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Nutritional profile of leaf litterfall as feed resource for grazing animals in semiarid regions

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Abstract: Leaf litterfall samples, monthly collected (2008) in two sites (Crucitas and Campus) at the Tamaulipan Thornscrub vegetation from northeastern Mexico, were subjected to chemical, *in vitro* fermentation assays. Thus, *in vitro* gas production at 24 hours, metabolizable energy and microbial protein synthesis were estimated. The presence of significant interactions for all variables (except the interaction site*month for the ash content) indicated that the studied factors are not independent among them. Considering the whole year and both sites, the neutral detergent fiber content were higher (41.6 vs 33%) and ether extract (3.7 vs 3.3%) in Campus site than in Crucitas site. Conversely, Campus site had higher ash content (13.6 vs 10.0%), crude protein (13 vs 9%), *in vitro* true organic matter digestibility (74 vs 62%), metabolizable energy (11.5 vs 9.5 MJ/kg dry matter), gas production (50 vs 65) and microbial protein (6.0 vs 5.4 µmol) than Crucitas site. In Crucitas site, the lower values for neutral detergent fiber fraction and higher values for crude protein content (28 vs 12.3%) were during the period of January to June, but in Campus site was just the opposite (42% vs 9.0%). It is concluded that the most of samples of litter fall in all months (except for crude protein in Campus site) have a good nutrient concentration and digestibility. Additionally, the estimated annual leaf litter production in Crucitas site and Campus site of 5.1 and 3.8 ton ha^{-1}, respectively is considered as enough biomass availability to meet the maintenance and weight gain requirements in adult range small ruminants.

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Keywords: chemical composition; *in vitro* gas production parameters; leaf litter; Tamaulipan Thornscrub vegetation.

1. Introduction

Litterfall and root systems produced by plants detritus during a period of time, represent a relevant portion of organic matter for the soil (Grayston and Prescott 2005; Rasse et al., 2005). Litter decomposition and the formation of humus are processes that are dependent on vegetation and the quality and quantity of its litter production (Del Valle-Arango 2003). In northeastern Mexico, the litterfall from the Tamaulipan thornscrub vegetation is composed mainly by leaves followed by twigs, reproductive structures and miscellaneous components (Gonzalez-Rodriguez et al. 2011). The annual mean of 4.5 ton ha^{-1} of leaf litter produced in this region, may serve as an alternative potential feed for range small ruminants during dry periods and scarcity of fodders (Dominguez-Gomez et al. 2011). High consumptions of fallen woody leaves by small ruminants have been indicated (Ramírez-Lozano 2012; Agetsuma et al., 2011). Despite the significance of litter fall as energy-rich plant materials to be used by faunal populations (Khanna et al., 2009), there is a paucity of scientific information regarding spatio-temporal evaluations of the nutritive value of leaf litterfall.in semiarid regions. *In vitro* assays represent a valuable tool to understand the dynamics of rumen fermentation and thus to accurately evaluate the potential feeding value of ruminant feeds (Muetzel et al., 2014). Thus, the aim of this study was to evaluate and to compare in space (sites) and time (months) the chemical composition and the *in vitro* fermentation profile of leaf litter collected in the semiarid regions of northeastern Mexico.

2. Materials y Methods

2.1. Study area

The study was conducted in two undisturbed sites. Crucitas site, was located in the ecotone of a *Quercus* sp. forest (24º46'N; 99º41'W; 550 m above sea level) and the Tamaulipan thornscrub vegetation, historical accumulated annual rainfall is of 915 mm and the mean annual temperature of 21°C. Campus site, was situated at the Experimental Research Station of the Department of Forest Sciences of the Autonomous University of Nuevo León (24º47' N; 99º32' W; 350 m above the sea level) with dominant vegetation known as Tamaulipan Thornscrub (SPP-INEGI 1986), historical total annual rainfall is of 851 mm and the mean annual temperature of 21.8°C. Both sites were located at Linares County in the state of Nuevo Leon, Mexico. The climate of the region is subtropical and semiarid with warm summer (González-Rodríguez et al. 2013).

