Chemical Composition, Antioxidant Compounds and Antioxidant Capacity of Ensiled Coffee Pulp

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ABSTRACT: Coffee pulp is produced in large quantities and its disposal can pollute the environment. However, its nutritional value and antioxidant content make it a good option for feeding animals. Therefore, the objective of this study was to determine the chemical composition, the presence of phenolic compounds and the antioxidant capacity of coffee pulp: fresh (FCP), ensiled for 140 days (ECP), and ensiled for 140 days and sun dried (EDCP). Dry matter (DM), crude protein (CP), ash, acid detergent fiber (ADF), neutral detergent fiber (NDF), lignin, phenolic compounds and the antioxidant capacity of FPC, ECP and EDCP were determined. Data were analyzed by analysis of variance, and means were compared with the Tukey test. The percentage of CP, FDN and FDA was higher ($P<0.05$) in ECP and EDCP than in FCP (CP: 10.85, 13.10, 13.24; NDF: 49.33, 50.95, 55.18; ADF: 41.90, 46.60, 52.14). There were no changes ($P>0.05$) in lignin content (FCP 30.97 %, ECP 31.25 %, EDCP 31.50 %). Ensiling and sun drying did not decrease ($P>0.05$) caffeine or tannins in coffee pulp. No differences ($P>0.05$) were found for caffeic acid (2.03, 5.10, 4.913 mg g-1 DM in FCP, ECP and EDCP, respectively). Chlorogenic acid (2.59 FPC; 5.36 ECP; 4.87 EDCP mg g-1) increased ($P<0.05$) in concentration with the ensiling process, but it was not affected by sun drying. Ethanol decreased ($P<0.05$) in ECP and EDCP, relative to FPC (FPC 15.88 %; ECP 7.04 %; EDCP 0.00 %), while antioxidant capacity was not affected ($P>0.05$) (FCP 2594.7; ECP 2486.3; EDCP 2769.9 nmol Trolox-1 mL). It is concluded that the ensiling process and sun drying did not affect the nutritional value or antioxidant capacity of coffee pulp.

Key words: Phenolic acids, Antioxidant capacity, Coffee pulp

INTRODUCTION

After harvest, coffee berries need to be processed before their use and commercialization. This can be done by two methods: dry and wet. In the industrial wet process, approximately 29% (dry weight) of the coffee berries remain as the first by-product. The berries have been used to produce bioethanol, biogas, compost, substrate for mushrooms, and animal feed. In addition, coffee pulp is an excellent source of natural antioxidants, which can prevent or decrease peroxidation of fatty acids, reduce oxidative stress in animals at critical physiological stages (Salinas *et al*., 2015) and increase shelf life of the meat of animals when it is included in their diets. The objective of this study was to assess the chemical characteristics, presence of antioxidant compounds and antioxidant capacity of coffee pulp: fresh, ensiled with 5 % molasses, and ensiled and sundried.

MATERIALS AND METHODS

Coffee pulp was obtained from a de-pulping plant that uses the wet method to remove the berry from the coffee (*Coffea Arabica*) bean. The plant is located in the municipality of Huatusco, Veracruz, Mexico. The coffee pulp was allowed to drain for 12 h to eliminate the water it absorbs during the process. The pulp was then placed in four plastic 1100 L capacity containers and 5 % molasses was added for fermentation during 140 days. When the silos were opened, samples were taken from the upper, middle and lower parts and mixed to obtain a single sample for later analysis. The silage from each container was sundried for 8 days. In this way, three treatments resulted (n=4): T1, fresh coffee pulp (FCP); T2, ensiled coffee pulp (ECP); and T3, ensiled sundried coffee pulp (EDCP).

FCP, ECP and EDCP were analyzed for dry matter (DM), crude protein (CP), ether extract (EE) and ash (AOAC, 1990), as well as neutral detergent fiber (NDF), acid detergent fiber (ADF) and lignin (Van Soest *et al.*, 1991). Ethanol content was determined following the technique of Davies and Chace (1969).

