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Invited Review

Leucaena leucocephala IN RUMINANT NUTRITION

[*Leucaena leucocephala* EN NUTRICIÓN DE RUMIANTES]

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SUMMARY

It is a common situation in extensive ruminant production systems in tropical countries to have low production indicators due to nutrient deficiencies in the diet. An economic alternative to increase animal production is the incorporation of legumes (fodder and fruits) in the diet. This review, presents an analysis of the positive and negative effects of *Leucaena leucocephala* consumption by ruminants, with particular emphasis on the secondary compound mimosine. Leucaena due to its high nutrient content, rumen by-pass protein supply and its possible effect on the reduction of greenhouse gas (attributed to tannins) has become one of the legumes most commonly used in ruminant feeding practices. However, in countries where leucaena has been introduced, its use is still limited to levels below 30% inclusion in the diet, due to the secondary compound mimosine and its isomers (3,4 and 2,3 DHP), which can induce toxicity, even when animals are inoculated with rumen fluid containing the bacteria *Synergistes jonesii* reported as responsible for degrading these compounds in the rumen. In the Yucatan Peninsula, Mexico, ruminants consuming leucaena can tolerate more than 50% inclusion in the diet, without having a negative impact on production, attributed intake to mimosine and its isomers. We conclude that in animals not adapted, the intake would be limited to low inclusion levels (less than 30% inclusion in the diet), mainly because of mimosine and its derivatives. The decrease in intake or diet digestibility seem to better explain the reduction in methane production, however, *in vivo* studies are required to clearly establish the mechanism of action. It has been reported the presence of different bacteria to *S. jonessi* that would have the ability to degrade mimosine and its derivatives, however, the activity of these bacteria and its effectiveness must be confirmed *in vivo*.

Key words: sheep, leucaena, tannins, mimosine, greenhouse gases.

RESUMEN

La producción de rumiantes en sistemas extensivos en países tropicales tiene indicadores productivos bajos debido al déficit de nutrientes en la dieta de los animales. Una de las alternativas económicas para obtener mayor producción es la incorporación de forrajes y frutos de leguminosas en la dieta. En esta revisión se presenta un análisis de los efectos positivos y negativos del consumo de leucaena por los rumiantes, con particular énfasis en el compuesto secundario mimosina. La leucaena debido a su alto aporte de nutrientes, paso de proteína no degradada al abomaso y su posible efecto en la disminución de los gases de efecto invernadero (atribuido a los taninos) se ha convertido en una de la leguminosas más usadas en la alimentación de rumiantes. Sin embargo, en países donde la leucaena fue introducida su consumo se ve limitado aún con niveles menores al 30% de inclusión en la dieta, debido principalmente al compuesto secundario mimosina y sus isómeros (3,4 y 2,3 DHP) los cuales inducen toxicidad, aún cuando los animales estén inoculados con liquido ruminal que contenga la bacteria *Synergistes jonesii* reportada como la responsable de degradar estos compuestos en rumen. En la península de Yucatán, los rumiantes que consumen leucaena pueden tolerar más del 50% de inclusión en la dieta, sin tener repercusiones negativas en la producción atribuida al consumo de mimosina y sus isómeros. Se concluye que en animales no adaptados a la ingesta de esta planta, el consumo puede ser limitado con niveles bajos de inclusión (menor a 30% de inclusión en la dieta), principalmente por causa de la mimosina y sus derivados. La disminución en el consumo o digestibilidad de la dieta parecen explicar mejor la reducción de la producción de metano, sin embargo se requiere estudios *in vivo* para establecer de manera clara los mecanismos de acción. Se ha reportado la presencia de bacterias diferentes a *S. jonessi* que tendrían la capacidad de degradar mimosina y sus derivados, sin embargo, la efectividad de estas debe ser corroborada *in vivo*.

Palabras claves: ovinos, leucaena, taninos, mimosina, gases de efecto invernadero.

INTRODUCTION

The leucaena is considered as the most widely consumed legume due to its characteristics such as high supply protein, energy, calcium and sulfur, the latter with a possible potentiating effect on rumen microbial populations (Aregheore, 1999). Additionally, in recent years some legumes such as leucaena, when used as feed for ruminants, has been attributed the effect of reducing emissions of greenhouse gases, an effect attributed to their content of secondary compounds such as tannins (Jayanegara et al., 2011).

