
CP	2	66.55	66.55	1.55	65.00	68.10	2	61.25	61.25	2.75	58.50	64.00	0	-	-	-	-	-	0	-	-	-	-	-
Performance Feed efficiency	3	8.16	9.37	1.64	5.71	9.41	5	8.06	8.43	0.78	6.45	9.15	1	-	-	-	-	-	1	-	-	-	-	-
ADG, kg/day	3	1.21	0.94	0.44	0.82	1.87	5	1.17	1.04	0.20	0.97	1.61	1	-	-	-	-	-	1	-	-	-	-	-

[1]DM, dry matter; BW, body weight; OM, organic matter; NDF, neutral detergent fiber; CP, crude protein; ADG, average daily gain.
[2]Number of means.
[3]Standard deviation.

Table 15. Range of feed intake, digestibility, and growth performance of cattle and sheep fed untreated and inoculated sugarcane silages.

The results found in Brazilian studies suggest a greater effect of inoculation when there is a positive response, compared to those from other countries. In Europe, a review of 14 studies reported increases in DM intake (+4.8%) and milk production (+4.6%) when animals were fed silage inoculated with *L. plantarum* strain MTD1 [49]. Similarly, a review of studies carried out between 1990 and 1995 reported that in 28, 53, and 47% of these studies, there were increases in DM intake (+4.8%), ADG (+4.6%), and milk production (+4.6%), respectively [48].

Item[1]	Number of treatments		Mean		Difference, %
	Total	Positive responses, %	Untreated	Inoculated	
Corn					
Intake of digestible nutrients, kg/day					
DM	3	33.3	4.44	4.98	+12.3
OM	3	33.3	4.64	4.83	+4.0
Digestibility, %					
DM	4	75.0	64.85	68.71	+6.0
OM	3	66.7	68.17	71.09	+4.3
Sugarcane					
Intake, kg/day					
DM	11	27.3	7.69	8.74	+13.7
Intake of digestible nutrients, kg/day					
NDF	4	25.0	1.96	2.09	+6.4
Digestibility, %					
NDF	4	25.0	52.70	55.20	+4.7
Performance					
Feed efficiency	5	80.0	9.39	8.46	−9.9
ADG, kg/day	5	80.0	0.88	1.06	+21.1

[1]DM, dry matter; OM, organic matter; NDF, neutral detergent fiber; ADG, average daily gain.

Table 16. Summary of positive responses of silage inoculants on the performance of cattle fed corn and sugarcane silages in experiments carried out in Brazil.

The results of the current survey are encouraging regarding the impact of bacterial inoculants on animal performance in tropical conditions. However, although the mean and median values for most variables measuring animal performance were very similar (which may indicate normal distribution of the data), this occurred because of the lack and/or low number of studies evaluated. Therefore, some caution should be taken when interpreting this data, as well as the great frequency of positive responses found, which is likely attributed to the low number of studies evaluated.

Regarding the factors responsible for enhancing animal performance, certainly improvements in DM digestion are closely linked to greater growth performance. In a review of the literature from 1985 to 1992, animal performance improved in 9 of 16 trials when inoculation improved DM digestion, but only 2 of 15 trials when digestion was not significantly affected [50].

In our survey, we did not observe a relationship between DM digestibility and growth performance, because the number of studies evaluated was quite low. However, there are other hypotheses related to the improvement of animal performance. The first suggests that improvements in silage quality could lead to increased animal performance. The second suggests that silage inoculants may provide a probiotic effect by inhibiting detrimental microorganisms in the silage and rumen, or by producing beneficial substances that may enhance the functioning of specific microbial populations in the rumen, leading to an increase in animal performance [47].

