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EVALUATION OF FERMENTATION PROGRESS DURING STORAGE OF MILLET STOVERS SILAGE BASED ON pH-INDICATORS

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ABSTRACT: This study aimed at evaluating the fermentation levels of pearl millet [*Pennisetum Glaucum* (L.) R. Br] stovers silage during storage based on pH evolution. A completely randomized experimental design in a 6×2×2 factorial scheme with three replications for each treatment was used to evaluate three factors (6 cultivars, 2 different cutting stages, and with or without salt addition to the cultivars). The silages were prepared in plastic bags and stored for 60 days at room temperature. The results revealed that the pH values of the treatments were significantly (P<0.05) higher on the first day than in the other periods and a rapid drop in pH, with significant differences (P<0.05), to levels below 4 was obtained on the third day of storage for the majority of local Sadoré and Siaka Millet silages (Niger). Four types of pH evolution were recorded and the variation was statistical significant among cultivars. Also, analysis of the relationships between pH, chemical composition parameters and In Vitro Digestibiliy of Organic Matter (IVDOM) showed that increasing pH values were associated with increasing Dry Matter content of stovers before silage (DM_BE), Dry Matter content of silages (DM_S), Neutral Detergent Fiber (NDF), Acid Detergent Fiber (ADF), Acid Detergent Lignin (ADL) values and decreasing Crude Protein (CP), Metabolizable Energy (ME), IVOMD, and Ash values. However, the pH values obtained for all silages showed that all the millet stovers used were suitable for silage. At the maturity stage, it is thus possible to use the grain for human consumption and to ensile the stovers for animal feed. This study also shows that monitoring the pH in the silo makes it possible to evaluate the quality of the fermentations to avoid losses on the farms.

Keywords: Dual-purpose varieties; Harvesting stage; Monitoring of pH; Silage; Stovers conservation.

INTRODUCTION

Rain-fed agriculture and agro-pastoralism systems are the source of employment for about 80-90% of the population in the West Africa Sahel (Bado et al., 2021). However, crop production alone cannot meet basic human needs for nutrition or income generation. The demand for livestock products is increasing due to an increasing human population and the dietary changes that are driven by demographic changes of an urbanizing society (Thornton, 2010; Bado et al., 2021). Livestock plays a crucial role, in generating income, nutrition and creating the means to purchase more diverse diets, and paying for health and other household needs. However, the increasing demand for feed and seasonal shortage in feed and water, particularly during the long dry season remain the major constraints limiting livestock production (Lamega et al., 2021; Amole et al., 2022).

In Niger, nearly 66 % of the national livestock, mainly small ruminants, are raised in agricultural areas, in a sedentary mode. The immediate consequence is chronic underfeeding of the animals, resulting in a decline in their performance (Sourabie et al., 1995). Increasing feed availability for livestock and the demand for food for the population is possible through the improvement of both grain and biomass production of the dominant crops, such as millet (Ouendeba and Siaka, 2004; Malam et al., 2019).

But the main constraint of millet stovers in small farming systems is the gradual loss of their nutritional value, ingestibility, and digestibility during storage (Cai et al., 2020). These losses can be controlled by managing storage conditions. Silage is one of the technologies that have been found to preserve the quality of crop residues (AFSSA, 2004). However, during the silage process, a number of parameters fluctuate and include: pH, temperature, types and numbers of microorganisms, fermentation products and chemical composition (Cherney and Cherney, 2003). Thus, when the pH value drops rapidly, the wet fodder is preserved from spoilage microorganisms (Rooke and Hatfield, 2003).

Therefore, monitoring acidity fluctuation over time is the simplest and quickest way to assess the fermentation and final product quality of silage (Sprague and Taylor 1977; Decruyenaere et al., 2008; Ishiaku et al., 2020). But how does pH relate to the quality of the final silage product? According to Cherney and Cherney (2003), the chemical composition of the silage influences its nutrient quality. Furthermore, assessing the progression of fermentation during the silage process

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provides information on its relative success with different types of treatments (Sprague and Taylor, 1977; Cherney and Cherney, 2003; Ahmed et al., 2022).

Therefore, this study was carried out to assess the progress of fermentation during the ensiling process by analyzing the evolution of pH as function of cultivars, cutting stage and adding salt. Specifically, the aim is to: i) Characterise the different types of pH evolution in millet stover silages; ii) Compare the different types of pH evolution in millet stover silages according to cultivars, cutting stage and salt addition; and iii) determine the relationship between pH and chemical composition parameters of millet stover silages.

MATERIALS AND METHODS

Study area and material

The experiment was conducted in Niger at the International Crops Research Institute for the Semi-Arid Tropic (ICRISAT) research station at Sadoré, which is located between 13° 14' N and 2° 16' E. During the experiment, the average minimum and maximum temperatures were 22.85±4.05°C and 36.42±3.91°C respectively, with an overall average of 29.64±3.51°C. In this period, the total annual rainfall was 715.29 mm. The pearl millet stover was obtained from pearl millet grown at the Sadoré experimental station from 29 June to 14 November 2019. The number of days between the flowering and maturity stage varied between 31 to 55 days depending on the variety (Table 1).

Table 1 - Number of days between sowing and harvest (NDAS) according to cultivar and cutting stage.