On the other hand, the intake of high amounts of leucaena may have negative impact on the productive indicators of animals mainly due:

1) Excess of nitrogen in the diet, which causes an imbalance in protein-energy ratio resulting in an inefficient microbial protein synthesis, and also high levels of ammonia in blood which can affect voluntary intake (Calsamiglia et al., 2010).

2) Presence of secondary compounds, such as mimosine, which can induce toxicity or death in ruminants (Adejumo y Ademosun, 1991; Ghosh et al., 2007a; Dalzell et al., 2012), and condensed tannins that form protein-tannin complex, inhibit the activity of rumen microorganisms and results in changes in the ecology of the rumen. These effects limit the degradation of nutrients and can cause a reduction in the production of volatile fatty acids (VFA) (Ramana et al., 2000; Salem et al., 2006; Galindo et al., 2009).

However, it has been shown that ruminants can tolerate up to 30% of leucaena in the diet without having negative impact on production (Yami et al., 2000; Ghosh et al., 2007b). This has been attributed to: i) the benefit obtained by forming tannin-protein complexes in the rumen which provides rumen by-pass protein leading to better utilization of protein for animal metabolic processes (Valdivia, 2006) and, ii) the ability of certain rumen bacteria able to degrade mimosine and its metabolites (Klieve et al., 2002). Thus Rincon et al. (2003) mention that *Synergistes jonesii* a bacteria isolated from the rumen of animals in Hawaii causes an isomerization reaction of 3-4 DHP that leads the 2-3 DHP (3-4 and 2-3 dihydroxypyridine; DHP). However, it has not yet confirmed its presence or absence in animals fed leucaena in other places where this plant is native. In the Yucatan Peninsula, Mexico, it has been shown that animals can tolerate high levels of leucaena in the diet (over 50%) without toxic effects attributable to mimosine and its derivatives (Ruiz-González et al., 2011; Arjona-Alcocer et al., 2012; Peniche-González et al., 2014). The aim of this review is to present an analysis of the positive and negative effects of intake of leucaena by ruminants, with particular emphasis on the secondary compound mimosine.

ORIGIN OF *Leucaena leucocephala* (LAM.) DE WIT.

Leucaena originated in the Yucatan Peninsula, Mexico, where their forage value was recognized over 400 years ago by Spanish conquistadores who carried leucaena forage and seed in their galleons to the Philippines to feed their cattle. From there, it has spread to most tropical and subtropical countries of the world where it is used in animal feed and human food, due to its success as forage of high nutritional value (Brewbaker et al., 1985)

EFFECTS OF INTAKE OF LEUCAENA IN RUMIANTES

The leucaena forage in ruminant nutrition is widely used due to its qualities such as a high content of crude protein, which varies between 24 and 30%, depending on the variety and time of year (García et al., 2008). The digestibility of the protein reaches 63% and digestibility of dry matter between 60 and

70% measured *in vivo* (Barros-Rodríguez et al., 2012). In this sense, the use of leucanea as a protein supplement in livestock farming systems in tropical countries is widely accepted (Galindo et al., 2009). In addition, it is a source of minerals such as sulfur, which is can act as enhancer of rumen microbial populations (mainly cellulolytic fungi and bacteria) (Aregheore, 1999).

An additional possible benefit from utilizing leucaena in ruminant feeding are the effects arising from the secondary compounds it contains (for example, tannins). This compounds when consumed in moderate amounts generally have positive effects and do not reduce voluntary intake. The phenolic hydroxyl groups of tannins bind to the dietary protein in aqueous solution, leading to the formation of a complex with proteins, mainly, and to a lesser extent with metal ions, amino acids and polysaccharides, avoiding their degradation in the rumen, increasing the amount of bypass protein to the lower parts of the gastrointestinal tract (abomasum) and the amount of essential amino acids supply, resulting in higher animal production (Waghorn et al., 1987). They can also be used as a natural anthelmintic agent for gastrointestinal nematodes (Torres-Acosta et al., 2008; Alonso et al., 2010). Additional, research with forage legumes, have suggested that condensed tannins may help to reduce rumen gas production (Monforte-Briceño et al., 2005).

Leucaena and greenhouse gases

Enteric methane production by ruminants is a byproduct of microbial fermentation process and represents a loss of 2-12% of the feeding energy. Emissions of carbon dioxide and methane (CO_2 and CH_4) are considered as one of the most important global environmental issues (IPCC 2001). Kurihara et al. (1999) indicate that the energy loss of methane in cattle fed tropical forage diets was higher than in those animals fed with temperate forages diets, due to the relatively high levels of fiber and lignin and a low level of non-structural carbohydrates in tropical forages. In addition, livestock in developing countries is predominantly fed on a high fiber diet with little or no grain-based concentrate supplementation resulting in increased ruminal methanogenesis.