The main species observed in Crucitas site were *Cordia boissieri, Havaria pallens, Randia ragocarpa, Sargenta greggii* and *Zantoxylum fagara* among other of less importance. In Campus site were *Acacia amentacea, Bernardia myricaefolia, Celtis pallida, Eysenhartia polystachya, Forestiera angustifolia, Havaria pallens,* and *Zanthoxylum fagara* among others of less importance (González Rodríguez et al. 2011).

2.2. Collection of samples

Litter fall samples were collected in ten litter traps (1.0 m^2), made of wooden sides fitted with a nylon net bottom (1 mm mesh size) that were randomly scattered in each collection site of about 2500 m^2. Trap contents were collected monthly from January to December 2008. Litter contents were manually separated into the following classes: leaves, twigs (<2 cm in diameter), reproductive structures (flowers, fruits and seeds) and miscellaneous residues. Due to high volume of the samples, at an initial step, the samples were dried at room temperature and later were placed in an air forced air oven until constant weight. Samples were then ground to pass a 1.0 mm mesh. Only leaf litter samples were subjected to chemical and digestion analysis because leaves were the main component and were present in all months.

2.3. Chemical and in vitro digestion analyses

Leaf litter samples, by triplicate, were analyzed to estimate crude protein (CP), ether extract (EE) and ash contents (AOAC 1997). Neutral detergent fiber (NDFom), acid detergent fiber (ADFom) and acid detergent lignin (ADLsa) were determined according to Van Soest et al. (1991). Cellulose (ADF–lignin) and hemicellulose (NDF–ADF) were obtained by difference. The *in vitro* gas production (GP24$_h$) in samples was determined using the procedures described by Getachew et al. (2005). The *in vitro* true organic matter digestibility (IVTOMD) in samples was determined in a Daisy^{II} incubator (ANKOM Technology, Macedon, New York, USA) using the technique described by Adesogan (2005). The metabolizable energy (ME) content was calculated in accordance with Menke and Steingass (1988):

$$\text{ME (MJ/kg dry matter)} = 2.20 + 0.136\ \text{GP24}_h + 0.057\ \text{CP} + 0.0029\ \text{EE}^2$$

Where: GP24$_h$ is gas production after 24 h of incubation (ml gas/200 mg dry matter); CP is the crude protein (g/kg dry matter); and EE is the ether extract (g/kg DM).

The microbial protein (MP) content in fermented samples was determined using the technique described by Makkar (2003).

2.4. Statistical analyses

The chemical composition and digestion data were analyzed using one-way analysis of variance with a bi-factorial arrangement with effects of sites (2), months of the year (12) and the double interaction (Montgomery, 2004). Statistical analyses were performed with the Statistical Package SPSS (2004) version 13, with the model:

$$Yijk = \mu + \tau i + \beta j + (\tau\beta)ij + uijk$$

Where: Yijk is the measured parameter of the ijk treatment, μ is the overall mean, τi and βj are the effects produced by the factor A (sites) and the factor B (months), respectively; (τβ)ij is the effect produced by the interaction among A×B, and uijk is the residual error term.

Regression coefficients (R) between the studied parameters were calculated .

3. Results and Discussion

3.1. Chemical composition

Ash content (except for the interaction site*month) was significantly different between sites and among months (Table 1). Samples in Crucitas site had higher ash content (13.6 vs 10%) than in Campus site. From the months of the end of summer until end of winter, ash content was higher in Crucitas site (15%), whereas in Campus site higher values were recorded at the beginning of winter, middle summer and at the end of fall (11%). These values are within the range reported by other authors who evaluated the litterfall in the Tamaulipan Thornscrub (Lopez-Hernandez et al. 2013) and in the Microphyllous Desert Scrub (González-Rodríguez et al. 2013).

Fiber fractions (NDF, ADF, ADL, cellulose and hemicellulose included) were significantly different between sites and among months and the interaction site*month was also significant (Table 1). The NDF content of litter fall was higher in the Campus site (42 vs 33%) than in Crucitas site. Related to months, from January to December, higher values were registered in Campus site except in August. The rest of the NDF components (ADF, LDA, cellulose and hemicellulose) had the same pattern. The NDF in forage could vary from 25 to 80% (Mertens 2003). In this study, the NDF varied from 25 to 46%. Ramirez et al. (2000) reported similar values in native forages collected in northeastern Mexico during the wet season. The NDF content has relevant implications in rumen functionality. Thus, the values for this variable are adequate to stimulate chewing, ruminal activities and establish an optimum pH for small ruminants (Zhao et al. 2011); resulting in a better environment for digestion of forages in the rumen.

