
INFLUENCE OF WILTING DURATION ON MICROBIAL PROFILE OF GRASS-LEGUME SILAGE IN UMUDIKE, SOUTHEAST, NIGERIA

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ABSTRACT

*Silage production has been considered to be best suited forage conservation technique in the rainforest zone of Nigeria due to high relative humidity and unpredictable sunlight which made it challenging to cure a good quality hay in such environment. During late rainy period legumes usually available in abundant and it could be utilized to improve quality of predominant grass species that are generally characterized with declined nutritive quality when there is moisture stress. Therefore, this experiment was conducted to evaluate microbial profile of grass-legume silage mixtures as affected by wilting periods. Different proportions of grass-legume silages mixtures were wilted for 6 and 12 hours namely: wilting period at two levels (6 and 12 hours after chopping) and forage composition in five different constituents (sole *Panicum maximum* (Guinea grass), sole *Pennisetum purpureum* (Elephant grass), *Panicum maximum*-*Centrosema molle* (*Panicum***Centro*) mixture, *Pennisetum purpureum*-*Centrosema molle* (*Pennisetum***Centro*) mixture and sole *Centrosema molle* (*Centro*). The experiment was a factorial arrangement laid out in a Completely Randomized Design (CRD). The microbial contents of silage were determined at 12 weeks after commencement of ensiling. The result of the study revealed that under anaerobic condition, the LABs suspected isolates were 14 (37.83 %) out of 37 total bacteria which is lower than the LABs suspected isolates 21 (42 %) out of 50 total bacteria under aerobic condition. That of *Clostridium* species were observed to be lower than LABs under both conditions. But a similar trend to that LABs suspected isolates was maintained when compared along the anaerobic and aerobic condition. The study showed that wilting period favoured production of LABs than *clostridium* species, though there was in slight increase in their population under aerobic condition.*

Keywords: wilting period, silage mixture and microbial profile

INTRODUCTION

Production of good silage require use of forage material posse high dry matter yield with better fermentative substrates. Among many forage species found in the humid tropics, *Pennisetum purpurem* has been recognized to be promising and high yield grass, giving dry matter yield surpass other grasses (Nymbati *et al.*, 2010). This grass specie is usually rejected by ruminants while grazing, but readily accepted when chopped and stall fed (Azevedo *et al.*, 2012). This make it appropriate material for silage production in the humid zone. Although, low crude protein and low amount of fermentable carbohydrates in has been a limiting factor to it efficient utilization in silage production. Hence, there is need to improve its nutritive quality with inclusion of legume and possibly mix with highly fermentable carbohydrates like grain to enhance it silage quality (Binuomote *et al.*, 2019). Before commencement of ensiling process, wilting the chopped forage materials is considered beneficial to silage preservation as it rapidly reduces moisture content and enhance dry matter content to about 27-30 %, which inadvertently impede the growth of undesirable micro-organisms that negatively affect the ensiled materials. Knowing the best wilting period of an ensile material would greatly improve the quality of silage. Therefore, this study was conducted to examine effect of wilting period and forage composition on microbial profile of silage produced in Umudike rainforest zone of Nigeria.

MATERIALS AND METHODS

EXPERIMENTAL SITE

The experiment was carried out in the Pasture Unit of the Teaching and Research Farm, Michael Okpara University of Agriculture, Umudike, Abia State. Umudike is located on latitude 05° 21'N and longitude 07° 33'E with an elevation of about 112m above sea level. The location has an annual rainfall of 177-2000mm per annum (April - October) and a short period of dry season (November - March) with a relative humidity of about 50 - 90% and monthly temperature range of 17°C - 36°C (Meteorological station NCRCI, Umudike, 2018).

EXPERIMENTAL DESIGN

The experiment comprised of two factors namely: wilting period at two levels (6 and 12 hours after chopping) and forage composition in 5 different constituents (sole *Panicum maximum* (Guinea grass), sole *Pennisetum purpureum* (Elephant grass), *Panicum maximum*-*Centrosema molle* (Centro) mixture, *Pennisetum purpureum*-*Centrosema molle* (Centro) mixture and sole *Centrosema molle* (Centro). Thus, the experiment was a factorial arrangement laid out in a Completely Randomized Design (CRD). There were 10 treatment combinations replicated three times.