Therefore, the use of species that contain secondary compounds [e.g. (leucaena: tannin/saponin and *Enterolobium cyclocarpum*: saponins)] as supplements in animal feeding, is a strategy that is being studied and implemented in the tropics in order to improve animal performance and reduce greenhouse gases emissions (Abdulrazak et al., 2000). The antimicrobial action and the adverse and beneficial effects on rumen fermentation of these compounds depend on its nature, biological activity and concentration in the feed. In addition, other factors such as the species, physiological state (animal and plant) and diet composition (Makkar, 2003) might modify these effects.

Two modes of action for the anti-methanogenic activity of the tannins have been proposed: i) direct effect on the activity or population of methanogens (direct reduction of methane emissions), and ii) indirect effect by the reduction in rumen hydrogen due to reducing feed degradation (Tavendale et al., 2005). Other studies report that tannins can decrease the number of protozoa in the rumen resulting in lower methane production (Makkar et al., 1995). However, Tiemann et al. (2008) did not found changes in the fermentation pattern or number of protozoa when methane was reduced in animals fed legumes rich in condensed tannins. Similarly, when 0.2% and 1.8% of condensed tannins was incorporated in the diet the methane production did not decreased (Sliwinski et al., 2002; Beauchemin et al., 2007). However by increasing the dose to 2.5% dietary tannin the methane production in cattle and sheep was decreased by 15%, but this effect could be partly attributed to a 5% reduction of fiber digestibility (Carulla et al., 2005; Waghorn and Woodward, 2006). Generally, *in vitro* studies using extracts of plants or quebracho tannins were a decreased population of microbes methanogens (Denman and McSweeney, 2006) and the production of greenhouse gases has been reported also a decreased digestion of organic matter was found (Jayanegara et al., 2008; Jayanegara, 2009).

Martinez et al. (2003) reported that the intake of condensed tannins can affect microbial fermentation as evidenced by a decreased production of gas and NH_3 and by lower dry matter digestion. However, the microbial biomass during *in vitro* incubation was not affected by a treatment with tannin. In addition to this, Hess et al. (2003) reported that the inclusion of tree legumes with high levels of tannins as a supplement of grasses with low protein levels did not contribute with degradable protein but decreased dry matter digestibility. Similarly, Galindo et al. (2009) found a higher number of total viable and proteolytic bacteria, as well as a higher concentration of VFAs in animals fed mixtures of herbaceous legumes without tannins compared with those fed leucaena. These authors mention that leucaena protein is relatively less degraded in the rumen due to the presence of condensed tannins, which were present in the diet at 1.80-5% DM.

Summarizing the reports, the effect of tannins on greenhouse gases production might attributed mainly to a reduction in the digestion of food (indirect effect). It is noteworthy that most of the reports were the effect of tannins both gas production and feed digestion is reduced were performed *in vitro*. On *In vivo* conditions where tannins are naturally contained in the feed these effects may be different.

LIMITATIONS OF *Leucaena leucocephala* IN RUMINANT FEEDING

Nitrogen supply

The intake of this legume in high quantities may be limited due to an excess of nitrogen in the diet causing a nutritional imbalance (protein-energy ratio), affecting microbial protein synthesis, and as consequence high levels of ammonia in blood which can affect voluntary intake (Calsamiglia et al., 2010).

Excess ammonia in the rumen indicates that their intake is greater than the capacity of rumen microorganisms to transform it into microbial protein or that existing microflora is unable to use resulting ammonia at its production rate. Ammonia levels in the blood are kept low due the liver rapidly converts ammonia in urea (ornithine cycle). If ammonia production exceeds the liver ability to transform it into urea, ammonia levels in the blood can become toxic. In addition, high blood ammonia levels influence on the appetite, so that limit feed intake, and if its presence is constantly high can cause permanent suboptimal production situations (Poppi and McLennan, 1995).

Detoxification of ammonia imposes an important enzymatic activity in the liver, involving the modification of plasma levels of the ornithine carbamyl transferase. This detoxification, in turn, requires energy consumption which is important at all times, but more critical in the case of high-producing dairy cows at the beginning of lactation. Ammonia detoxification has a cost for the animal of about of 12 kcal/g of nitrogen (Arias and Nesti de Alonso, 1999).

