EVALUATION OF SEASON OF HARVEST AND WATER ADDITION ON FORAGE QUALITY OF WILLOW (*SALIX CAROLINIANA*) PROCESSED FOR SILAGE AT DISNEY'S ANIMAL KINGDOM

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Abstract

The objective of this study was to evaluate two seasons of harvest and water addition at the time of ensiling on silage quality of Carolina willow (Salix caroliniana). If successful, willow silage could be used as a supplement during the winter months for some browse-consuming animals at Disney's Animal Kingdom. The experiment consisted of two harvest seasons and two moisture concentrations for the willow in a two by two factorial design. Willow was harvested in April and August (beginning and middle of the Florida willow-growing season), chipped to reduce particle length, and packed into plastic-lined 208-liter drums for ensiling. At each harvest month, water was added to half of the willow just prior to packing in order to reduce dry matter content and potentially improve fermentation. After approximately 300 days of storage, samples were collected and analyzed for nutrients and fermentation characteristics. Approximately 80% of the sugar of the fresh willow (10% of dry matter (DM)) was used during fermentation. Willow harvested in the spring experienced a slightly less favorable fermentation based upon lower lactic acid concentrations (1.19 vs. 1.52% of silage DM), greater butyric acid concentrations (0.043 vs. 0.0% of silage DM), and elevated pH (4.97 vs. 4.48). This may have been due to greater concentrations of starch and lower concentrations of ash in willow harvested in the summer compared to that in the spring. Decreasing the DM concentration by 2 to 3%units by adding water at the time of ensiling resulted in lower concentrations of lactic acid (1.03 vs. 1.68% of silage DM) and greater concentrations of acetic acid (0.64 vs. 0.38% of silage DM). Adding water to willow prior to ensiling may improve silage quality when willow is harvested in the summer due to increased concentration of acid. The concentrations of lactic acid and acetic acid in willow silage were lower than that expected in corn or legume silages, although the lactic acid to acetic acid ratio fell within the range reported for corn silage. Carolina willow can be successfully ensiled. Its use as a potential dietary item during the winter months for those animals requiring browse in their diets remains to be determined.

Introduction

In recent years, an effort has been put forth to ensile browse in order to provide appropriate herbaceous material to browsing species held in zoological institutions over the winter months. Ensiled browse has been used to feed such animals as black rhinoceros at the Zurich Zoo,⁴ an assortment of ruminant browsers such as okapi, giraffe, kudu, bongo and tufted deer at the Rotterdam Zoo,⁵ and giraffe and moose at the Toronto Zoo.³ All three facilities have been able to successfully ensile willow and other woody plants to feed to the browsers during months when fresh browse is not available. Up to this point, all silage projects involving zoological

institutions have been conducted in Northern climates where fresh browse is available only in late spring, summer and early fall.

A small scale silage project was previously conducted at Disney's Animal Kingdom (DAK),⁶ the results of which will be compared with the current project. The growing season at DAK in Orlando, Florida is substantially longer than those in more northern locations. In spite of the warmer climate and longer growing season, there still exists a period of time when fresh Carolina willow (*Salix caroliniana*), the most extensively used browse species at DAK, is not available. In order to maintain a minimum level of browse in the diet of browsing species such as giraffe (*Girafa camelopardalis*), kudu (*Tragelaphus strepsiceros*), bongo (*Tragelaphus eurycerus isaaci*), nyala (*Tragelaphus angasii*) and black rhino (*Diceros bicornis minor*), willow was ensiled with the intent that it could be used as a supplement with the limited assortment of fresh browse available for purchase from December to March. It was hoped that the ensiled willow would help to lower food costs during the winter months as most browse available to the animals during this time had to be purchased at a premium price from an outside vendor.