Cultivars	Stage	NDAS	NDFM
Chakti	Flowering	45	31
	Maturity	76	
Millet of Siaka	Flowering	73	42
	Maturity	115	
ICRI-Tabi	Flowering	72	40
	Maturity	112	
Alambana	Flowering	71	55
	Maturity	126	
Maywa	Flowering	88	50
	Maturity	138	
Local Sadoré	Flowering	75	41
	Maturity	116	

NDAS: Number of Days After Sowing; NDFM: Number of Days between Flowering and Maturity.

Experiment design

A completely randomized experimental design was used in a 6x2x2 factorial scheme with three replications. Three factors were evaluated: cultivar, stage of cutting stover, and inclusion or not of salt (i) the cultivar, with six modalities consisting of four improved dual-purpose varieties Chakti, ICMV167005 (Millet of Siaka), ICMV167006 (ICRI-Tabi), ICMH177111 (Alambana) and two local varieties (Maywa and Local Sadoré). The four improved dual-purpose varieties were selected because they have been approved and evaluated in the field, while the local varieties were selected because they are widely used in the study area and generally in all Niger. (ii) Two stages of cutting were considered: flowering and maturity. Silages obtained from stover cut at the flowering stage were used as a basis for comparison, as this stage is probably the best time to ensile millet stover (Morales et al., 2015). (iii) The addition of salt was also studied in two ways (without salt and with salt). Salt was used at a rate of 10 kg per ton of fresh silage (Tamboura et al., 2005).

The treatments tested consisted of a combination of different modalities of the three factors, i.e. 6x2x2 giving 24 treatments. For each treatment, three repetitions were considered (Kim and Adesogan, 2006), giving 72 repetitions for the whole trial.

Silage- making process and data measurements

The stovers were harvested, using a machete, at different periods depending on the cultivar and desired stage of cutting for ensiling (Table 1). The stovers were chopped using a chopping machine, to obtain particles of approximately 2 to 3 centimeters (Trevisoli et al., 2017; Kang et al., 2018; Ishiaku et al., 2020). A 20 kg of fresh material was used for each repetition (Morales et al., 2011). Thus, 20 kg of stover was put and compacted, using a manual compactor, in 100 kg capacity plastic bags. The filled plastic bag was hermetically sealed with string and tape. Then introduced in a second bag to improve the anaerobic conditions. The plastic bags were kept in a covered area, at room temperature for 60 days (Costa et al., 2012). Before ensiling and at 60 days, when the plastic bags were opened, fresh silage samples were taken for dry matter determination in an oven at 65°C for 72 hours (Trevisoli et al., 2017) and stored in a freezer for laboratory pH measurements. The dried samples were then ground with a 1 mm grid for the determination of the chemical composition and the In Vitro Digestibility of Organic Matter (IVDOM).

The Direct pH and temperature of the silage were recorded on the 1st, 3rd, 45th, and 60th day of storage, using a Hanna instrument HI99161 portable pH meter, equipped with a penetration electrode for food products. The pH electrode FC2023 is equipped with an integrated temperature sensor for direct reading of both parameters. In this regard, Sprague and Taylor (1977), Cherney and Cherney (2003) and Shan et al. (2021) reported that pH and temperature during the fermentation process in experimental silos or on-farm silos can be monitored effectively using pH probe electrodes, for acceptable moisture levels.

For each repetition, the bag was perforated to place the pH meter electrode inside the silage. The flashing light disappears to indicate the stabilization of the device for pH and temperature readings. Thus, for each repetition, this operation was repeated three times, changing the position of the probe to take into account the heterogeneity inside the silage. Then averages were calculated for each repetition to find the pH and temperature corresponding to a given

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measurement period. At the end of each measurement, the pH meter probe was properly cleaned with distilled water and the hole formed at the introduction of the latter inside the bag was automatically and hermetically sealed with adhesive tape. For the laboratory pH measurement, 10 g of fresh silage was mixed with 100 ml of distilled water using a blender. After filtration, the laboratory pH was measured with a "WTW Multi 9620 IDS pH meter". The chemical composition and IVDOM were determined by Near Infra- Red Spectrometry (NIRS) at the International Livestock Research Institute (ILRI) Laboratory in Burkina Faso.

Statistical analysis of data

The analysis of the pH evolution pattern was performed using SPSS chi-deux tests according to the different factors.

A principal component analysis (PCA) was carried out using XLSTAT 2014. Relationships between Factors (cultivars, cutting stage and adding salt), direct pH, Laboratory pH, IVDOM and some parameters of the chemical composition of silages were established. The DM content was determined according to the ratio between the weight of the dried matter (Pf) and the weight of the initial sample (Pi) (Cherney and Cherney, 2003). According to the factors and their modalities, the frequencies of the different types of pH evolution have been calculated using the following formula:

$$\boldsymbol{FRtm} = \frac{\boldsymbol{NOTEm}}{\boldsymbol{TNSf}} \times \boldsymbol{100}$$

FRtm: Frequency of the type of evolution in the modality under consideration;

NOTEm: Number of Occurrence of the Type of evolution in the modality;

TNSf: Total Number of Samples for the considered factor.

RESULTS

Characteristics of pH evolution in millet stover silages

The stovers were ensiled at DM levels ranging from 19.06 to 41.69% (Table 2). In general, as a function of the storage period, the pH decreased from day 1 to day 60 in all treatments (Figures 1 and 2). The pH values of the treatments were significantly higher on the first day than in the other periods. A rapid drop in pH, with significant differences, to levels below 4 was obtained on the third day of storage for the majority of the Local Sadoré and Millet of Siaka silages (Table 2). Overall, from this period onwards, the pH evolved until the 60th day of storage with a tendency to stabilize for the majority of silages (Table 2).