Mimosine

Mimosine is a secondary compound that affects ruminants, can induce toxicity and cause death if animals not adapted to leucaena are feed above 30% dietary DM. Depending of the variety, the foliage of leucaena contains between 2.3 to 12% mimosine (Jones, 1994; Fortes et al., 2003; García et al., 2008). Mimosine is a free non-protein amino acid, an alanine β-substituted with an aromatic ring of 3-hydroxy-4(1H)-pyridone (3,4-DHP), structurally analogous to tyrosine. Due its similarity to tyrosine, presumably the mimosine functions as an inhibitor or antagonist in many processes involving tyrosine as an intermediary (Hammond, 1995). The aromatic ring of 3,4 DHP is in free state in the rumen and in the blood of intoxicated animals, indicating that mimosine is easily hydrolyzed in the rumen (Kudo et al., 1984). Mimosine is present in leucaena and if ingested by ruminants in large quantities can have a toxic effect or cause death (Hammond, 1995). Among the most common symptoms attributed to mimosine intoxication are: alopecia, anorexia, weight loss, deep salivation, esophagus lesions, necrotic papillae in the rumen and reticulum, thyroid hyperplasia, and low levels of circulating hormone thyroxine (T_4), early embryonic mortality and perinatal death (Allison et al., 1992; Al-dehneh et al., 1994). Usually, these symptoms are evident when the mimosine levels in diet are > 0.015% live weight (Szyszka and Termeulen, 1985).

DEGRADATION OF MIMOSINE BY RUMINAL MICROORGANISMS

In 1991 was first isolated, from animals fed leucaena, a bacterium belonging to the genus *Clostridium* which had degrading capacity of 3-4 DHP (Domínguez-Bello and Stewart, 1991). Later, Allison et al. (1992) identified and characterized a bacterium isolated from the rumen of animals (Hawaii), capable of degrading 3-4 DHP and its isomer 2-3 DHP, which was named *Synergistes jonesii*.

Rincón et al. (1998); Rincón et al. (2003) in research conducted in the United States and Venezuela respectively, corroborated that the *S. jonesii* had degrading capacity of the metabolite derived of mimosine 3-4 DHP, causing isomerization that lead to the 2-3 DHP which is degraded by the same bacteria, leading to fusion reactions and oxidation which yield propionic acid as final result. The formation of ornithine, which has two amino groups in their structure, as an intermediate product of the catalytic degradation of 2-3 DHP suggests the existence of transamination reactions.

When this bacterium was inoculated into the rumen of animals not adapted to the consumption of leucaena the toxic effects was reduced (Jones and Megarrity, 1986; Klieve et al., 2002; Palmer et al., 2010). However, despite inoculation, animals of various latitudes (e.g. Australia, Indonesia and Ethiopia) can only tolerate up to 30% of dietary leucaena (Yami et al., 2000; Dalzell et al., 2012). In such cases, there has been hypothesized that ruminal bacterial populations capable of degrading mimosine to DHP are saturated by the high intake levels of leucaena (mimosine). Large amounts of non-degraded mimosine is rapidly absorbed into the bloodstream, producing a severe impact on cell division that causes hair loss and sometimes fatal damage to internal organs (Prasad and Paliwal, 1989).

It is known that the bacteria *S. jonesii* has degrading ability of 3-4 and 2-3 DHP, however, has not confirmed their presence or absence in animals fed leucaena in other places where this plant is native. Furthermore, it is unknown whether there are some bacteria with mimosine degrading capacity, as the aforementioned bacteria only has ability to degrade the 3-4 and 2-3 DHP but not mimosine (Ram et al., 1994; Rincón et al., 1998; Domínguez-Bello et al., 1997; Rincón et al., 2003; Jetana et al., 2011). In addition, Dalzell et al. (2012) suggested that there may be other bacteria with degrading capacity for mimosine and its metabolites.

Degradation of mimosine is attributed to an enzyme named mimosinasa that is found in leucaena leaf, even though the mechanism of activation is not known, it is believed that at the moment of be chewed the leaf this enzyme it activated, hydrolyzing mimosine and yielding 3-4 DHP (Tangendjaja et al., 1986). It has also been reported the presence of hydrolytic activity in the rumen of animals fed leucaena; therefore, it is presumed that there are some bacteria that could degrade mimosine (Tangendjaja et al., 1983; Kudo et al., 1984; Rincón et al., 2003).