Methods and Materials

Carolina willow was grown and harvested in 2007 from the willow browse farm at DAK. Four treatment groups were arranged in a 2 by 2 factorial design with season of harvest and water addition at the time of ensiling as the two factors. The harvest seasons were April (beginning of growing season) and August (middle of growing season). Water was mixed with the willow in order to increase the moisture content from approximately 60% for controls to 63.5%. Branches smaller than 3 cm in diameter and in full leaf were cut and processed through a wood chipper (Vermeer BC625A Brush Chipper, Pella, IA) as described by Schlegel (2005). Approximately 300 kg of willow was chipped onto a large tarp and thoroughly mixed. For the water treatment, water was sprayed on the chipped willow to increase the moisture content of the willow was determined by sampling willow harvested the week prior to the trial and dried in a freeze dryer (Labconco model 77530, Kansas City, MO).

Chipped willow was added to 208 liter plastic drums (Model T55C400UD23LL, Orlando Drum, Orlando, FL) lined with 2 Food and Drug Administration (FDA) approved 10 mil drum liners (model L375310 from linersandcovers.com) with a shovel. Willow was manually compacted (stomped) throughout the filling process to ensure adequate compaction. Each drum was fitted with an airtight lid and sealed with a metal clasp. The weight of each drum was recorded empty and full (Ohaus bench scale ES100L, Pine Brook NJ). Six drums were prepared for each of the 4 treatments.

Drums were randomly opened to assess silage quality after 300 ± 50 days of storage. Drums were completely emptied and samples collected from the top, middle and bottom levels of each barrel. Within each drum, samples were composited across sampling level prior to chemical analysis. For determination of fermentation profile, samples from each level of collection were analyzed. Silage and unensiled samples were sent to Dairy One (Ithaca, NY) for determination of the following: dry matter (DM), crude protein (CP), acid detergent fiber (ADF), neutral detergent fiber (NDF), acid detergent insoluble crude protein (ADCIP), non fiber carbohydrates

(NFC), relative feed value (RFV), rumen degraded nitrogen (RDN), net energy of lactation (Nel), net energy of maintenance (Nem), net energy of gain (Neg), metabolizable energy (ME), and minerals (calcium (Ca), phosphorus (P), magnesium (Mg), potassium (K), sodium (Na), iron (Fe), zinc (Zn), copper (Cu), manganese (Mn), molybdenum (Mo)) lignin, starch, total sugars, lactic acid, acetic acid, lactic:acetic, propionic acid, butyric acid, iso-butyric acid, volatile fatty acid (VFA) score, total acids, pH, ammonia nitrogen, and mycotoxins (aflatoxin, vomitoxin, T-2, zeaalonone, 15-Acetyl deoxynivalenol (DON), 3-acetyl DON).

Any mold found in the barrel was removed, as was 5 cm of silage below the mold. The moldy samples were sent to Dairy One to obtain a fermentation profile and mycotoxins screen. Non-moldy silage was returned to the barrel fitted with a new 10 mil liner, sealed, and stored in the Animal Nutrition Center browse cooler at 4°C until results of the mycotoxins screen were provided.

Results and Discussion

There was little variation in the nutrient content of the fresh and ensiled willow averaged across treatments with the exception of sugar which decreased from 10.0% in the fresh willow to 1.8% (DM) in the ensiled willow (Tables 1 and 4). This decrease was to be expected as sugar is used by microbes to complete the ensiling process. There were small increases in the amount of ADF, NDF and lignin and a small decrease in the average content of crude protein due to ensiling. Iron concentration of the silage (62 ppm) was much greater than that of the fresh willow (29 ppm). A similar change in iron concentration was not reported by other researchers reporting on the ensiling of other browse species.^{1,6}