The CP was significantly different between sites and among months and the interaction site*month was also significant (Table 2). The CP of litter fall was higher throughout the year in Crucitas site (13.4 vs. 9.2%) than in Campus site, except in January. Both sites experience the higher values of CP at the end of spring and in summer months (July to September), which are the rainy months in the studied area. Differences in CP of forage content may be due to inherent characteristics of each species related to their ability to extract and accumulate nutrients (Camacho et al., 2010; Safari et al., 2010) in sites of different nature. In general, most of samples had CP values that satisfy and, in some months, exceeded (eg. in Crucitas site) the requirements of maintenance and weight gain in adult range small ruminants (7-12%; NRC, 2007; Yang et al. 2014).

The EE content was affected significantly by sites and among months and the interaction site*month was also significant (Table 1). The EE content was higher in Campus site (3.7 vs 3.2) than in Crucitas site. It is important to notice successive monthly increases and decreases in EE content from February to august in both sites. It seems that differences in chemical composition between sites may be related to differences in the botanical composition between sites, and differences among months may be related to changes in the climatic conditions occurred in the region (Gonzalez-Rodriguez et al., 2011).

3.2. In vitro true digestibility

Content of IVTOMD (Table 2) and ME (Table 3) were affected by the studied factors as well as the double interaction (Table 3). The IVTOMD was higher in Crucitas site (74 vs. 62%) than in Campus site, except in September. In the Crucitas site, a progressive decrease throughout January to September (from 82 to 60%) of the IVTOMD was recorded; in contrast, no important variations were observed throughout the year in Campus site (range from 57 to 64%). Al-Masri (2013) who studied five native drought-tolerant range perennial shrub that grow naturally on the south-eastern semi-desert of Syria, reported similar IVTOMD ranges (48-70%). In addition, Alicata et al. (2002) found also comparable values (53-66%) for *Atriplex halimus.*

Crucitas site had higher ME (11.5 vs 9.5 Mj/kg DM) compared to Campus site. Since the IVTOMD and ME content are closely and positively related, ME content of litter fall experience the same monthly trend that the IVTOMD. Jung and Allen (1995) suggested that the quality of feeds depends primarily on digestibility and ME content; in this regard, values of IVTOMD and consequently ME, are higher (10.6 vs 8.2 MJ ME/kg DM; NRC, 2007) than those required to guarantee the good performance of range small ruminants. Al-Masri (2013) reported a similar ME value in *C. spinosa* (9.1 MJ ME/kg DM). In this study, even though the fiber fractions were not significantly correlated with the IVTOMD and ME, in general, the samples that had lower NDF, ADF and ADL, had higher IVTOMD and ME, which is in agreement with Acero et al. (2010) and Al-Masri (2011) while studying leaves of native shrubs. Thus, leaf litter samples appear to be a good source of ME to the grazing livestock in semiarid regions.

The wide variations in chemical composition throughout the year, sites and species may be explained by soil type, fertility, leaf maturity tissue and botanical composition of the sites (Madibela et al*.,* 2002). Environmental changes also play a capital roll altering the nutritional quality of plants; indeed, high temperatures and the development of water transport system (xylem) in plants increased the fiber content and diminish other valuables nutritional components of foliage (Raven et al., 2005). Additionally, in general the lower biomass production in the Tamaulipan Thornscrub occurs in late winter (60%) and the highest in the last third of the spring (90%) when temperatures are favorable. However, the phenological stages are variable and can produce alternating periods of growth, development and latency (Garcia 1997).

