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Nutrient Digestibility and Metabolizable Energy Content of *Mucuna pruriens* Whole Pods Fed to Growing Pelibuey Lambs

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ABSTRACT: The nutrient digestibility, nitrogen balance and *in vivo* metabolizable energy supply of *Mucuna pruriens* whole pods fed to growing Pelibuey lambs was investigated. Eight Pelibuey sheep housed in metabolic crates were fed increasing levels of *Mucuna pruriens* pods: 0 (control), 100 (Mucuna100), 200 (Mucuna200) and 300 (Mucuna300) g/kg dry matter. A quadratic (p<0.002) effect was observed for dry matter (DM), neutral detergent fibre (aNDF), nitrogen (N) and gross energy (GE) intakes with higher intakes in the Mucuna100 and Mucuna200 treatments. Increasing *M. pruriens* in the diets had no effect (p>0.05) on DM and GE apparent digestibility (p<0.05). A linear reduction in N digestibility and N retention was observed with increasing mucuna pod level. This effect was accompanied by a quadratic effect (p<0.05) on fecal-N and N-balance which were higher in the Mucuna100 and Mucuna200 treatments. Urine-N excretion, GE retention and dietary estimated nutrient supply (metabolizable protein and metabolizable energy) were not affected (p>0.05). DM, N and GE apparent digestibility coefficient of *M. pruriens* whole pods obtained through multiple regression equations were 0.692, 0.457, 0.654 respectively. *In vivo* DE and ME content of mucuna whole pod were estimated in 11.0 and 9.7 MJ/kg DM. It was concluded that whole pods from *M. pruriens* did not affect nutrient utilization when included in an mixed diet up to 200 g/kg DM. This is the first *in vivo* estimation of mucuna whole pod ME value for ruminants. (**Key Words:** Lambs, *M. pruriens* Pods, Digestibility, Retention Coefficient)

INTRODUCTION

Currently, there is a renewed interest to evaluate feed potential of *M. pruriens* in ruminant animals (Castillo-Caamal et al., 2003; Matenga et al., 2003; Chikagwa-Malunga et al., 2009a, b, c, d; Loyra-Tzab et al., 2011, García-Galvan et al., 2012).

Mucuna whole pods or beans alone could be used as supplement to ruminants fed poor quality roughage diets in many tropical countries. This option would be particularly important during the dry season when there is a reduction in productive capacity of animals due to inadequate availability and low quality of pastures (Castillo-Caamal et al., 2003; Matenga et al., 2003).

Although anti-nutritional factors have been detected in *M. pruriens* (particularly in beans), there is no evidence of a detrimental effect of these compounds when *M. pruriens* is

fed to ruminants in large quantities (Castillo-Caamal et al., 2003; Mendoza-Castillo et al., 2003; Pérez-Hernández et al., 2003).

The M. pruriens bean contains a high amount of crude protein (CP) while its husk has a lower CP and a high content of lignin and fiber (Ayala-Burgos et al., 2003; Sandoval-Castro et al., 2003). The M. pruriens pods contain 56% grain and 44% husk in terms of dry matter (DM) (Sarmiento et al., 2006 unpublished data). Ayala-Burgos et al. (2003) reported effective DM degradability in sacco being 81% and 53% for bean and husk respectively. Higher values were obtained by Sandoval-Castro et al. (2003) by an in vitro digestibility technique (97.94 and 78.96% for bean and husk respectively). The organic matter (OM) digestibility from bean and husk were 96.02 and 78.85% respectively. Although, degradability values from the husk are inferior, they are still higher than those of common grasses used for ruminant livestock in many tropical areas of Mexico (Ayala-Burgos et al., 2003; Sandoval Castro et al., 2003).

