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# **Effects of supplementing local plants on rumen fermentation, microbial protein synthesis, digestibility and voluntary feed intake in beef cattle steers**

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## **Paper I**

## **Paper II**

## **1. Introduction**

In the tropics and subtropics, majority of feed resources for ruminant consist of leftovers from the grain harvest, grasses and foliages growing on roadsides or waste land. The grasses are generally with high fiber and low protein contents. This results in poor animal performance,

especially in the dry season. However, alternative feed resources and crop-residues are locally available for used to increase livestock production in the tropical and subtropical areas. Cassava chip/pellets and dried leaves, baby corn stovers, kapok meal, cottonseed meal, broken rice and leuceana leaves are good example, while cereal crop, rice straw and urea-treated rice straw are excellent roughages for ruminants during the dry season. Efficient supplementation of locally mixed concentrate, grains or protein foliages, has been demonstrated to improve rumen ecology, dry matter intake and subsequently meat and milk quantity and quality (Wanapat, 1999). The extent to which tree foliage protein is degraded in, or escapes from the rumen is extremely important. If the tree foliage protein is totally degraded then it provides only ammonia and minerals for microbial growth (Leng, 1993). However, many tropical foliages or legumes contain secondary plant compounds, such as tannins and/or saponins, which may reduce their potential value as feeds, and there is an increasing awareness of the effects of these compounds on feed quality and animal production.

## 2. Objectives

- To investigate the level of CT and/or CS concentrations in local plants and their relationship with the end-products of fermentation by using the *in vitro* gas technique (Exp.I)
- To study the effects of crude condensed tannins and/or saponins in local plants on rumen microorganisms, fermentation end-products, microbial protein synthesis and feed digestibility using beef cattle steers (Exp.II)

## 3. General discussion

### 3.1 Feed resources for ruminants in the tropics

Feed resources available for livestock production in the tropics can be categorized into four groups : ([www.iaea.org/programmes/nafa/d3/public/review-1.pdf](http://www.iaea.org/programmes/nafa/d3/public/review-1.pdf))

#### 3.1.1 High fiber-low protein feeds

These include fibrous residues arising from crops grown for human consumption, such as straws and stovers from rice, millet, sorghum and maize, and sugarcane bagasse. Crop residues are characterised by their high fiber content (>700 g of cell wall material/kg DM), low metabolizable energy (<7.5 MJ/kg dry matter), low levels of crude protein (20.60 g of crude protein/kg DM) and minerals and low to moderate digestibility (<30.45% organic matter digestibility). Most residues are also deficient in fermentable carbohydrates, reflected by the relatively low organic matter digestibility.

#### 3.1.2 High fiber-high protein feeds

By-products derived from crop production (tops and haulms from ground nut, sweet potato vine, cassava leaves, bean straw) and industrial processing (bran from cereal milling: rice, wheat and maize bran, brewer's grain), fall into this category of feeds. They are generally less fibrous (below 700 but above 400 g of cell wall material/kg of DM) than those in the first category but have relatively high amounts of crude protein (> 60 g/kg DM). Leaves from tree legumes and browse plants such as *Glyricidia*, *Leucaena* and *Erythrina*, that have around 250 g/kg of crude protein in DM, can also be considered in this category.

#### 3.1.3 Low fiber-low protein feeds

These include feed resources derived from crops grown for renewable energy such as sugarcane by-products and root crops. They are generally rich in energy and low in protein content. Examples of this category would be molasses, oil palm juice and waste material arising from the fruit processing industry (citrus pulp, pineapple waste) and root crops (cassava).

#### 3.1.4 Low fiber-high protein feeds

These are the feeds traditionally called *concentrates* and include oilseed meals and cakes (coconut cake, soybean meal, cotton seed cake, groundnut meal/cake) and animal byproducts (fishmeal, blood meal, feather meal). They are valuable sources of good quality protein for both ruminant and non-ruminant animals. Oil seed meals and cakes may contain variable amounts of crude protein: coconut meal contains around 200 g crude protein/kg of dry matter while decorticated oil seed meals such as groundnut meal, and cotton seed meal (or cake) may contain as much as 400 g of crude protein/kg of dry matter.

