Bacterial population in *Crotalaria ochroleuca* hay subjected to different drying management and storage times.

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Introduction: The haying of forage legumes is still a practice that is underused due to the management demanded in the drying process. *Crotalaria ochroleuca* is a legume that has been cultivated in large areas as a predecessor to the soybean crop due to its potential of controlling nematodes. The green mass that is produced can be preserved for later use in animal feeding. Crotalaria is an annual plant, with upright shrub growth, which can reach up to 2.0 meters in height, presenting average yield potential of 7 to 17 tons per hectare (t ha^{-1}) of dry matter (Amabile et al., 2000). It is known that, after the forage reaches the ideal dry matter for storage (above 800 g kg-1), possible losses are due to the action of microorganisms such as fungi and bacteria. Therefore, this study intended to evaluate the bacterial population in the hay of *Crotalaria ochroleuca* produced with and without turning in the field drying process and its storage times.

Materials and Methods: The experiment was conducted in the experimental area of the Professor Antonio Carlos dos Santos Pessoa Farm, belonging to the Center for Agricultural Sciences, State University of Western Paraná, whose campus is located in the municipality of Marechal Cândido Rondon - PR. Randomized blocks were employed as the experimental design, with plots subdivided in time and using two drying methods of management: turning and no turning (during dehydration of forage legume), with subplots being allocated 0 (baling), 30, and 90 days of storage time, with four repetitions. The planting was done in rows with 0.50 m spacing and 10 kg ha-1 of *Crotalaria ochroleuca* seeds. The plants were cut on the pre-flowering stage at 82 days after emergence, using a conditioner mower with free-finger beaters and production of 16,566.68 Kg ha^{-1} DM. After the crotalaria cut (February), the plants remained in the field for three days for dehydration. Turning management was performed on the second day, at 14:00. After reaching the ideal dry matter for storage (820 g kg-1), the dehydrated material was collected and stored on pallets, in a covered and ventilated barn. In total, 24 bales were stored, with the removal of 8 bales every 30 days for evaluation. After completing each storage period, the bales were evaluated, and samples were taken for microbiological laboratory analysis of bacteria (*Clostridium* sp., Enterobacteria, lactic acid bacteria, and total bacteria) to count the population of microorganisms, which were determined by culture techniques according to Silva et al. (2007). The results were submitted to statistical analysis using the Statistical Analysis System with the MIXED procedure of SAS® University Edition, where the Tukey test at 5% significance level was utilized.

Results and Discussion: There was no interaction between drying systems and storage times (Table 1). Regarding the management, there were no differences in the population of microorganisms with turning and no turning of plants in sun drying. A high population of the bacterial genus Clostridium 7.08 log CFU and a smaller population of BAL were observed. The evaluated storage times did not change the population of Clostridium, lactic acid bacteria, and total bacteria. However, the enterobacteria population decreased with storage time, going from 6.42 at 30 days to 4.14 log CFU g at 90 days. This microorganism is a facultative anaerobic bacterium

(grows in aerobiosis and anaerobiosis), glucose fermenter, and reduces nitrate to nitrite present in plants (Rhoden, 1974), therefore, it is undesirable in the haying process. This effect of higher enterobacterial rates at 30 days of storage may be explained by the inhibition of this agent during more extended storage periods, due to the pH effect and absence of oxygen present in the samples (França et al., 2015), which slows the growth of these bacteria in the hay. What promoted these lower rates of microorganisms in the hay during the storage process was the effect of rapid dehydration and decreased cellular breathing of the plants during the field drying stage, which favored the paralysis of the metabolic processes of the plant and, as a result, that of the microorganisms in the hay (Collins and Coblentz, 2007). Due to its decreasing amounts of enterobacteria during storage, it furthered to produce hay with less contamination and higher sanitary quality. Although not statistically significant, the other evaluated bacteria, that is, *Clostridium* sp., lactic acid bacteria, and total bacteria, were present during the hay storage process.

Table 1. Population of microorganisms (log UFC g-1) in *Crotalaria ochroleuca* hay submitted to drying managements and three storage times

	Turning		Time (days)			Average	SEM	P- value [§]		
	Yes	No	30	60	90			V	T	V*T
CLOS	7.32a	6.84a	7.05a	7.16a	7.04a	7.08	0.1556	0.0653	0.9529	0.1874
ETB	5.31a	5.26a	6.42a	5.30b	4.14c	5.28	0.2358	0.8769	0.0002	0.5413
LAB	4.46a	4.53a	4.62a	4.19a	4.69a	4.50	0.1034	0.7879	0.0803	0.7481
TB	7.09a	7.05a	7.56a	6.73a	6.92a	7.07	0.1661	0.9154	0.1547	0.3574

[§]V – Turning effect; T – Effect of storage time; V*T – Effect of interaction between turning and storage times. CLOS – *Clostridium sp.*, ETB - Enterobacteria, LAB – Lactic acid bacteria, TB – Total bacteria. SEM= standard error of the mean. Means followed by lowercase letters in the row differ from each other according to the Tukey test ($P \leq 0.05$).

Conclusions: The drying management does not alter the population of microorganisms evaluated during the storage of *Crotalaria ochroleuca* hay. In the timeframe of 30 to 90 days of storage time, enterobacteria have its population reduced.

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