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Studies evaluating nutrient digestibility or nitrogen retention from pods or bean alone of *M. pruriens* in ruminants are scarce (Chay-Canul et al., 2009; Chikagwa-Malunga et al., 2009d; Loyra-Tzab et al., 2011). Information available on metabolizable energy (ME) concentration from Mucuna bean and husk has been estimated from results obtained with *in vitro* digestibility techniques. Thus, for the beans Burgos et al. (2002) and Sandoval-Castro et al. (2003) reported an ME value for ruminants of 11.9 and 13.9 MJ/kg DM respectively and for husk 11.4 MJ/kg DM (Sandoval-Castro et al., 2003). These results suggest that *M. pruriens* could be employed as a source of nitrogen (N) and energy by the rumen microorganisms as there are no effects upon rumen microbial activity (Chikagwa-Malunga et al., 2009a).

It would be attractive for farmers to use the whole pod rather than only the bean, while discarding the husk, since the husk appear to have high digestibility. It is likely that including mucuna whole pod in ruminant diets will not cause major changes in their performance as compared with traditional feeds (Garc ía-Galván et al., 2012). However, in order to incorporate mucuna in commercial formulations, an estimate of energy retention and metabolizable energy values are needed. In a previous work (Loyra-Tzab et al., 2011) *in vivo* estimation of mucuna bean ME content was reported. However, a ME value for mucuna whole pod has not been estimated.

Thus, the objectives of this study were i) to determine apparent digestibility of Mucuna whole pod (DM, N, and gross energy (GE)), ii) to estimate N and GE retention coefficients when the *M. pruriens* whole pods was incorporated in a diet and iii) to estimate *in vivo* Mucuna whole pod ME value in diets for growing Pelibuey lambs.

MATERIALS AND METHODS

The experiment was conducted at the Faculty of Veterinary Medicine and Animal Science (University of

Yucatan), located in the central region of the state of Yucatan, Mexico (21° 15' N lat., 83° 32' long.). The region has an annual average rainfall of 953 mm. Annual average temperature and humidity of 26.5°C and 72% respectively.

The methodology employed was similar to that reported by Loyra-Tzab et al. (2011) where mucuna bean was evaluated. In this experiment mucuna whole pods (husk and bean) were evaluated. The methodology is described below.

Animals

Eight Pelibuey male lambs $(29.37\pm3.24 \text{ kg live weight})$ were housed in metabolic cages allowing a complete separation and recovery of faeces and urine. Lambs were treated for internal parasites by giving an oral dose of albendazole (7.5 mg/kg live weight (LW)) before the start of the trial.

Experimental diets

At maturity M. pruriens pods were harvested under tropical conditions of Yucatán, Mexico. Star grass (C. nlenfuensis) was harvested at 4 wk of re-growth. Both star grass and M. pruriens pods were sun dried and ground through a 3 mm sieve. Sorghum grain, soybean meal and CaCO₃ were obtained from a commercial company of the region. M. pruriens pod was mixed with the corresponding amounts of a control diet in a horizontal mixer. Control diet consisted of 545.4, 110.1, 312.6, 29.8 and 2.1 g/kg DM of sorghum grain, soybean meal, star grass hay, molasses sugarcane and CaCO₃ respectively. Experimental diets consisted of ground M. pruriens pods in the following proportions 0 (control diet); 100, 200 and 300 g/kg DM. Levels were selected to allow the estimation of energy value of Mucuna beans using the methodology of Schneider and Flatt (1975) and taking into account feasible feed formulation inclusion levels. Diets had similar CP and GE contents. The chemical composition of experimental diets is shown in Table 1. Lambs were fed daily ad libitum (at 09:00 h), allowing for a 20% increase over the DM

Table 1. Chemical composition of dietary ingredients and experimental diets and *M. pruriens* pods (g/kg DM)^a

Nutriment	Sorghum	Soybean meal	Star grass hay	Mucuna pruriens pods	Sugar cane Molasses	CaCO ₃	Control diet	Mucuna100	Mucuna200	Mucuna300
DM	884.6	889.6	872.7	893.4	780	900	876.5	877.4	882.9	880.4
СР	97.0	534.0	60.0	141.7	-	-	138.2	140.8	131.1	129.2
NDF	383.7	103.0	906.5	495.6	-	-	380.8	422.9	416.1	493
ADF	48.0	87.7	498.2	266.1	-	-	199.2	201.4	211.7	223.4
Lignin	2.3	0	99.4	35.6	-	-	48.8	44.7	44.8	41.7
Ash	15.9	76.3	44.4	43.9	-	100	38.5	37.6	38.7	39
GE	-	-	-	-	-	-	17,181	17,508	17,083	17,241