The pH values on the first day of storage did not vary significantly between treatments, except Maywa, which was significantly lower than the others for all the different interactions between factors, and Local Sadoré for all the silages at the maturity stage with and without salt. On the forty-fifth day of storage, for all silages at the flowering stage with and without salt, there were no significant differences between all cultivars for the pH means. For maturity stage silages with and without salt, the pH of Chakti and Maywa varieties was significantly lower than the others. At day 60 of storage, significant differences were recorded between treatments. For silages at the flowering stage with salt, ICRI-Tabi, Alambana, Chakti, and Millet of Siaka recorded the lowest pH values, while for silages at the flowering stage without salt, Millet of Siaka, ICRI-Tabi, Local Sadoré, and Alambana gave the most acidic silages. For maturity silages with salt, the varieties Chakti, Maywa, and Millet of Siaka recorded significantly lower pH values than the other varieties, while for maturity silages without salt, the same result was obtained with Chakti and Local Sadoré (Table 2).

Comparison between the different types of pH evolution in millet stover silages according to cultivars, cutting stage, and salt addition

Four types of pH evolution over time were identified, one normal (type 1) and three atypical (types 2-4). The type 1 showed a continuous fall in pH until stabilization, from the 1st to the 60th day of storage. The second type recorded pH drop from the 1st to the 3rd day, followed by a slight increase until the 45th day, then a slight decrease until the 60th day. Type 3 had a pH drop from the 1st to the 45th day, followed by a slight increase until the 60th day while the type 4 registered a pH falls from the 1st to the 3rd day, followed by an increase until the 60th day (Figures 1 and 2). Overall, type 1 evolution of pH was dominated in silages, followed by type 3 and then type 2, whereas the type 4 is weakly recorded (Table 3). However, this global analysis of the type of pH evolution hides several disparities depending on the stage of cutting, the addition of salt, and the cultivar. Regarding the cutting stage, although there was no significant difference (P > 0.05), type 2 has was dominated in silages from stover cut at flowering stage, while in silages from stover cut at maturity stage, type 1 dominated. Type 4 was only record by silages from stover cut at maturity (Table 3). There was no significant difference among silage from pearl millet stover with inclusion or not of salt (P > 0.05), but in these silages the type 1 was dominated. Specifically, in silages without salt type 3, was dominated compared to type 2, while the opposite was observed with silages with inclusion of salt (Table 3). The type of pH evolution was statistically different between cultivars (P < 0.05). Chakti silages have all recorded a type 1 pH evolution (Table 3). 50 % of silages from millet of Siaka showed type 2 pH evolution, while the 50% remaining were equally distributed in, types 1 and 3 pH evolution. In silages of ICRI-Tabi pH evolution was in order to types 1, 2 and 3. In the Alambana silages, the pH evolved in order according to type 1 and type 3. As for Maywa silages, the pH evolution was type 1 and type 3. The type 4 pH evolution was only recorded in the Local Sadoré silages (Table 3).

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Table 2 - Comparison of Dry Matter and pH values according to treatment and conservation period