Phenolic acids and caffeine were determined by HPLC. Caffeine was quantified by isocratic analyses using HPLC. Tannins were analyzed with a spectrophotometer at an absorbance wavelength of 725 nm.

FRAP (ferric reducing antioxidant power) was determined in extracts of FCP, ECP and EDCP (Benzie and Strain, 1999).

The experimental design used was completely randomized with three treatments. The software PROC GLM (SAS, 2002) was used, and means were compared with Tukey test (P<0.05). All of the variables were subjected to a normality test with the Univariate procedure.

RESULTS AND DISCUSSION

Table 1 shows the chemical composition of the treatments. Dry matter content was affected (P<0.05) by the ensiling and drying processes. The lowest value was observed in ECP. The DM value (22.95 %) in FCP is similar to the 23.20 % reported by other authors (Elias, 1978), as was the value found in ECP (18.84 %) which is close to the 15.40 % obtained by others (Bautista *et al.*, 2005).

The concentrations of crude protein (CP) tended to increase (P<0.05) in FCP (10.85 %), ECP (13.10 %) and EDCP (13.24 %), although the last two were not different (P>0.05). Similar results have been reported, with increments in CP from FCP (9.12 %) to ECP (13.14 %) and 11.60% in FCP and 12.00% in ECP. The ensiling process increased the percentage of CP, probably due to a decrease in carbohydrates.

The ether extract of FCP was not affected by the ensiling and dehydration processes (P>0.05), FCP (1.20%), ECP (1.48%) and EDCP (1.72%). The results of ether extract in ensiled coffee pulp coincide to those found by other authors (Moreau *et al.*, 2003). In sun dried coffee pulp, there were values of 2.8 and 1.34%, which are similar to those observed in our study, while in FCP, the value was below 3.86% but above 0.48 (Noriega *et al.*, 2012).

Table 1. Chemical composition of coffee pulp

Parameters	Coffee pulp				
(%)	FCP	ECP	EDCP	SEM	P < F
Dry matter	22.95b	18.84c	91.13a	0.240	0.0001
Crude protein	10.85b	13.10a	13.24a	0.070	0.0001
Ether extract	1.20	1.48	1.72	0.357	0.6403
NDF	49.33b	50.95b	55.18a	0.864	0.0003
ADF	41.90c	46.60b	52.14a	0.667	0.0001
Lignin	31.50	30.97	31.25	0.282	0.4152
Ash	7.40c	8.76b	10.82a	0.107	0.0001
pH	4.25a	3.90b	ND	0.034	0.0001

FCP: fresh coffee pulp, ECP: coffee pulp ensiled with 5% molasses, EDCP: coffee pulp ensiled and sun dried, NDF: neutral detergent fiber, ADF: Acid detergent fiber. ND: not determined.
a,b,c Different letters in the same row indicate significant differences (P<0.05).
SEM: Standard error of the mean

Values for NDF and ADF increased (P<0.05) in EDCP, although there were no differences (P>0.05) between FCP and ECP for NDF. The increase in ADF and NDF may be related to the presence of coffee stems or leaves in the silage, increasing cell walls. Although the percentage of lignin was similar (P>0.05) in FCP (30.50%), ECP (31.97%) and EDCP (30.25%), the results were higher than those published by other authors (Oliveira *et al.*, 2007). The high content of lignin in the silage of this experiment may have been due to the presence of coffee plant residues in the silage, in addition to the age at harvest, environmental conditions and the variety of coffee used.

Ash content had significantly different (P<0.05) values in ECP (8.76%), EDCP (10.82%) and FCP (7.40%). The increase could be due to the contamination with soil where the coffee pulp was sun dried.

Table 2 shows the results for caffeine, tannins and ethanol. An increasing trend in caffeine content was observed in FCP, ECP and EDCP (18.60, 25.49 and 30.29 mg g-1 DM, respectively), although the values were not statistically different (P>0.05). The ensiling and drying processes did not modify the initial levels of caffeine. However, even though caffeine is a factor that may limit coffee pulp as animal feed, no negative effects have been observed in ruminant productive variables when fed fresh or dehydrated coffee pulp in different proportions of the diet (Salinas-Rios *et al.*, 2015; Bautista *et al.*, 2005).