^a Except DM expressed in g/kg Fresh Base and GE in KJ/kg dry matter. Control diet, 545.4, 110.1, 312.6, 29.8 and 2.1 g/kg DM of sorghum grain, soybean meal, star grass hay, molasses sugarcane and CaCO₃ respectively. Experimental diets control diet: *M. pruriens* pods at: 1,000:0 (control diet); 898.9:101.1 (Mucuna100); 798.1:201.9 (Mucuna200); and 697.5:302.5 (Mucuna300) g/kg DM.

consumed the previous day. Water was freely available.

Feed intake and apparent digestibility

The amounts of both consumed and refused feed were recorded daily and samples were taken for chemical analyses. DM, N and GE intakes were measured as the difference between the amounts offered and those refused. Faeces were collected daily, weighed and samples were kept. All samples of feeds and faeces were stored at -20°C until analysis.

N and GE retention and N balance

Daily urinary excretion was weighed and a proportional sample was taken from each animal. The urine samples were kept at a pH below 3.0, by adding 80 ml of a diluted solution of Sulphuric acid (4 M) per liter. N content of feed, feces and urine were used to estimate N balance and retention. In addition, energy excreted through urine was estimated according to McDonald et al. (2002) from urine-N content as 23 kJ/g N excreted. A constant level of 0.08 of GE was considered to be lost as methane.

Chemical analyses

Daily feed samples and orts were proportionately pooled to obtain a single sample for each period. Samples were analyzed for DM (#7.007), CP (N*6.25) (#2.057) and ash (#7.009) (AOAC, 1980). GE was determined in an adiabatic calorimetric chamber (Parr, 1988). Neutral detergent fiber (aNDF) (Mertens, 2002), acid detergent fiber (ADF) (VanSoest et al., 1991) and lignin (sa) (Robertson and Van Soest, 1981) were also determined. aNDF was analyzed without sulphite, with amylase and ash inclusive. Faecal samples were analyzed for DM, GE and N. In addition, urine-N was determined. The methods utilized were the same as above.

Statistical analysis

A balanced block design (Mead, 1988) with two periods of fifteen days each, ten days for adaptation and five days for measurements were used. On each period, two animals were randomly allocated to each of the four treatments. A total of four animals (replicates) per treatment was obtained after the two experimental periods. One week interval was allowed between both periods where animals were fed the control diets.

Data were analyzed using a generalized linear model (Minitab, 2007). The model considered the effects of treatments (4 Mucuna inclusion levels) and experimental periods were considered as blocking factors (2 periods). Total number of replicates per treatment was 4. Degrees of freedom for experimental error = 11. Linear, quadratic and cubic effects were determined. Significant effects were

accepted if $p \le 0.05$. In addition, DM, CP and GE apparent digestibility coefficients, as well as digestible protein (DP), digestible energy (DE) and metabolizable energy (ME) content values from *M. pruriens* beans were calculated using multiple regression equations (Schneider and Flatt, 1975).

 $y = a + bX_1 + cX_2$

where y = DM, CP or GE in the diet (g or kJ/kg DM), a = intercept, b = digestibility coefficient or nutrient content of *M. pruriens* (X₁) (DM, CP or GE), c = digestibility coefficient or nutrient content of control diet (X₂) (DM, CP or GE), X₁ = amount of nutrients (DM, CP or GE) provided by *M. pruriens* in the ration (g or kJ/kg DM), or inclusion level (the later for nutrient content calculation), and X₂ = amount of nutrients provided by control diet (g or kJ/kg DM) or inclusion level (the later for nutrient content calculation).