		DM				pH					
Factors interaction		**Conservation period (day)**									
Cutting Stage × Adding Salt	**Cultivar**	**1**	**60**	***P*-value**	**Sign. level**	**1**	**3**	**45**	**60**	***P*-value**	**Sign. level**
Flowering × With Salt	Alambana	20.93^{cB}	23.57^{abA}	0.015	*	6.39^{aA}	4.42^{abcB}	4.33^{aB}	3.67^{bcB}	0.002	**
	Chakti	22.91^{bcA}	21.77^{bB}	0.038	*	6.62^{aA}	5.71^{aB}	4.31^{aC}	3.76^{bcC}	0.000	***
	ICRI-Tabi	27.75^{aA}	23.02^{bB}	0.022	*	6.67^{aA}	4.18^{bcB}	4.31^{aB}	3.64^{cB}	0.000	***
	Local Sadoré	26.03^{abA}	21.21^{bB}	0.049	*	6.49^{aA}	3.45^{cC}	4.38^{aB}	4.09^{abB}	0.000	***
	Maywa	26.05^{abA}	26.11^{aA}	0.969	NS	5.89^{bA}	5.08^{abB}	4.15^{aC}	4.30^{aC}	0.000	***
	Millet of Siaka	19.63^{cB}	22.19^{bA}	0.006	**	6.58^{aA}	3.56^{cC}	4.03^{aB}	3.80^{bcBC}	0.000	***
	P-value	0.000	0.000			0.001	0.002	0.517	0.002		
	Significance level	***	***			**	**	NS	**		
Flowering × Without Salt	Alambana	19.51^{aA}	17.36^{bA}	0.128	NS	6.71^{aA}	5.78^{aB}	4.11^{aC}	4.14^{bcC}	0.000	***
	Chakti	22.17^{aA}	17.77^{bA}	0.138	NS	6.86^{aA}	5.67^{aB}	4.65^{aC}	4.54^{aC}	0.000	***
	ICRI-Tabi	22.96^{aA}	22.15^{aA}	0.561	NS	6.59^{aA}	4.55^{bcB}	4.27^{aB}	3.97^{bcB}	0.000	***
	Local Sadoré	24.61^{aA}	19.24^{abA}	0.051	NS	6.71^{aA}	3.61^{cC}	4.22^{aB}	3.97^{bcBC}	0.000	***
	Maywa	22.47^{aA}	23.59^{aA}	0.316	NS	5.87^{bA}	5.39^{abB}	4.30^{aC}	4.31^{abC}	0.000	***
	Millet of Siaka	19.06^{aA}	20.74^{abA}	0.055	NS	6.74^{aA}	3.76^{cB}	4.43^{aB}	3.87^{cB}	0.000	***
	P-value	0.057	0.003			0.000	0.000	0.109	0.001		
	Significance level	NS	**			***	***	NS	**		
Maturity × With Salt	Alambana	38.90^{abA}	37.75^{abA}	0.684	NS	6.39^{abA}	5.56^{aB}	5.26^{aB}	5.09^{aB}	0.003	**
	Chakti	31.65^{dA}	26.46^{dB}	0.004	**	6.62^{aA}	5.39^{aB}	4.34^{cC}	3.98^{cC}	0.000	***
	ICRI-Tabi	41.69^{aA}	41.06^{aA}	0.782	NS	6.64^{aA}	5.56^{aB}	4.83^{bC}	4.78^{abC}	0.000	***
	Local Sadoré	33.33^{bcdA}	29.71^{cdB}	0.035	*	5.93^{bA}	5.13^{abB}	4.56^{bcC}	4.72^{abC}	0.000	***
	Maywa	31.90^{cdA}	32.97^{bcA}	0.187	NS	5.12^{cA}	4.79^{bB}	4.37^{cC}	4.29^{bcC}	0.000	***
	Mil of Siaka	37.47^{abcA}	39.41^{aA}	0.314	NS	6.11^{abA}	4.77^{bB}	4.46^{bcB}	4.40^{bcB}	0.000	***
	P-value	0.000	0.000			0.000	0.000	0.000	0.000		
	Significance level	***	***			***	***	***	***		
Maturity × Without Salt	Alambana	30.26^{bcA}	29.83^{bcA}	0.747	NS	5.92^{cA}	5.37^{abB}	4.70^{abC}	4.65^{bC}	0.000	***
	Chakti	30.96^{bcA}	24.87^{cB}	0.010	*	6.75^{aA}	4.92^{bB}	4.30^{bBC}	4.18^{cC}	0.000	***
	ICRI-Tabi	39.61^{aA}	36.44^{aA}	0.356	NS	6.60^{abA}	5.81^{aB}	4.96^{aC}	5.46^{aBC}	0.000	***
	Local Sadoré	29.44^{cA}	27.22^{cB}	0.005	**	6.08^{bcA}	3.58^{cC}	4.31^{bBC}	4.47^{bcB}	0.000	***
	Maywa	28.56^{cA}	30.91^{abcA}	0.287	NS	5.37^{dA}	5.07^{abAB}	4.83^{aBC}	4.60^{bC}	0.003	**
	Millet of Siaka	35.12^{abA}	35.98^{abA}	0.562	NS	6.24^{abcA}	4.81^{bB}	4.30^{bC}	4.57^{bBC}	0.000	***
	P-value	0.000	0.000			0.000	0.000	0.001	0.000		
	Significance level	***	***			***	***	**	***		

In each column and according to parameter and interaction between the modalities, means with at least one similar lower case letter in superscript are not statistically different at 5% level. In each row, according to parameter, means with at least one similar capital letter in superscript are not statistically different at the 5% level. Flowering x With Salt: Interaction between Flowering and With Salt ; Flowering x Without Salt : Interaction between Flowering and Without Salt ; Maturity x With Salt : Interaction between Maturity and With Salt; Maturity x Without Salt : Interaction between Maturity and Without Salt; Sign.: Significant; NS: Not Significant; *: *P-value < 0.05;* **: *P-value < 0.01;* ***: *P-value < 0.001.*

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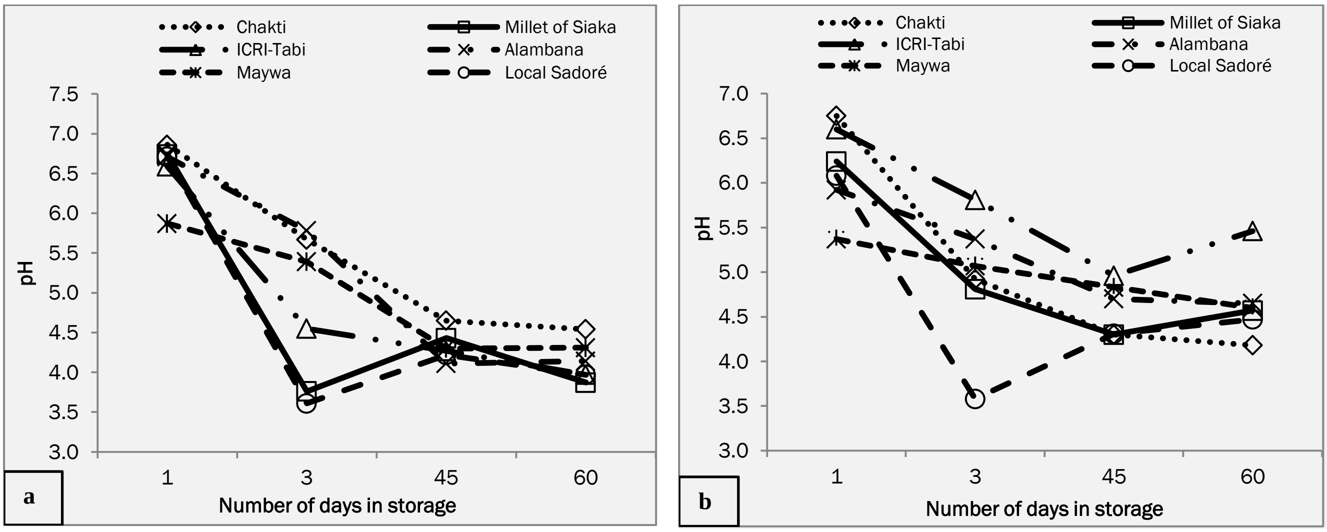