SOURCE OF EXPERIMENTAL FORAGE MATERIALS FOR SILAGE PREPARATION

Fresh forage materials were harvested manually from natural vegetation around Umudike environment in May 2021 for this study. The harvested forage materials include two varieties of grass species and one leguminous specie namely: *Panicum maximum* (Guinea grass), *Pennisetum purpureum* (Elephant grass) and *Centrosema molle* (Centro). The chopped materials were spread and allowed to wilt in a room with an average temperature of 27.5°C for 6 hours and 12 hours respectively to reduce moisture content. A total of ten (10) treatment combinations were fully filled and properly compacted by exerting pressure on the chopped materials in thirty buckets (mini silos). In addition, dried grains measuring 48g were added to the fills as a fermentable substrate. Thereafter, the silos were covered with black nylon and sands were placed on the top and closed with a steal tape.

Ensiling process of the forage materials

After the silos had been properly filled and covered with airtight material, ensiling commenced on May 23rd, 2021 and was completed within the first four weeks after the silos were filled. Thereafter, the silos were kept in a room at normal temperature for another 8weeks period of storage.

RESULTS AND DISCUSSION

Table 1 reveals the interaction effects of wilting period and forage composition on total bacteria count ($\times 10^6$ CFU) under anaerobic and aerobic condition using De man Rogosa sharpe (MRS) and egg yolk (EY). There were no significant differences in the mean values presented across all treatment groups except in EY under anaerobic condition, where sole Centro wilted for 12 hours had the highest value (2.00×10^6 CFU) and that of the three mixtures silage wilted for 12 hours recorded the lowest value (0.00×10^6 CFU). Meanwhile, in the table 2, it was observed that under anaerobic condition, the LABs suspected isolates were 14 (37.83 %) out of 37 total bacteria which is lower than the LABs suspected isolates 21 (42 %) out of 50 total bacteria under aerobic condition. That of Clostridium species were observed to be lower than LABs under both conditions. But a similar trend to that LABs suspected isolates was maintained when compared along the anaerobic and aerobic condition

CONCLUSION AND RECOMMENDATION

Silage produced from grass-legume mixtures had higher total bacterial count for Lactic acid forming bacteria than sole grass or sole legume. And wilting improved the total bacteria count in both conditions. It could be recommended that grass-legume silages wilted for 12 hours before ensiling process should be adopted by farmers for good quality silage production.

Table 1: Interaction effects of wilting period and forage composition on total bacteria count (x 10⁶ CFU) of silage produced under both conditions

Wilting period	Treatments Forage Composition	Anaerobic condition			Aerobic condition		
		Egg yolk	De Man shape	Rogosa	Egg yolk	De Man shape	Rogosa
6 hours	Sole Panicum	1.300 ^{abc}	8.100		1.200	5.100	
6 hours	Sole Pennisetum	1.467 ^{ab}	5.900		0.667	7.967	
6 hours	Panicum x Centro	0.967 ^{abc}	6.067		0.000	5.700	
6 hours	Pennisetum x Centro	0.500 ^{bc}	6.600		0.300	6.167	
6 hours	Sole Centro	0.100 ^{bc}	6.667		0.000	5.867	
12 hours	Sole Panicum	1.467 ^{ab}	8.900		0.467	3.900	
12 hours	Sole Pennisetum	0.000 ^c	7.067		0.080	4.600	
12 hours	Panicum x Centro	0.000 ^c	5.567		0.000	5.367	
12 hours	Pennisetum x Centro	0.000 ^c	8.800		0.000	5.300	
12 hours	Sole Centro	2.000 ^a	6.067		1.100	5.200	
SEM		0.283	1.150		0.307	0.871	

a,b,c: means in the same column with different superscripts are significantly different (P<0.05), SEM= Standard error of mean.

Table 2: Percentage occurrence of microbial isolated of silage produced under both conditions

Suspected isolates	Anaerobic condition Number of positive	Aerobic condition Number of positive	Total number of positive	Percentage (%)
<i>Lactobacillus brevis</i>	4	9	13	14.9
<i>Lactobacillus fermentum</i>	7	3	10	11.5
<i>Lactobacillus lactis</i>	2	5	7	8.0
<i>Lactobacillus casei</i>	1	4	5	5.7
<i>Clostridium perfringen</i>	3	3	6	7.0
<i>Clostridium septicum</i>	1	7	8	9.2
<i>Aspergillus niger</i>	4	4	8	9.2
<i>Aspergillus flavus</i>	9	2	11	12.6
<i>Rhizopus stolonizer</i>	3	1	4	4.6
<i>Rhodotorula species</i>	2	7	9	10.3
<i>Saccharomyces cerevisiae</i>	1	5	6	7.0
Total	37	50	87	100

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