RESULTS

Nutrient intake and digestibility of experimental diets

A quadratic (p<0.002) effect was observed for dry matter (DM), neutral detergent fibre (aNDF), nitrogen (N) and gross energy (GE) intakes with higher intakes in the Mucuna100 and Mucuna200 treatments. Increasing *M. pruriens* in the diets had no effect (p>0.05) on DM and GE apparent digestibility (p<0.05). A linear reduction (p<0.04) in N digestibility was observed as mucuna whole pods increased in the diet. However, no effects were found on estimated nutrient supply (DP, DE and ME) (Table 2).

Nutrient retention and nitrogen balance

Although N balance was positive in all treatments, a linear reduction in N digestibility and N retention was observed with increasing mucuna whole pod level. However, urine-N excretion was not affected (p<0.5). These resulted in a quadratic effect (p<0.05) on fecal-N and N-balance which were higher in the Mucuna100 and Mucuna200 treatments,

Nutrient digestibility and energy content of raw *M*. *pruriens* pods

Average DM and GE digestibility coefficients of *M. pruriens* pods were higher than that of control diet being 0.69 ± 0.060 and 0.65 ± 0.058 vs 0.51 ± 0.042 and 0.51 ± 0.040 respectively. However CP digestibility coefficient of *M. pruriens* pods was lower than that from the control diet (0.46 ± 0.070 vs 0.54 ± 0.046). The estimated DE and ME values for mucuna pods were 11.0 ± 1.08 and 9.7 ± 1.08 MJ/kg DM respectively.

		Mucuna pods inclu	usion level (g/kg D	M)	р	р
	0	100	200	300	Linear	Quadratic
Intake						
DM^{a}	79.83	135.33	112.64	84.68	0.8553	0.0013
NDF ^a	31.60	53.20	51.79	40.27	0.1774	0.0012
GE^b	1.37	2.33	1.96	1.48	0.9336	0.0015
Nitrogen balance						
N intake ^a	1.71	2.89	2.44	1.81	0.8666	0.0015
Fecal N ^a	0.54	1.16	0.88	0.70	0.6145	0.0006
Urine N ^a	0.27	0.46	0.40	0.38	0.3804	0.1675
N balance ^a	0.90	1.28	1.16	0.72	0.2978	0.0102
Digestibility						
DM	0.69	0.62	0.68	0.65	0.1738	0.0797
Nitrogen	0.67	0.60	0.63	0.62	0.0433	0.0356
GE	0.67	0.60	0.66	0.64	0.3830	0.0689
Retention						
Nitrogen	0.50	0.43	0.47	0.39	0.0395	0.8618
GE	0.59	0.52	0.57	0.55	0.3400	0.0776
Nutrient supply						
DP ^c	90.53	80.47	85.81	84.19	0.1912	0.0813
DE^d	11.58	10.42	11.45	11.17	0.8384	0.0846
ME^d	10.12	8.96	9.98	9.65	0.7165	0.0923

Table 2. Nutrient intake, retention and digestibility in diets containing M. pruriens pods

Control diet, 545.4, 110.1, 312.6, 29.8 and 2.1 g/kg DM of sorghum grain, soybean meal, star grass hay, molasses sugarcane and CaCO₃ respectively. Experimental diets *M. pruriens* pods at: 1,000:0 (control diet); 898.9:101.1 (Mucuna100); 798.1:201.9 (Mucuna200); and 697.5:302.5 (Mucuna300) g/kg DM.

^a g/kg LW^{0.75}. ^b Mj/kg LW^{0.75}. ^c g/kg DM. ^d MJ/kg DM.

DISCUSSION

Nutrients intake

Inclusion of *M. pruriens* pods in diets did not have a negative effect in total DM intake possibly due to the similar chemical composition and nutrient digestibility among M. pruriens pods and control diets. Castillo-Caamal et al. (2003) supplemented a basal diet of Pennisetum purpureum using three levels (165, 331 and 497 g/kg LW) of *M. pruriens* pods and reported a linear increase in total DM intake, without an effect on forage DM intake. On the other hand, Perez-Hernandez et al. (2003) reported a tendency to decrease the digestibility in P. purpureum basal diet when a concentrate was substituted by M. pruriens pods on 50% and 100% level, which was explained to be due to the increase in fiber (NDF) from the pod husk. However, although the diets were formulated to resemble a commercial lamb fattening concentrate diet, the resulting higher NDF concentration (particularly in Mucuna300 diet) appeared to be a limiting factor for DM intake. The reduction in DM intake was not attributed to an antinutritional factor effect as the intake level was similar to that achieve with the control diet and no symptoms of toxicity were observed.