Figure 1 - pH of silages over time, (a) silages from stover cut at flowering stage, without addition of salt, and (b) silages from stover cut at maturity, without addition of salt.

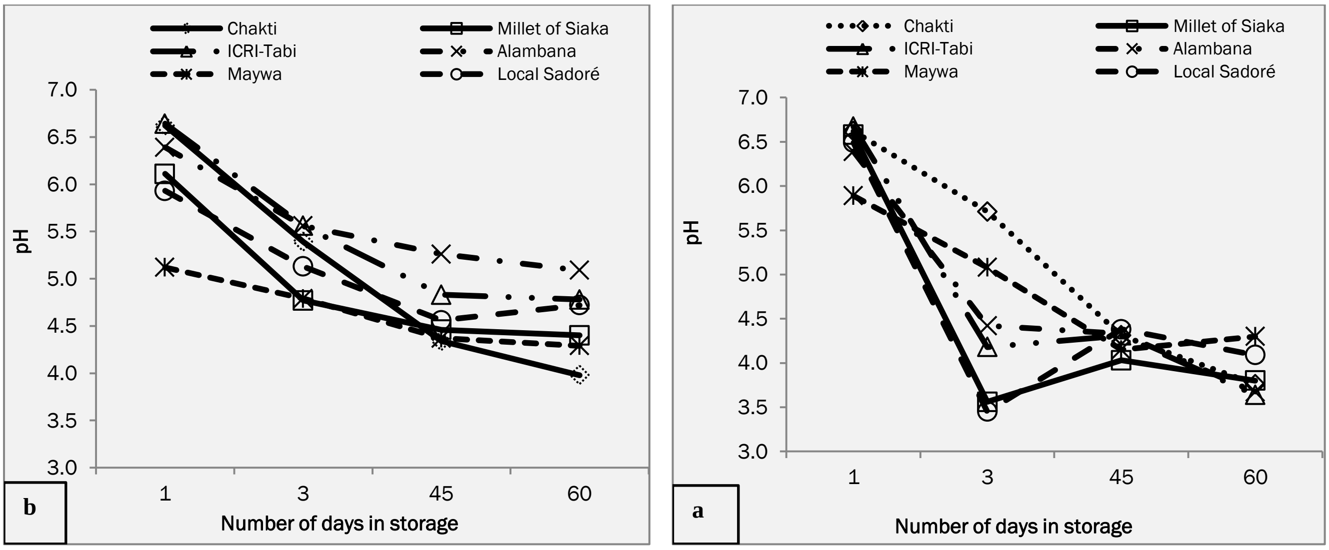

Figure 2 - pH of silages over time, (a) silages from stover cut at flowering, with added of salt (a) and (b) silages from stover cut at maturity, with added salt (b).

Table 3 - Mean of Dry Matter content at first day, mean of pH at day 60 and frequencies of different types of pH evolution according to variety, cutting stage and salt addition

Factors	Modalities	DM_BE_M (%)	pH_f	Types of pH evolution				
				Type 1 (%)	Type 2 (%)	Type 3 (%)	Type 4 (%)	Total (%)
Cultivars	Chakti	26.92^{a}	4.12^{a}	100	0	0	0	100
	Millet of Siaka	27.82^{a}	4.16^{ab}	25	50	25	0	100
	ICRI-Tabi	33.00^{b}	4.46^{c}	50	25	25	0	100
	Alambana	27.40^{a}	4.39^{c}	75	0	25	0	100
	Maywa	27.25^{a}	4.37^{c}	50	0	50	0	100
	Local Sadoré	28.35^{a}	4.31^{bc}	0	50	25	25	100
	P-value	0.000	0.000			0.000		
	Significance level	***	***			***		
Cutting stages	Flowering	22.84^{a}	4.00^{a}	33.3	41.7	25	0	100
	Maturity	34.08^{b}	4.60^{b}	66.7	0	25	8.3	100
	P-value	0.000	0.000			0.062		
	Significance level	***	***			NS		
Adding salt	Without salt	27.06^{a}	4.39^{b}	41.7	16.7	33.3	8.3	100
	With salt	29.85^{b}	4.21^{a}	58.3	25	16.7	0	100
	P-value	0.000	0.000			0.53		
	Significance level	***	***			NS		
Global Frequencies				50	20.8	25	4.2	100

DM_BE_M : Mean dry matter content at first day ; pH_f : Mean pH at day 60; NS: Not Significant; ***: *P-value < 0.001.*

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Relationships between pH and pararameters of chemical composition and In Vitro Digestibily of Organic Matter of millet stover silages