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Item[1]	Number of treatments		Mean		Difference, %
	Total	Positive responses, %	Untreated	Inoculated	
Corn					
Intake, kg/day					
DM	7	57.1	1.07	1.21	+ 13.0
OM	4	50.0	0.96	1.02	+ 6.4
NDF	4	50.0	0.41	0.44	+ 8.0
CP	4	50.0	0.13	0.14	+ 7.8
Digestibility, %					
CP	5	20.0	60.50	66.00	+ 9.1
Performance					
ADG, kg/day	2	50.0	0.20	0.21	+ 4.0
Grass					
Intake, % BW					
DM	6	50.0	1.43	1.97	+ 38.0
Digestibility, %					
DM	11	36.4	49.95	54.14	+ 8.4
NDF	9	33.3	54.71	60.64	+ 10.8
CP	11	9.1	53.07	68.76	+ 29.6
Sugarcane					
Intake of digestible nutrients, kg/day					
NDF	1	100.0	0.35	0.48	+ 36.9
Digestibility, %					
NDF	1	100.0	57.50	64.90	+ 12.9

[1]DM, dry matter; OM, organic matter; NDF, neutral detergent fiber; CP, crude protein; ADG, average daily gain; BW, body weight.

Table 17. Summary of positive responses of silage inoculants on the performance of sheep fed corn, grass, and sugarcane silages in experiments carried out in Brazil.

A probiotic can be defined as a culture of live microbes, that when fed to the animals, beneficially affects the host by improving the properties of the native gut microflora [48]. Indeed, a recent study displayed greater microbial protein synthesis in lambs fed silage inoculated with *L. buchneri*, applied either alone or associated with *L. plantarum* in corn silage [51], which is likely related to changes in the microbial community in the rumen.

3. Implications

The data summarized from Brazilian studies displays a recent increase in interest from researchers addressing bacterial inoculants as an alternative to improve silage quality. But although the number of studies remains quite low compared with the international literature, data of this survey revealed some trends for improved fermentation and nutritive value regarding the group of bacterial inoculant used at ensiling and crop.

Considering an overall mean, homolactic inoculation unaffected DM losses in corn, grass, HMC, and sorghum silages, but reduced DM loss in alfalfa silages. However, an unexpected increase in aerobic stability of grass silage was reported due to homolactic inoculation. The greater frequency of positive response was also observed for grass silages when treated with ^{ho}LAB. Conversely, heterolactic inoculation revealed to be more interesting than homolactic inoculants to reduce fermentation losses in sugarcane silage, and positive responses were found most often. In addition, enhanced aerobic stability was reported for corn and HMC silages when they were treated with ^{he}LAB. Overall, the results of the current survey regarding fermentation patterns of inoculated silages are encouraging, mainly for grass and sugarcane silages. Otherwise, the impact of bacterial inoculant on silage quality (i.e., fermentation patterns, chemical composition, and nutritive value) appeared to diminish as the quality of ensiled crop increased.

Despite of animal performance and considering the overall means, inoculation consistently depressed DM intake in cattle fed corn, grass, and sugarcane silages, but DM intake increased in sheep due to inoculation. There were not a consistent effect of bacterial inoculants on silage digestibility, which largely varied depending the animal and crop evaluated. Conversely, cattle fed inoculated sugarcane silage had a greater frequency of positive response on ADG. The performance of animals consuming inoculated silages has been investigated in Brazil only a few times, but the data suggest a greater impact of bacterial inoculants on DM intake and weight gain in cattle and sheep than that indicated under temperate conditions. However, the number of studies evaluating animal performance still remains quite low, especially for dairy cows fed inoculated silage, and this survey did not provide a definitive conclusion about the effect of bacterial inoculants on animal performance (cattle and sheep).

Finally, we need caution to interpret the data of the current survey because the potential of bacterial inoculants measured by studies containing positive responses were highly variable and deeply associated with number of studies. Hence, a greater frequency of positive responses was often observed when there were a low number of studies evaluated. Additionally, positive responses were clearly impacted by the group of microorganisms (homo and heterofermentative LAB) and it determined the success of bacterial inoculant applications in silage. In this way, the compatibility between the plants and microorganisms used at ensiling should be taken into account in further studies, as well as its applicability on farm. In addition, further studies may consider assessing animal performance and sanitary aspects related to the use of bacterial inoculants since there is a lack of data about it.

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