Nutrient digestibility

A similar DM and GE digestibility between all

experimental diets was expected due to their similar nutrient composition (Table 2). Although Ayala-Burgos et al. (2003) indicated that according to its chemical composition *M. pruriens* husk was classified as a roughage. The *in vitro* and *in sacco* DM degradability of Mucuna husk (0.79 and 0.53 respectively; Ayala-Burgos et al., 2003; Sandoval-Castro et al., 2003) is higher than those from tropical grasses (Mero and Udén, 1997; Mero and Udén, 1998a). This was in agreement with previous findings on legume pod digestibility (Pieltain et al., 1996; Bruno-Soares and Abreu, 2003). Hence, DM digestibility seems not to be affected by including the husk fraction of legume pods making feasible its routine use for feeding ruminants.

Digestibility, retention and supply of nutrient from diets containing *M. pruriens* pods

DM and GE digestibility of diets formulated with *M. pruriens* pods were higher than those from most diets based on tropical grasses. Although its N digestibility was lower than those from diets based on young (re-growth) grass, it was similar or higher to values obtained with mature tropical grasses (Mero and Udén, 1998a; Archimède et al., 2000; Mendoza and Sandoval, 2003).

Digestibility and N retention of Mucuna bean diets has been previously compared with diet containing soybean meal with satisfactory results (Chikagwa-Malunga et al., 2009d; Loyra-Tzab et al., 2011; García-Galvan et al., 2012). Although a positive N balance was observed among all treatment, similar to previous results (Chikagwa-Malunga et al., 2009d; Loyra-Tzab et al., 2011), N balance was lower at higher inclusion levels. It is possible that soybean meal provides a better amino acid balance as suggested by Chikagwa-Malunga et al. (2009d) but it is also possible the result of accounting L-dopa as a protein-N source. As L-dopa would not accumulate in blood or tissue (Chikagwa-Malunga et al., 2009c), nor utilized for protein accretion, it would be excreted and result in lower N digestibility, MP supply and retention (Table 2). Although as with any other protein rich feed, efficiency of N utilization might also be influenced by diet presentation (*e.g.* particle size) (Chay-Canul et al., 2009), and grinding whole pods is likely to result in a larger particle size than grinding beans alone.

In vivo metabolizable energy value

To our knowledge, this is the first report of an in vivo ME value for Mucuna whole pod. Its 9.7 MJ/kg DM value compares favorably with many green crops such as rape, maize, sugarcane and lucerne, as well as with other feed such as clover hay (McDonald et al., 2002). It is lower than an estimation based on ME values for bean and husk when these fraction where evaluated as separated fractions (Sandoval-Castro et al., 2003), possibly due to a lower digestibility when incorporated in whole diets. Additionally, in vitro and in situ digestibility estimates represent the potential a feed can achieve which usually are higher than in vivo measurements as samples cannot escape rumen digestion. Nevertheless current estimate provides a value which can be readily incorporated into commercial formulation as has been validated by García-Galván et al. (2012), who found this ME value reliable for feed formulation in growing lambs.

CONCLUSION

Nutrient apparent digestibility and retention in diets incorporating *M. pruriens* whole pods are similar to those from diets made from conventional feedstuff when included in diets for lambs up to 200 g/kg DM without detrimental effects on nutrient digestibility and retention by growing lambs. The ME value of *M. pruriens* was estimated to be 9.7 MJ/kg DM.

REFERENCES

- AOAC, 1980. Official methods of analysis, 13th ed. Association of Official analytical Chemists, Washington, DC, USA.
- Archimède, H., M. Boval, G Alexandre, A. Xandé, G Aumont, and C. Poncet. 2000. Effect of regrowth age on intake and digestion of *Digitaria decumbens* consumed by Black-belly sheep. Anim. Feed Sci. Technol. 87:153-162.