The results of the PCA between the factors (Cultivar, Salt addition, and Cutting stage), the pH was measured directly on the 60th day of storage (Direct pH), the pH was measured in the laboratory on the 60th day of storage (Laboratory pH), Chemical composition parameters (DM_BE, DM_S, CP, Ash, NDF, ADF, ADL, ME) and In Vitro Digestibility of Organic Matter (IVDOM) was indicated that there were strong significant correlations between some parameters (Table 4 and Figure 3). Figure 3 shows that all parameters are well represented on the first two axes of the plan, which together account for 81.02% of the information. Each parameter was well correlated positively or negatively with one of the two axes. Thus, all the studied parameters are well correlated with axis 1 (70.21% of the information), except for Salt addition and cultivar which are best represented on axis 2 (10.81% of the information). The analysis of Figure 3 and Table 4 shows a strong positive correlation between direct and laboratory pH. Two groups of correlation can be distinguished according to the analysis of the evolution of pH values on axis 1. The analysis of these relationships showed that the increase in pH values was associated with the increase in DM_BE, DM_S, NDF, ADF, and ADL. All these parameters move in opposite directions with CP, EM, MDMVI, and ash. The increase in these parameters is caused by the decrease in pH values. The analysis of Table 4 and Figure 3, also allows us to deduce that all the parameters of the chemical composition and the IVDOM studied can contribute to characterise the cutting stage, while only Ash contributes to separate the groups according to the addition of salt and no parameter allows identifying the cultivar groups. The results also show that there are no significant interactions between the three factors compared in pairs (Table 4).

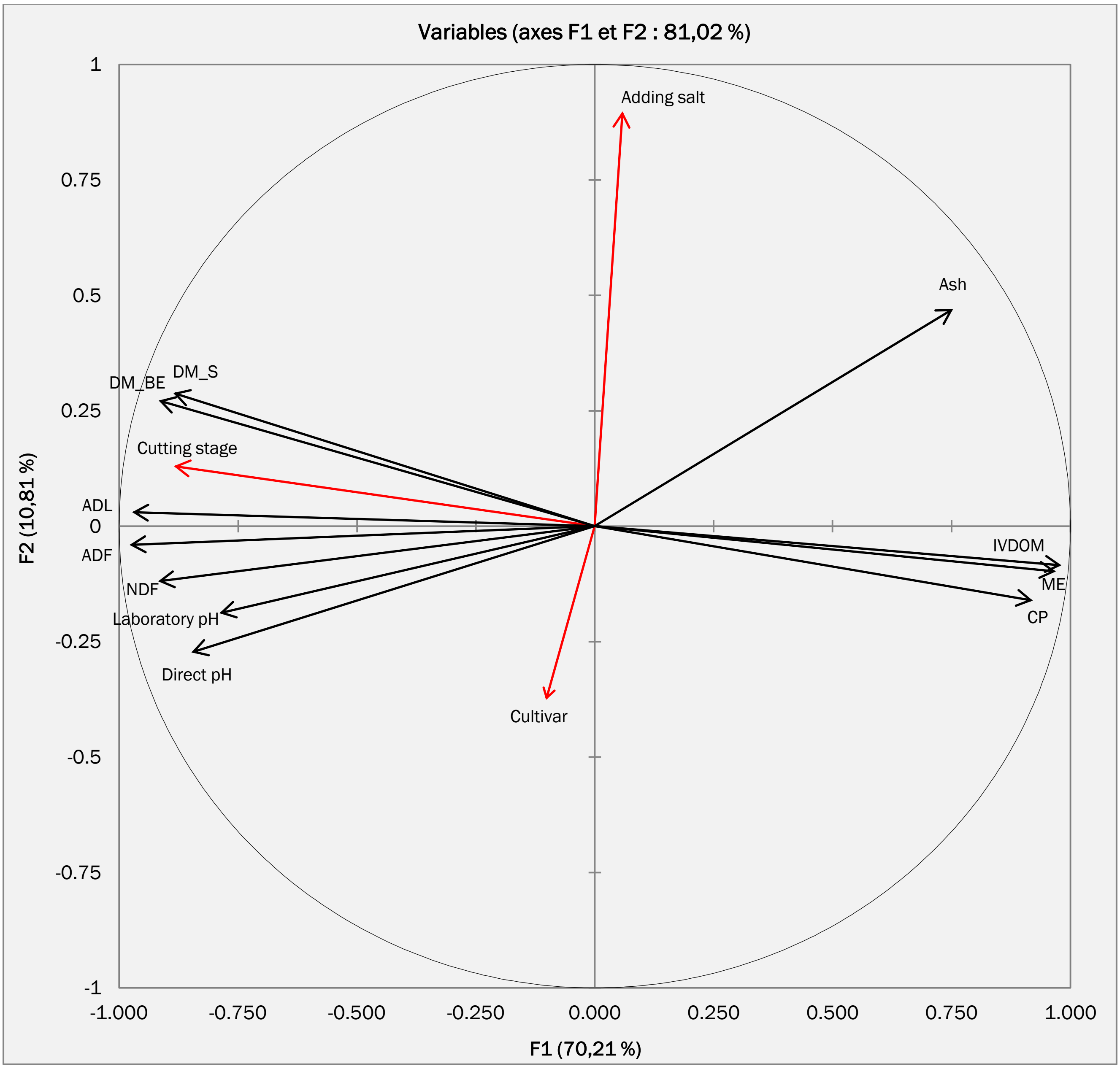

Figure 3 - Spatial representation of the relationships between some silage parameters. Direct pH: Hydrogen potential measured directly in silages at 60 days of storage; Laboratory pH: Hydrogen potential of silages measured in the laboratory after 60 days of storage; DM_BE: Dry Matter content of stovers before silage; DM_S: Dry Matter content of silages; CP: Crude Protein; NDF: Neutral Detergent Fiber; ADF: Acid Detergent Fiber; ADL: Acid Detergent Lignin; ME: Metabolizable Energy; IVDOM: In Vitro Digestibility of Organic Matter.