On the other hand, as stated by Navar et al. (2002), the Tamaulipan thornscrub is quite dense and diverse and from the total annual biomass (6.3 ton ha^{-1}) estimated by these authors, the foliage production represents 4.5 ton h^{-1} $year^{-1}$ in this area. Thus, the biomass availability in the studied sites support the assertion that this biomass production and chemical composition of studied samples throughout the year, is enough to meet the maintenance and weight gain requirements in adult range small ruminants and white-tailed deer.

3.3. Fermentation parameters

The GP_{24h} and MP values in all samples were significantly different between sites and among months, and the double interaction was significant (Table 3). Crucitas site had higher GP_{24h} (65 vs 50 ml/200 mg) than the Campus site. It seems that the GP_{24h} diminished from January to September in Crucitas site, whereas in Campus site remain relatively stable throughout the year.

The production of gas at 24 hours and the chemical composition of feeds are closely associated with metabolizable energy values measured *in vivo* (Menke and Steingass, 1988), which is in accordance to the close relationship ($r = 0.89$; data not shown) between *in vitro* gas production and ME content in litter fall samples observed herein. Thus, higher GP_{24h} might support a superior nutritive value of the litter fall samples collected from Crucitas. Values of MP were higher in Crucitas site (5.9 vs 5.4 µmol) compared to Campus site. Higher MP values were recorded in Crucitas site during winter (from January to march), at the beginning of spring (April) and in the fall season (November and December). In terms of ruminant nutrition practices, it is recommended a good conversion of feed into microbial mass in the rumen. Thus, the concept of efficiency in the synthesis of microbial protein implies an efficient use of N and C content of feeds (Van Soest, 1994). In this study, the MP values were higher than values reported by Dominguez-Gomez et al. (2011) and Guerrero-Cervantes et al. (2012). This fact may be due to the better synchronization between the fermentable soluble carbohydrates (FSC) = 100 - (NDF + CP + ash) and the CP content during the *in vitro* incubation period of samples. In Crucitas site, the relationship was 39.7% and 13.6% (ratio = 2.9), whereas in Campus site was 38.1% and 9.2% (ratio = 4.1), respectively. In this way, it is commonly accepted that the maximum fermentation is achieved when forages or diets contain 37% or more of FSC (Stokes et al., 1991; NRC, 2001). Since the greater ruminal microbial protein production promotes a greater flow of protein to the duodenum, thus, forages that promote greater ruminal microbial protein synthesis are important to support productivity in grazing ruminants (Carro, 2001). In this study none of studied parameters were significantly correlated.

4. Conclusion

Data related to important biomass production as well as crude protein, *in vitro* organic matter digestibility, gas production at $_{24h}$, microbial protein synthesis and metabolizable energy and low content of cell wall constituents; support the potential of leaf litter for range small ruminants in semiarid regions of northeastern Mexico, mainly in site 1. Knowledge on the nutritional attributes of leaf litter of the Tamaulipan Thornscrub vegetation may become a useful tool for those interested in range small ruminant nutrition practices, with economic benefits with a reduction in the cost of the rations.

Table 1. Monthly means of the chemical composition (% dry matter) of leaf litter samples

Sites		Ash	NDF	ADF	LDA	Cellulose	Hem
Crucitas	January	15.0	29.4	19.1	8.0	11.2	10.3
	February	14.7	31.7	19.2	8.3	10.9	12.5
	March	12.0	30.6	22.0	10.4	11.7	8.8
	April	13. 1	25.2	16.3	6.8	9.5	8.9
	May	12.4	25.8	17.4	7.0	10.4	8.1
	June	12.9	27.7	19.4	9.0	10.4	8.4
	July	13.0	34.9	23.9	10.0	13.9	11.0
	August	12.2	40.5	27.7	12.6	15.1	12.8
	September	14.1	40.9	29.4	14.2	15.2	11.4
	October	14.3	38.9	30.2	13.6	16.6	8.7
	November	14.1	35.2	25.7	11.1	14.5	9.5
	December	16.0	35.6	25.0	12.6	12.4	10.5
Mean site		13.7	33.0	22.9	10.3	12.7	10.1
Campus	January	10.8	41.6	29.4	14.7	14.7	12.2
	February	10.9	45.8	32.8	17.4	15.4	12.9
	March	9.5	41.9	29.9	11.5	18.5	11.7
	April	10.3	43.3	30.6	12.8	17.8	12.6
	May	8.3	43.1	30.2	14.7	15.5	12.9
	June	9.3	39.5	27.9	12.4	15.5	11.6
	July	10.1	40.7	28.8	13.3	15.6	11.9
	August	11.0	39.3	27.7	14.0	13.6	11.6
	September	9.2	43.3	30.9	15.6	15.3	12.4
	October	10.3	40.5	27.1	11.4	15.7	13.4
	November	9.4	40.7	27.0	12.0	15.0	13.7
	December	10.8	40.3	26.4	12.7	14.7	13.8
Mean site		10.0	41.7	29.1	13.5	15.6	12.6
Total mean		11.8	37.3	26.0	11.8	14.1	11.3
SEM		0.4	0.3	0.3	0.2	0.2	0.2
		Probability					
Effects	Sites (A)	***	***	***	***	***	***
	Month (B)	*	***	***	***	***	***
	A x B	NS	***	***	***	***	***