Tannin content was similar (P>0.05) in the three treatments, although there was an increase from FCP to ECP and a decrease in EDCP (3.5, 4.49 and 1.18 mg g^{-1} DM, respectively). The lack of differences may have been because the fermentation process in the silo did not modify the tannins and their concentrations were maintained.

Differences (P<0.05) were observed in the quantity of ethanol in FCP and ECP (15.88 and 7.04 g 100^{-1} DM, respectively), but none was detected in EDCP. The ethanol found in our study is within the acceptable range of 10 a 30 g kg^{-1} DM for silage.

Eight phenolic acids were found (Table 2). Ferulic, caffeic and chlorogenic acids were found in greater proportion, which increased over the ensiling process, but decreased when silage was sun-dried. Differences (P<0.05) were found only for chlorogenic acid, although in ECP and EDCP it was similar (P>0.05). Chlorogenic acid was not the most abundant, as found in other studies (Murthy and Naidu, 2010) because the quantity varies with the degree of maturation, the species and other factors associated with coffee quality, such as altitude and the presence or absence of shade, as well as resistance to some diseases (Humphrey and Macrae, 1987).

Table 2. Caffeine, tannin, antioxidant compounds and antioxidant capacity (FRAP) in coffee pulp

	Coffee pulp				
	FCP	ECP	EDCP	SEM	P < F
Caffeine (mg g^{-1} DM)	18.60	25.493	30.296	2.78	0.1014
Tannins (mg g^{-1} DM)	3.50	4.490	1.189	0.945	0.1173
Ethanol (g 100 g^{-1} DM)	15.880a	7.040b	0.000c	0.307	0.0001
Antioxidants (mg g^{-1} DM)					
p-hydroxybenzoic acid	0.070a	0.018b	0.017b	0.010	0.0311
Chlorogenic acid	2.593b	5.368a	4.875a	0.34	0.0052
Ferulic acid	4.256	8.503	4.256	2.45	0.3642
Caffeic acid	2.031	5.103	4.913	0.851	0.0950
Syringic acid	0.062a	0.036b	0.0394b	0.003	0.0037
Gallic acid	0.058b	0.144a	0.00b	0.018	0.0050
Vanillic acid	0.006	0.016	0.010	0.006	0.5326
p-cumaric acido	0.002	0.002	0.0004	0.001	0.2917
FRAP (nmol Trolox-1 mL)	2769.9	2594.7	2486.3	136.47	0.4382

FCP: fresh coffee pulp, ECP: coffee pulp ensiled with 5% molasses, EDCP: coffee pulp ensiled and sun dried

a, b: Different letters in the same row indicate significant differences (P<0.05).

SEM: Standard error of the mean

Antioxidant capacity was not affected (P>0.05) by ensiling or sun drying (FCP: 2769.9; ECP: 2594.7; EDCP: 2486.3 nmol $Trolox^{-1}$ mL) (Table 2), coinciding with findings of other authors (Salinas *et al.*, 2015) in fresh and ensiled coffee pulp. Using microorganisms (*Aspergillus tamarii*) to ferment the coffee pulp, extracts from fermented coffee pulp had greater antioxidant capacity despite the lower content of total phenolic compounds and hydroxycinnamic acids, which were metabolized by this fungus. Our study found no differences in some of the phenolic acids because no microorganism was added during ensiling or drying to favor fermentation.

CONCLUSIONS

Ensiling increased the percentages of CP, ADF, NDF and ash without affecting the content of lignin and EE. The highest concentrations of CP, ADF, NDF, ash and lignin were observed in ensiled sun-dried pulp. P-hydroxybenzoic and syringic acids decreased with the ensiling process and with sun drying afterward, but chlorogenic acid increased. Gallic acid increased with fermentation, but it was imperceptible in the sun-dried pulp. Ferulic, caffeic, vanillic and p-cumaric acids were not affected. Antioxidant capacity did not vary in fresh, ensiled or dried ensiled pulp. Therefore, it is recommended the use of fresh, ensiled, and dried ensiled coffee for feeding ruminants.

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