- Ayala-Burgos, A. J., P. E. Herrera-Díaz, J. B. Castillo-Caamal, C. M. Rosado-Rivas, L. Osornio-Muñoz, and A. M. Castillo-Caamal. 2003. Rumen degradability and chemical composition of velvet bean (*Mucuna* spp.) grain and husk. Trop. Subtrop. Agroecosyst. 1:71-74.
- Bruno-Soares, A. M., and J. M. F. Abreu. 2003. Merit of *Gleditsia* triacanthos pods in animal feeding: Chemical composition and nutritional evaluation. Anim. Feed Sci. Technol. 107:151-160.
- Burgos, A., I. Matamoros, and E. Toro. 2002. Evaluation of Velvet bean (*Mucuna pruriens*) Meal and Enterolobium ciclocarpum Fruit Meal as Replacement for Soybean Meal in Diets for Dual Purpose Cows. In: Food and Feed from Mucuna: Current Uses and the Way Forward (Ed. B. M. Flores, M. Eilitta, R. Myhrman, L. B. Carew, and R. J. Carsky). Proceedings of the Centro Internacional de Información sobre Cultivos de Cobertura (CIDICCO). April 26-29, 2000. Tegucigalpa, Honduras. pp. 228-236.
- Castillo-Caamal, A. M., J. B. Castillo-Caamal, and A. J. Ayala-Burgos. 2003. Mucuna bean (*Mucuna* spp.) supplementation of growing sheep fed with a basal diet of Napier grass (*Pennisetum purpureum*). Trop. Subtrop. Agroecosyst. 1:107-110.
- Chay-Canul, A., A. J. Ayala-Burgos, J. C. Kú-Vera, and J. G. Magaña-Monforte. 2009. Efecto del tamaño de partícula sobre, consumo, digestibilidad y balance del nitrógeno en ovinos pelibuey alimentados con dietas basadas en fríjol terciopelo (*Mucuna pruriens*) y grano de maíz. Trop. Subtrop. Agroecosyst. 10:383-392.
- Chikagwa-Malunga, S. K., A. T. Adesogan, M. B. Salawu, N. J. Szabo, R. C. Littell, S. C. Kim, and S. C. Phatak. 2009a. Nutritional characterization of *Mucuna pruriens*. 2. *In vitro* ruminal fluid fermentability of mucuna pruriens, mucuna Ldopa and soybean meal incubated with or without L-dopa. Anim. Feed Sci. Technol. 148:51-67.
- Chikagwa-Malunga, S. K., A. T. Adesogan, L. E. Sollenberger, L. K. Badinga, N. J. Szabo, and R. C. Littell. 2009b. Nutritional characterization of *Mucuna pruriens*. 1. Effect of maturity in the nutritional quality of botanical fractions and the whole plant. Anim. Feed Sci. Technol. 148:34-50.
- Chikagwa-Malunga, S. K., A. T. Adesogan, L. E. Sollenberger, S. C. Phatak, N. J. Szabo, S. C. Kima, C. M. Huisden, and R. C. Littell. 2009c. Nutritional characterization of *Mucuna pruriens*.
 4. Does replacing soybean meal with *Mucuna pruriens* in lamb diets affect ruminal, blood and tissue L-Dopa concentrations. Anim. Feed. Sci. Technol. 148:124-137.
- Chikagwa, M. S. K., A. T. Adesogan, N. J. Szabo, R. C. Littell, S. C. Phatak, S. C. Kim, K. G Arriola, C. M. Huisden, D. B. Dean, and N. A. Krueger. 2009d. Nutritional characterization of *Mucuna pruriens*. 3. Effect of replacing soybean meal with *Mucuna* on intake, digestibility, N balance and microbial protein synthesis in sheep. Anim. Feed. Sci. Technol. 148: 107-123.
- García-Galván, A., R. Belmar-Casso, L. Sarmiento-Franco, and C. A. Sandoval-Castro. 2012. Evaluation of the metabolizable energy value for growing lambs of the *Mucuna pruriens* seed and whole pod. Trop. Anim. Health Prod. 44:843-847.
- Loyra-Tzab, E., L. A. Sarmiento-Franco, C. A. Sandoval-Castro, and R. H. Santos-Ricalde. 2011. Nutrient digestibility and

metabolizable energy content of *Mucuna pruriens* beans fed to growing Pelibuey lambs. Anim. Feed Sci. Technol. 169:140-145.