SEM = standard error of the mean; NDF = neutral detergent fiber; ADF = acid detergent fiber; ADL = acid detergent lignin; Hem = hemicellulose; *(P< 0.05); ***(P<0.001); NS = no significant

Table 2. Monthly contents of crude protein (CP, % DM), ether extract (EE, % DM) and *in vitro* organic matter digestibility (IVTOMD, % DM) of leaf litter samples

Sites	Months	CP	EE	IVTOMD
Crucitas	January	9.0	4.4	81.9
	February	10.8	2.9	77.4
	March	12.0	4.0	78.6
	April	11.0	2.5	84.3
	May	14.3	3.9	75.0
	June	16.9	2.3	79.6
	July	14.9	4.0	76.0
	August	13.7	2.3	67.4
	September	14.1	5.5	60.0
	October	14.3	2.6	65.0
	November	14.4	2.4	71.5
	December	15.1	2.6	71.4
Mean site		13.6	3.3	74.0
Campus	January	10.8	3.1	61.3
	February	8.3	3.2	57.3
	March	8.2	4.1	58.2
	April	6.9	3.2	58.5
	May	12.2	4.9	60.4
	June	7.6	3.3	64.9
	July	9.1	4.6	64.5
	August	11.9	3.3	64.3
	September	9.1	3.8	61.5
	October	8.9	3.4	62.1
	November	8.7	4.6	64.4
	December	9.2	3.8	64.6
Mean site		9.2	3.7	61.8
Total mean		11.3	3.5	67.9
SEM		0.2	0.1	0.5
		Probability		
Effects	Sites (A)	***	***	***
	Month (B)	***	***	***
	A x B	***	***	***

SEM = standard error of the mean; ***(P<0.001).

Table 3. Monthly means of the in vitro gas production (GP 24h, ml/200 mg), metabolizable energy (ME, MJ/kg) and microbial protein (MP, µmol) of leaf litter samples

Sites	Months	GP $_{24h}$	ME	MP
Crucitas	January	76	13.1	7.6
	February	70	12.4	8.1
	March	78	13.4	6.7
	April	89	13.5	9.7
	May	68	9.9	5.6
	June	70	12.2	3.7
	July	71	12.3	6.0
	August	53	12.4	6.7
	September	44	8.8	5.7
	October	52	9.8	4.0
	November	59	10.8	3.5
	December	52	9.9	4.4
Mean site		65.2	11.5	6.0
Campus	January	45	9.0	5.9
	February	44	8.7	5.3
	March	50	9.5	6.2
	April	46	8.0	6.2
	May	50	9.7	5.9
	June	59	10.8	6.6
	July	57	10.5	6.1
	August	54	10.0	5.3
	September	52	9.8	7.3
	October	53	10.0	4.3
	November	50	9.5	2.4
	December	44	8.8	4.0
Mean site		50.3	9.5	5.5
Total mean		58	10.6	5.9
SEM		0.9	0.1	0.1
		Probability		
Effects	Sites (A)	***	***	***
	Month (B)	***	***	***
	A x B	***	***	***

SEM = standard error of the mean; ***($P \leq 0.001$).

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