According to the above system most of the substrates, which used in paper I fall into the second category, except for bitter cucumber, green banana fruit and rice straw fall into the third and first category, respectively.

#### 3.2 Some examples of using local feed resources as livestock feeds

##### *Crop-residues*

Wanapat *et al.* (1985) successfully demonstrated the efficient use of urea-treated rice straw as a roughage source when supplemented with various level of concentrate in fattening beef cattle. It was also confirmed on-farm that the use of urea-treated rice straw as a sole source of roughage for fattening beef proved to be economical in the prevailing village system (Wanapat *et al.*, 1992).

Preston and Leng (1987) reported that ensiling chopped sugarcane-tops (*Saccharum officinarum*) with urea improved digestibility of nutrients. Other researcher concluded that sugarcane-tops silage had a similar nutritive value to dried sugarcane-tops (Sritakoonpech, 1990). Moreover, Yuangklang *et al.* (2005) studied on alternative ways to use sugarcane-top, and suggested that dried-chopped and pelleted sugarcane-top increased digestibility and feed intake in beef cattle.

##### *By-products of agricultural industry*

The early work by Preston and Willis (1974) demonstrated that diets containing a large proportion of molasses and small inputs of bypass protein dramatically increased growth rate and feed efficiency of cattle.

Studies using extracted rice bran by Wora-anu *et al.* (2000) concluded that urea-treated rice straw and extracted rice bran, at 40:60 ratio, gave better results in terms of intake and digestibility in both cattle and buffaloes.

In addition, Mungbean bran, kapok meal and cotton seed meal have been shown to be comparable to soybean meal when supplemented in heifer rations (Promkot, 2003).

##### *Forage and tree fodders*

Cassava hay (*Manihot esculenta*, Crantz) was demonstrated to be an excellent foliage for feeding to ruminants in the dry season as earlier reported by Wanapat *et al.* (1997). Higher level of cassava hay supplementation especially at 2-3 kg/hd/d, improved efficiency, reduced production cost and increased profits (Wanapat *et al.*, 2000). However, Wora-anu *et al.* (2004) observed that feeding fresh cassava foliage as full-feed resulted in anorexia and ataxia, as well as frequent urination.

*Leucaena leucocephala* leaves have been widely used in diets of all animal species. Cheva-Israkul (1988) reviewed the use of leucaena as a feed. Leucaena can be fed fresh (Wongsrikeao and Wanapat, 1986) or dried (Sarawish *et al.*, 1988). Sarawish *et al.* (1988) reported that supplementing the rice straw diet of growing native cattle with dried leucaena leaves produced good results.

In work on mulberry foliage (*Morus alba*) Liu *et al.* (2001) reported that the benefits resultant from supplementation with mulberry leaves in sheep diets included an increased intake of the basal diet, lower consumption of concentrate and an increased income.

Using *Samanea saman* as a feed at 2 kg samanea pods to a basal rice straw diet of buffaloes enabled body weight to be maintained throughout the dry season (Seedtakosed *et al.*, 1988)

However, Leng and Preston (1985) suggested that in designing a diet based on crop residue or by-product, optimizing animal productivity rather than designing the diet to meet productive requirements should be considered.

### 3.3 Plant secondary compounds

There is substantial current research activity to evaluate the potential of secondary plant constituents as natural means of modifying ruminal fermentation. Plants contain various secondary compounds which protect them from attack by fungi, bacteria, herbivorous insects and vertebrates. Classes of compounds known to act in this way include saponins and tannins (Makkar *et al.*, 1995; Pell *et al.*, 2001), which are prevalent in many tropical fodder plants.

#### 3.3.1 Tannins

Tannins (polyphenols) are a significant group of the secondary plant ingredients. According to the classical definition by Bate-Smith and Swain (1962), tannins are water-soluble phenols with a molecular weight between 500 and 5,000 which apart from phenolic reactions have the property of precipitating alkaloids, gelatins and proteins (Leinmuller *et al.*, 1991). Polyphenols do not constitute a unified chemical group, but have a large variety of molecular structures; in general they are subdivided in hydrolyzable and condensed tannins (McLeod, 1974).