There were some significant differences in the fermentation characteristics of the willow due to season and treatment (Table 2). As planned, adding water at the time of ensiling decreased the dry matter concentration of willow silage from 39.9 to 36.5% (P<0.001). Unexpectedly, adding water appeared to reduce the extent of fermentation across both harvest seasons, as evidenced by lower concentrations of lactic acid (1.03 vs. 1.68% of silage DM) and elevated pH (4.81 vs. In addition, the efficiency of fermentation was reduced by shifting from a 4.64). homofermentative toward more of a heterofermentative-type as evidenced by an increased concentration of acetic acid (0.63 vs. 0.39 % of silage DM) in willow mixed with water prior to ensiling (P<0.001). Compared to willow ensiled in the spring, willow ensiled in the summer contained less dry matter (39.4 vs. 37.0%), less CP (7.1 vs. 6.3%), and less ammonia (0.15 vs. 0.08%) (P<0.001). In addition, willow harvested in the spring experienced a slightly less favorable fermentation based upon lactic acid concentrations (1.19 vs. 1.52% of silage DM), butyric acid concentrations (0.043 vs. 0.0% of silage DM), and pH (4.97 vs. 4.48) (P<0.01). Adding water at the time of ensiling appeared to be advantageous for summer-harvested willow because the total acid content of the willow silage was returned to control concentrations whereas water addition to willow harvested in spring resulted in lower acid production (season by water addition interaction, P=0.03). Concentrations of total acid of the willow silage were lower than what is typically detected in corn or legume silages.² Expected concentrations of lactic acid are greater than 4% in corn silages and 3% in legume silages (DM). Average lactic acid content of willow silages were 1.36%. This is greater than the 1.21% reported by Schlegel¹ for willow ensiled for 84 days, but lower than the 2.32% for willow reported by Baertsche.²

Concentrations of acetic acid expected for corn and legume silages are less than 3%.² The ensiled willow in this study averaged 0.51% acetic acid (% DM) which is greater than previous values of 0.21% reported by Schlegel,¹ but less than the 1.87% reported by Baertsche.² The expected lactic acid to acetic acid ratio in corn and legume silage is between 1.5 and 4 and between 2 and 3, respectively.² Our ratio of 3.3 falls within the acceptable range of corn silage. Concentrations of propionic and butyric acids found in the willow silage were similar to those reported previously for DAK willow⁶ and below the expected values for both corn and legume silages.² The expected total acids for silage are between 5 and 10%. Total acid concentration of ensiled willow of 2.08% fell well below that, but exceeded previously reported values of 1.68%.⁶

Concentration of acetate and butyrate in the silage were not the same throughout the drums. Willow ensiled in the summer with added water had greater concentrations of acetate in the top and middle sections of the drums than silage sampled from the bottom of the drums whereas acetate concentrations did not differ across sampling levels in the other treatments (treatment by level interaction, P=0.05; Table 3). In a similar pattern, butyrate concentration was greater in the top third of the silage prepared from the spring harvest compared to that in the bottom third.

On average, willow silage was 6.8% CP, 62.9% NDF, 53.2% ADF, 19.7% lignin, 1.4% starch, 1.8% sugar, 2.1% ether extract, and 5.2% ash (Table 4). Calcium (1.3%) and K (0.75%) were found in greatest concentration of the 13 minerals measured. The digestibility of willow silage will be relatively low due to its high concentration of lignin. There was little effect of harvest season or addition of water on the chemical composition of willow silage (Table 4). Addition of water at the time of ensiling tended to decrease (P=0.06) the DM concentration from 38.3 to 36.3%, averaged across harvest seasons. Starch was greater (2.0 vs. 0.8% of DM) and ash was lower (4.9 vs. 5.4% of DM) in the willow harvested in the summer compared to the spring. The decrease in ash concentration was mainly due to lower concentrations of phosphorus, magnesium, potassium and sodium. The micro minerals of manganese (9.3 vs. 12.7 ppm) and zinc (94 vs. 103 ppm) also were lower in summer silage compared to spring silage.