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Table 4 - Matrix of correlation between factors, pH, different parameters of chemical composition and In Vitro Digestibility of Organic Matter

	Cultivar	AS	CS	Ash	CP	NDF	ADF	ADL	ME	IVDOM	Direct pH	DM_S	DM_BE	Laboratory pH
Cultivar	1													
AS	-0.024	1												
CS	0.024	0.000	1											
Ash	-0.210	0.447*	-0.582**	1										
CP	0.003	-0.028	-0.803**	0.600**	1									
NDF	0.081	-0.221	0.751**	-0.730**	-0.924**	1								
ADF	0.062	-0.117	0.830**	-0.716**	-0.908**	0.943**	1							
ADL	0.026	-0.088	0.920**	-0.709**	-0.915**	0.917**	0.967**	1						
ME	-0.115	-0.045	-0.837**	0.662**	0.922**	-0.913**	-0.967**	-0.964**	1					
IVDOM	-0.076	-0.013	-0.874**	0.689**	0.925**	-0.917**	-0.977**	-0.982**	0.994**	1				
Direct pH	0.204	-0.208	0.670**	-0.715**	-0.651**	0.707**	0.823**	0.750**	-0.766**	-0.772**	1			
DM_S	0.020	0.237	0.812**	-0.626**	-0.844**	0.727**	0.804**	0.825**	-0.844**	-0.855**	0.696**	1		
DM_BE	0.026	0.210	0.847**	-0.596**	-0.852**	0.758**	0.861**	0.871**	-0.888**	-0.896**	0.735**	0.918**	1	
Laboratory pH	0.206	-0.109	0.640**	-0.574**	-0.590**	0.610**	0.769**	0.680**	-0.705**	-0.716**	0.947**	0.660**	0.708**	1

*: The correlation is significant at the 0.05 level; **: The correlation is significant at the 0.01 level. AS: Adding Salt; CS: Cutting Stage; CP: Crude Protein; NDF: Neutral Detergent Fiber; ADF: Acid Detergent Fiber; ADL: Acid Detergent Lignin; ME: Metabolizable Energy; IVDOM: In Vitro Digestibility of Organic Matter; Direct pH: Hydrogen potential measured directly in silages at 60 days of storage; DM_S: Dry Matter content of silages; DM_BE: Dry Matter content of stovers before silage; Laboratory pH: Hydrogen potential of silages measured in the laboratory after 60 days of storage.

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DISCUSSION

Analysis of the different types of pH evolution in silages

The variation of pH during the storage period revealed four types of evolution reflecting the silage preparation conditions. Similar types of pH evolution in silage was reported by AFSSA (2004). The type 1 evolution suggests good silage preparation conditions and involve high concentration of soluble sugars in stover, which would allow sufficient production of lactic acid throughout the silage process. This probably led to a continuous decrease in pH from the first to the sixtieth day of storage. Indeed, Kang et al. (2018) reported that Plant carbohydrates are the substrates of the fermentation process, so their concentration in the original forage has a strong influence on the extent and nature of silage fermentation. Thus a high concentration of soluble sugars in fresh forage would result in good- quality silage as this would lead to high production of lactic acid which would be the basis for the gradual fall to stabilization of the pH of the silage. For type 2 pH evolution, after a drop in pH to values below 4 between days 1 and 3 corresponding to stability pH, the product becomes stable according to Adesogan and Newman (2021) because the development of undesirable microorganisms is inhibited. But, the slight increase in pH observed between the 3rd and 45th day suggests that during this period, the undesirable microorganisms have resumed their activities following air entry into the silos, because according to Adesogan and Newman (2021) the quality of the silage and consequently the stability pH can be maintained during storage as long as the silo remains sealed and air does not penetrate it. However, the rapid depletion of silo air and the acceptable level of soluble sugar in the product resulted in a slight decrease in pH between days 45 and 60. For pH evolutions of types 3 and 4, soluble sugars were probably insufficient in the stover to allow continuous production of lactic acid, to maintain a decrease of silage pH until the end of the process. Soluble carbohydrates play an important role in the fermentation process as they accelerate the acidification process. Their action, lead to a complete inhibition of any microbial activity (even that of the lactic flora) and of any proteolytic activity (Rooke and Hatfield, 2003; Baumont et al., 2011).

The rapid decrease of pH was observed in most of our silages between day 1 and 3 followed by pH stability during the storage period indicates a good fermentation process as was reported by Hassanat et al. (2007), Alhaag et al. (2019) and Hanif et al. (2020). Generally, pH is one of the quickest and simplest ways of evaluating silage quality (Ishiaku et al., 2020). The majority of pH values obtained in our silages at day 60 show that the making process would have gone well. This result shows good fermentation levels in these silages as was reported by Hassanat et al. (2007). Normally, good - quality silage is defined by a pH < 4 (Demarquilly et al., 1998).