Synergistes jonesii

The *S. jonesii* is a strictly anaerobic gram-positive bacterium. Molecular techniques for the detection and sequencing of *S. jonesii* give greater knowledge of the geographical distribution and genetic variability of bacteria. Four strains have been isolated from the ruminal sample that was carried to Australia (extracted from a Hawaiian goat in 1982), including the strain designated as: 78,1 (ATCC 49833) and other 3 types (100-6, 113-4, 147-1) (Allison et al., 1992). The strains were shown to have differential specificity for degrading 3,4 - DHP and 2,3- DHP (Jones, 1994).

Currently using PCR amplification with specific primers of *S. jonesii* of 16S DNAr (small subunit ribosomal - RNA single stranded), which was used as a molecular phylogenetic classification. The theoretical limit of detection of *S. jonesii* PCR is 10^3 cells/mL. However, a realistic amplification by PCR of DNA genomic of rumen liquid is 10^4-10^5 cells/ml due to co-precipitation of contaminants inhibitors (Halliday et al., 2013). These same authors mention that this problem was overcome by a second round of PCR (Nested PCR) in the initial PCR products, which increased the sensitivity of theoretical limits of detection. The sequencing of the amplified products (nested) not only confirmed the identity of *S. jonesii* but also mutations detected in this segment of the 16S gene. These changes appeared as discrete mutations or ‘Single nucleotide polymorphisms’ (SNPs), which can be correlated with their ability to degrade DHP relating to the strain type. The SNPs can be random or occur constantly in the same locus, named ‘hot points’ (Halliday et al., 2013). Jones (1994), mentions about the inconsistent ability of *S. jonesii* to degrade DHP of rumen fluid of animals from different locations (and continents). The question remains whether these molecular changes also modify the ability of these strains to degrade DHP.

TOXIC EFFECT OF METABOLITES DERIVED MIMOSINE (3.4 AND 2.3 DHP) IN RUMINANTS

3-4 DHP

The primary metabolite of mimosine is the compound 3-hydroxy-4(1H)-pyridine (3-4 DHP) (Hegarty et al., 1976), in the presence of certain rumen microbes, can be further converted into its isomer 2,3-dihydroxypyridine (2-3 DHP) (D'Mello, 1992). The DHP is a potent goitrogenic, because of its peroxidase activity. By inhibition of the peroxidase and lactoperoxidase catalyzed reactions essential (Christie et al., 1979; Lee et al., 1980), the iodination of tyrosine in the binding step is inhibited in the thyroid. This step is crucial to the synthesis of thyroidal hormones, as the thyroxine (T_4) and triiodothyronine (T_3), resulting in levels of T_4 being depressed, causing overstimulation and expansion (up to 4 times) of the thyroid gland (goiter) (Hegarty et al., 1979; Jones, 1979; Megarrity and Jones, 1983). Studies have also shown that continuous intake of DHP increases the uptake of iodine into the hyperplastic thyroid, confirming the antithyroid effects of DHP (Hegarty et al., 1979). One effect of the low levels of T_4 and T_3 is the reduction of appetite, and ultimately a reduction in live weight gain. The T_4 levels lower than 13 nmol/L can even cause death (Jones et al., 1978).

The reduced availability of iodine due to DHP can also affect the salivary glands. Confounding the goitrogenic effects of DHP, is the fact that both isomers also chelate metal ions (Tsai and Ling, 1971), the complex form with Zn, Cu and Fe, lead to excretion and depletion of these minerals (Ghosh and Samiran, 2007). Zinc deficiency was shown to be responsible for skin lesions (Mills, 1978; Paul, 2000), increased salivation (Mills, 1978; Puchala et al., 1996) and abnormal hair growth (Hashiguchi and Takahashi, 1977). The Zn deficiency may also be responsible for the inhibition of DNA replication (Perry et al., 2005) and may adversely affect spermatogenesis (Yamaguchi et al., 2009). All this suggests that chelation of essential minerals is an important toxic effect of DHP. Deficiencies of essential minerals in the diet can accelerate the manifestation of clinical signs of toxicity arising from the intake of leucaena. Nevertheless, the toxicity of

DHP is a function of both the amount of leucaena in the diet as of length of time ingesting leucaena. Clinical symptoms may take up to 8 weeks to manifest (Quirk et al., 1988). Although there may also be excretion of DHP in the urine in large quantities without clinical sign of goiters (Phaikaew et al., 2012; Halliday et al., 2013b).