No mycotoxins were detected in any of the silage samples or in any of the samples collected from mold found on the silage. Mold was present in all but 2 of the silage drums. The drums without mold were prepared on the last day of collection in August, and both were from the moisture-added treatment. Both barrels also had plastic from the drum liners covering the silage, creating a barrier between the silage and the drum lid. Most of the other barrels had the plastic of the drum liner removed to facilitate easier sealing of the drums.

Conclusions

Nutrient composition of ensiled willow was similar to that of willow prior to ensiling with the exception of the expected changes of decreased concentrations of sugars and protein and increased concentrations of ADF, NDF and lignin. Willow harvested in the spring and summer seasons were successfully ensiled using 208 liter plastic drums although fermentation was not extensive. However, willow harvested in the spring experienced a slightly less favorable fermentation based upon lower lactic acid concentrations (1.19 vs. 1.52% of silage DM), greater butyric acid concentrations (0.043 vs. 0.0% of silage DM), and elevated pH (4.97 vs. 4.48). This may have been due to greater concentrations of starch and lower concentrations of ash in willow

harvested in the summer compared to that in the spring. Decreasing the DM concentration by 2 to 3% units by adding water at the time of ensiling resulted in lower concentrations of lactic acid (1.03 vs. 1.68% of silage DM) and greater concentrations of acetic acid (0.64 vs. 0.38% of silage DM) and therefore a lower lactic acid to acetic acid ratio compared to willow ensiled without the addition of water. Adding water to willow prior to ensiling may improve silage quality when willow is harvested in the summer due to increased concentration of acid. Future studies could include the ensiling of willow with a silage enhancer to increase production of lactic and acetic acids. The palatability and intake potential of the silage with the target species of browsers must also be addressed.

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Nutrient (% DM)	Chipped Willow April	Chipped Willow August
DM %	40.99 ± 0.03	40.28 ± 0.01
CP %	7.3 ± 0.0	7.25 ± 0.49
ADF %	54.15 ± 0.65	49.15 ± 1.48
NDF %	62 ± 0.4	62.7 ± 0.14
Lignin %	15.85 ± 0.45	16.45 ± 0.64
Starch %	1.25 ± 0.15	2.6 ± 0.71
Sugar %	10.45 ± 0.55	9.6 ± 0.0
Ca %	1.30 ± 0.03	1.21 ± 0.0
P %	0.18 ± 0.01	0.17 ± 0.1
Mg %	0.13 ± 0.01	0.1 ± 0.01
К %	0.82 ± 0.01	0.69 ± 0.01
Na %	0.15 ± 0.01	0.14 ± 0.01
Fe ppm	29.0 ± 4.0	28.5 ± 2.12
Zn ppm	107.5 ± 0.5	87 ± 4.24
Cu ppm	7.0 ± 0.0	5.0 ± 0.0

Table 1. Average nutrient composition of willow chipped in April (n=2) and August (n=2) with standard deviation

Table 2. Effect of harvest season and water addition on the fermentation characteristics of willow.

	Season					Statistical contrasts		
	Spring		Sum	mer		Season	water	intrxn
Item (% DM)	No	+	No	+	SEM	P value		
	water	water	water	water				
DM, %	40.7	38.1	39.1	35.0	0.5	< 0.001	< 0.001	0.15
СР %	7.0	7.2	6.8	5.9	0.2	< 0.001	0.15	0.01
Ammonia %	0.15	0.16	0.09	0.08	0.02	< 0.001	0.97	0.42
Ammonia, % of CP	2.1	2.2	1.4	1.4	0.2	0.001	.082	0.72
pН	4.88	5.06	4.41	4.56	0.06	< 0.001	0.01	0.85
Lactate %	1.58	0.80	1.78	1.27	0.13	0.01	< 0.001	0.30
Acetate %	0.35	0.42	0.44	0.84	0.05	< 0.001	< 0.001	0.001
Lactate: acetate ratio	4.8	2.2	4.1	2.1	0.4	0.41	< 0.001	0.52
Propionate %	0.037	0.033	0.002	0.006	0.005	< 0.001	1.00	0.44
Butyrate %	0.037	0.049	0.0	0.0	0.005	< 0.001	0.21	0.21
Iso-butyrate %	0.14	0.26	0.09	0.19	0.03	0.09	0.002	0.61
Total acid %	2.14	1.56	2.32	2.30	0.12	0.001	0.02	0.03

Table 3. Effect of sampling level within the ensiling drum on concentrations of acetate and butyrate in willow ensiled during spring or summer with or without added water.