Analysis of the different types of pH evolution in millet stover silages according to cultivars, cutting stage, and salt addition

For the cultivar, relatively low average pH (<4.5) and high frequencies of normal evolution (type 1) of pH recorded in the silages of Chakti (100%) and Alambana (75%) show that these silages were made under very good conservation conditions and that these cultivars produced stovers sufficiently rich in soluble sugars to make good silages. Moreover, the average pH of less than 4.5 obtained in the silages of the cultivars Millet of Siaka, Local Sadoré, Maywa and ICRI-Tabi, suggest that these cultivars produced stovers that are easily ensiled. However, the process of storage did not go well for some of the samples, which would have resulted in types 2, 3, and 4 of pH evolution in silages of these cultivars.

Regarding the cutting stage, normal (type 1) pH evolution dominated in silages at the maturity stage (66.7%), while atypical evolution of types 2 and 3 (66.7%) was predominantly recorded in silages at the flowering stage. However, the average pH (Table 3) obtained in silages at the flowering - stage (4.00) was statistically lower ($p<0.001$) than that recorded at the maturity stage (4.60). The frequency of atypical pH evolution in flowering stage silages would indicate inadequate conditions in the preparation and conservation process of these silages. On the other hand, the frequency of normal pH evolution obtained in silages from the maturity stage allows us to conclude that they were prepared under better conditions and that, according to the quality grid based on pH and DM proposed by Decruyenaere et al. (2008), the average pH of 4.6 and the average DM content of 34.08% (Table 3) would indicate that the stovers from the maturity stage are suitable for silage. The variations of the average pH according to the cutting stage can be explained, on the one hand, by the DM rate which is significantly higher in the silages of the maturity stage (Table 3), which would lead to a weak decrease of the pH in these silages and, on the other hand, by the fact that the stovers of the flowering stage contain more soluble sugars to allow a good decrease of the pH. These results are similar to those of Morales et al. (2014), who reported a variation in silage quality depending on the cutting stage. Costa et al. (2012), Morales et al. (2014 and 2015) noted an increase in silage pH and DM with plant age.

As for adding salt, silages with salt had a significantly lower pH ($p<0.001$) than silages without salt and a predominantly normal pH evolution (58.3%) compared to 41.7% in control silages. This shows that salt improved the fermentation conditions of the silages. These results corroborate those of Cai et al. (1997) who observed, by adding salt (NaCl), a decrease in pH and an improvement in silage quality. However, Shockey and Borger (1991) reported a negative effect of salt on silage storage. This can be explained by the differences in the amount of salt used in the two trials and the types of lactic acid bacteria present in the silage. Indeed, Amar and Manaa (2016) obtained three types of lactic acid bacteria growth depending on the salt dose.

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Analysis of Relationships between pH and parameters of chemical composition and In Vitro Digestibility of organic matter of millet silages

Our results showed that increasing pH values were associated with increasing DM_BE, DM_S, NDF, ADF, ADL values and decreasing CP, ME, IVOMD, Ash values. Indeed, Smith (1954), Seglar (2003) and DuPonte et al. (2016) reported that silage quality is closely related to the degree of acidity obtained during fermentation. According to Wilkinson et al. (2003) and Cai et al. (2020), low pH values were associated with stable, good-quality silages. Furthermore, hay and silage quality can be defined in several ways. It is associated with nutrients; protein, NDF, ADF and ADL, minerals, vitamins, lipids, carbohydrates, energy, and digestibility, and also sometimes animal production (Charmley 2001; Newman et al. 2009). Ephrem et al. (2015) reported that the CP content of the forage is the most important nutrient parameter to consider. Thus, the results of our study suggest that good quality silage is characterized by low pH, high CP, ME, IVOMD, Ash contents and low DM_BE, DM_S, NDF, ADF, ADL contents.

Also, CP, ME, NDF, ADF and ADL contents of silages were the most correlated with IVOMD. These results corroborate those of Yang et al. (2018) and Aïssa et al. (2021), who showed that nutrient digestibility and energy parameters are positively correlated with forage CP content, but negatively correlated with NDF and ADF content. These authors also showed that the CP, NDF and ADF contents of the forage most determine the digestibility of the nutrients.

CONCLUSION

This research revealed that the stover of six pearl millet varieties has the potential to provide good- quality silages based on their pH analysis as a function of DM before ensiling. Stovers cut at flowering gave the best silages. However, silages at the maturity stage are also suitable. This means that the grain can be used for human consumption and the stover silages for animal feed. The addition of salt at 1% has the benefit to improve the quality of silage. The dual-purpose pearl millet varieties Chakti and Siaka produced silage of better quality. However, the varieties of Local Sadoré and Siaka produced the most suitable stover for silage. On the other hand, this study has shown that the pH evolution in the silo can be monitored directly with an adapted pH meter in order to prevent silage fermentation problems. However, this study needs to be completed with fermentation profile of the silages.

DECLARATIONS

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Authors' contributions

HS KOROMBÉ contributed to protocol development, trial conduct, data analysis, manuscript writing, review, and validation. **VB BADO, N ABDOU, C UMUTONI** and **AS GOURO** participated in protocol development, data analysis, manuscript review, and validation of the manuscript. **A IBRAHIMA** participated in protocol development, data collection, review and validation of the manuscript.

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Competing interests

The authors declare that they have no competing interests.

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