- Matenga, V. R., N. T. Ngongoni, M. Titterton, and B. V. Maasdorp. 2003. Mucuna seed as a feed ingredient for small ruminants and effect of ensiling on its nutritive value. Trop. Subtrop. Agroecosyst. 1:97-103.
- McDonald, P., R. A. Edwards, J. F. D. Greenhalgh, and C. A. Morgan. 2002. Animal nutrition. 6th ed. Prentice Hall. Edimburgh, UK.
- Mead, R. 1988. The design of experiments. Cambridge University Press, Cambridge, UK.
- Mendoza-Castillo, H., J. B. Castillo-Caamal, and A. Ayala-Burgos. 2003. Impact of Mucuna bean (*Mucuna* spp.) supplementation on milk production of goats. Trop. Subtrop. Agroecosyst. 1: 93-95.
- Mendoza-Nazar, P., and C. Sandoval-Castro. 2003. In vitro gas production and nylon bag rumen degradation as predictors of the *in vivo* apparent digestibility and voluntary intake of tropical hays fed to sheep. J. Appl. Anim. Res. 23:103-116.
- Mero, R. N., and P. Udén. 1997. Promising tropical grasses and legumes as feed resources in Central Tanzania II. *In sacco* rumen degradation characteristics of four grasses and legumes. Anim. Feed Sci. Technol. 69:341-352.
- Mero, R. N., and P. Udén. 1998a. Promising tropical grasses and legumes as feed resources in Central Tanzania III: Effect of feeding level on digestibility and voluntary intake of four grasses by sheep. Anim. Feed Sci. Technol. 70:79-95.

- Mertens, D. R. 2002. Gravimetric determination of amylasetreated neutral detergent fibre in feeds with refluxing beakers or crucibles: collaborative study. J. Assoc. Off. Assoc. Chem. Int. 85:1217-1240.
- Minitab. 2007. Minitab release 15. Minitab Reference Manual. Philadelphia, Minitab, State college. USA.
- Parr Instruments Co. 1988. Instructions for the 1241 adiabatic calorimeter. Moline, Il. USA.
- Pérez-Hernández, F., A. J. Ayala-Burgos, and R. Belmar-Casso. 2003. Performance of growing lambs supplemented with *Mucuna pruriens*. Trop. Subtrop. Agroecosyst. 1:119-122.
- Pieltain, M. C., A. A. R. Castañón, M. P. Flores, and J. I. R. Castañón. 1996. Nutritive value of postharvest bean byproducts (*Phaseolus vulgaris* L.) for ruminants. Anim. Feed Sci. Technol. 62:271-275.
- Robertson, J. B., and P. J. Van Soest. 1981. The detergent system of analysis. In: The analysis of dietary fibre in food (Ed. W. P. T. James, and O. Theander). Marcel Dekker. New York, USA. pp. 123-158.
- Sandoval-Castro, C. A., P. Herrera, C. M. Capetillo-Leal and A. J. Ayala Burgos. 2003. *In vitro* gas production and digestibility of Mucuna bean. Trop. Subtrop. Agroecosyst. 1: 77-79.
- Schneider, B., and W. Flatt. 1975. The evaluation of feeds through digestibility experiments. The University of Georgia Press, Athens. USA. pp. 161-167.
- Van Soest, P. J., J. B. Robertson, and B. A. Lewis. 1991. Methods for dietary neutral detergent fiber and nonstarch polysacacharides in relation to animal nutrition. J. Dairy Sci. 74:3583-3597.