	Season					Statistical contrasts				
	Spr	ing	Summer			Season	Water	Level	Trt *	
									level	
Sampling	No	+	No	+	SEM					
level in	water	water	water	water		P value				
drum										
Acetate						< 0.001	< 0.001	0.06	0.05	
Тор	0.36 ^a	0.39 ^a	0.50^{a}	1.04 ^b	0.08					
Middle	0.36^{a}	0.39 ^a	0.43 ^a	0.91 ^b	0.08					
Bottom	0.33 ^a	0.46^{a}	0.39 ^a	0.55^{a}	0.08					
Butyrate						< 0.001	0.21	0.10	0.36	
Top	0.05^{a}	0.06^{a}	0	0	0.008					
Middle	$0.04^{a,b}$	0.04^{b}	0	0	0.008					
Bottom	0.02^{b}	0.04 ^b	0	0	0.008					

^{a, b} Means with different superscripts for each dependent variable are different, $P \le 0.05$.

	Season					Statistical contrasts		
	Sp	ring	Sur	nmer		season	water	intrxn
Measure (as %	No	+ water	No	+ water	SEM		P value	
DM)	water		water					
DM %	39.1	37.4	37.6	35.3	1.0	0.08	0.06	0.74
CP %	7.2	7.0	6.8	6.2	0.4	0.13	0.31	0.56
NDF %	62.9	63.1	60.9	64.9	1.0	0.91	0.07	0.09
ADF %	53.4	50.9	54.0	54.7	1.9	0.29	0.65	0.43
Lignin %	19.7	20.0	18.6	20.7	1.2	0.86	0.34	0.45
NFC %	24.9	24.9	27.1	24.7	0.6	0.12	0.07	0.06
Starch %	1.0	0.6	1.8	2.2	0.2	< 0.001	1.0	0.07
Sugar %	1.5	2.0	2.7	0.9	0.5	0.99	0.27	0.05
Ether extract %	2.1	2.1	2.3	1.8	0.2	0.94	0.26	0.20
Ash %	5.5	5.4	4.9	4.9	0.2	0.02	0.81	0.84
Ca %	1.33	1.26	1.28	1.38	0.05	0.48	0.80	0.10
Р%	0.19	0.17	0.16	0.16	0.01	0.001	0.29	0.18
Mg %	0.14	0.14	0.10	0.10	0.004	< 0.001	1.0	1.0
К %	0.83	0.81	0.68	0.68	0.01	< 0.001	0.60	0.45
Na %	0.16	0.17	0.14	0.13	0.01	< 0.01	0.90	0.28
S %	0.12	0.11	0.10	0.10	0.007	0.04	0.27	0.71
Fe ppm	89	47	53	58	10	0.24	0.10	0.04
Zn ppm	101	105	95	94	4	0.03	0.65	0.53
Cu ppm	7.0	6.8	6.8	6.0	0.7	0.46	0.46	0.62
Mn ppm	13.7	11.7	9.3	9.3	0.7	< 0.001	0.17	0.17
Mo ppm	0.3	0.3	0.3	0.2	0.04	0.69	0.25	0.43
Se ppm	0.025	0.005	0.008	0.018	0.005	0.74	0.32	< 0.01
Co ppm	0.07	0.03	0.05	0.03	0.01	0.58	0.09	0.36

Table 4. Effect of harvest season and water addition on the chemical composition of ensiled willow.