2-3 DHP

The isomer 2-3 DHP was originally thought to be transient, and indicative of incipient colonization of *S. jonesii* in the rumen (Ford et al., 1984; Jones et al., 2009). Recent data from Australia, Thailand and Indonesia contradict this notion (Dalzell et al., 2012; Phaikaew et al., 2012; Graham et al., 2013; Halliday et al., 2013b) and indicate that it is usually the predominant isomer in ruminants fed leucaena during long periods of time. Phaikaew et al. (2012) reported markedly high levels (> 1000 mg/L) of 2-3 DHP in the urine of ruminants fed for extended periods with leucaena (> 3 months). In general, it is considered that 3-4 DHP and 2-3 DHP are equally harmful (Lee et al., 1980; Ghosh et al., 2008). The latter has been shown that reduces the intake (McSweeney et al., 1984), and reduces milk production in dairy cows (Ghosh et al., 2007), and can be fatal if administered intraruminally in its pure form (Puchala et al., 1995).

In a study conducted in Australia, steers fed diets with 25 to 100% of leucaena excreted 2-3 DHP in urine at levels greater than 100 mg/L two weeks after the beginning of leucaena intake and it was until the seventh week that DHP excretion decreased (Halliday et al., 2013a). Gupta et al. (1992) reported that mimosine levels between 0.8 and 2.3% were to be considered as potentially toxic to animals. However, Kudo et al. (1990) mentioned that despite high levels of mimosine, their presence in feces might be low, because both mimosine and its metabolites derivatives can be rapidly absorbed. Additionally urinary excretion of these compounds is fast, which may explain why their toxic effects in ruminants are not frequently reported (as long as intake is lower than 1.0 g/mimosine/kg $LW^{0.75}$ or lower than a 30% inclusion level of leucaena) (Adejumo and Ademosun, 1991). The geographical variation in toxicity arising from leucaena intake reported by Jones (1981) was attributed to the presence or absence of bacteria capable of degrading the 3-4 DHP. Furthermore, Dominguez-Bello and Stewart (1991) indicate that there were rumen bacteria capable of degrading and/or utilizing this compound when its concentration in the diet is not high.

The cumulative levels and varying proportions of 2-3 DHP in ruminants, both previously exposed and not exposed to *S. jonesii*, may suggest that: a) rumen microbes other than *S. jonesii* are capable of degrading 3-4 DHP and 2-3 DHP. b) the *in vitro* inoculum of S. jonesii produced in Australia may have lost effectiveness or the strains have mutated. c) there are other environmental factors (including gene regulation) involved in metabolism of DHP that affect the optimum ability of *S. jonesii* to fully degrade this compound (Halliday et al. 2013a).

PERSPECTIVES

New in vitro studies report that some bacteria such as: *Streptococcus bovis* and *Klebsiella spp* are capable of degrading mimosine and its isomers (Chhabra et al., 1998; Aung et al., 2011). However, the *in vivo* degradation of mimosine and DHP has not been demonstrated. In Mexico, there have been studies that show that ruminants can tolerate the intake of leucaena in a high proportion (up to 50-60% inclusion) without negative impacts on animal health which could be attributed to DHP (Ruiz-González et al., 2011; Arjona-Alcocer et al., 2012; Contreras-Hernández et al., 2013; Ruz-Ruiz et al., 2013). Although Leucaena is native from the Yucatan Peninsula, Mexico, there are no studies demonstrating the presence/absence of *S. jonesii* in ruminants of this region.

Leucaena have potential for increasing productivity and sustainability of farming systems using ruminant in the tropics either as a high quality forage or possibly contributing in the reduction of greenhouse gases emissions. However, there is much skepticism in its widespread use due to some factors as discussed above. A strategy to potentiate the nutrient supply from this plant to ruminants, it is the association with local feed as energy sources, and strategy that have obtained positive results in ruminant production (Valdivia, 2006). Thus, in silvopastoral systems (with sheep and cattle), increased LW gain and milk yield been reported (Wencomo, 2008; Barros-Rodríguez et al., 2012).

From this background and the limted information on rumen ecology of animals consuming in Mexico (cow, sheep, and goat), the following questions are raised: Is there, in the rumen of animals in Mexico, a microbial consortia that promote the degradation of mimosine and its metabolites? Does intake of leucaena induces changes in the ecology of the rumen affecting digestion and nutrient utilization? Does diets based in leucaena cause a reduction of methanogenic archaea?

Finally, the interaction of secondary compounds contained in Leucaena with different compounds contained in tropical products such as *Enterolobium cyclocarpum* fruits (rich in saponins) (Hess et al.,

2003) might have further potential to modify rumen microbial population and activity. The *in vivo* synergism or antagonism effects of secondary compounds when simultaneously fed to ruminants deserve further studies.

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