Hydrolyzable tannins are polyester compounds of carbohydrates such as glucose and the phenolated carboxylic acids of gallic acid (gallotannin) or of hexahydroxydiphenyl acid (ellagitannin).

Condensed tannins (proanthocyanidins, CT) are polymeric compounds of flavan-3-ol (catechin) or flavan-3,4-diol (leucoanthocyanidin) or their derivatives, which are linked by C-C or C-O-C bonds. Condensed tannins polymerize on heating in acids to the amorphous phlobaphenes (Haslam, 1989). The linkage between the monomers, typically a carbon condensation, is relatively stable under the conditions which cleave ester linkages in hydrolyzable tannins.

#### *Tannins in ruminant feedstuffs*

Tree foliages represent an important source of cellulosic biomass for feeding ruminants throughout the world. Fodder from trees and shrubs have been an important source of protein for grazing animals. In plant species that contain tannins, maceration of the plant causes the tannins which are in the vacuoles to be released and these subsequently bind to the plant material, with tannins binding more strongly to protein than to fiber (McLeod, 1974). Some plants probably evolved tannins production as a defense strategy, first against invasion by bacteria and fungi, then against being eaten by insects and finally against being eaten by herbivores, with the

mechanism being related to the protein-precipitating properties of tannins (Barry, 1989; Terrill *et al.*, 1992).

Animals respond differently to dietary tannins in part because of the variation in the biological activity of the tannins themselves (Reed, 1995; Makkar *et al.*, 1995). It has been believed for some considerable period that tannin above 5 % can become a serious anti-nutritional factor in plant materials fed to ruminants (McLeod, 1974). If the protein-tannin complex dissociates under acid conditions then the protein can be digested in the lower gut. Barry (1983) and his colleagues have demonstrated with *Lotus pedunculatus* that the ideal concentration of condensed tannins in this forage legume is between 2-4 % of the diet dry matter, at which level they bind with the dietary proteins during mastication and appear to protect the protein from microbial degradation in the rumen. Thus a low level of tannin in plant materials has been usually accepted as being able to protect protein of forages and allow a higher efficiency of feed utilization by the animal. However, recent results throw some doubt on this. Tannins may indirectly effect rumen function by reducing rumen ammonia levels to below a critical concentration in the rumen of 80 mg N/l, and then fiber digestibility may be depressed and digestibility is reduced well below 10 mg N/l (Leng, 1993).

#### *Effects of tannins on nutritive value of forages*

Tannins in forage have both negative and positive effects on nutritive value (Reed *et al.*, 1990; Mueller-Harvey and McAllan, 1992). Tannins in high concentrations reduce intake, digestibility of protein and carbohydrates, and animal performance (Reed *et al.*, 1990). Tannins in low to moderate concentrations prevent bloat and increase the flow of non-ammonia nitrogen and essential amino acids from the rumen (McNabb *et al.*, 1993). The positive effects of tannins on protein utilization have practical importance because problems associated with extensive proteolysis and(or) deamination in the rumen limit production in modern feeding systems (Beever *et al.*, 1989).

#### *Negative Effects*

*Intake.* Tannins may reduce intake of forage legumes by decreasing palatability or by negatively affecting digestion. Astringency is the sensation caused by the formation of complexes between tannins and salivary glycoproteins. Astringency may increase salivation and decrease palatability. Waghorn *et al.* (1994a) suggested that decreased ruminal turnover and rate of digestion was more important than palatability in reducing intake of sheep fed pure diets of *Lotus pedunculatus* in comparison to sheep fed *L. pedunculatus* along with polyethylene glycol ( PEG) the latter binds the tannins making them relatively inactive in the rumen.

*Growth.* Rate of gain of young animals reflects total intake and availability of nutrients in the diet. Low growth rates because of low total feed intake were observed in animals eating fruits of *A. sieberiana* and *A. nilotica*, which contained high levels of tannins (Tanner *et al.*, 1990). Low total intake and low growth rates were also observed in animals eating *A.sieberiana* pods and leaves of *A. cyanophylla* (Reed *et al.*, 1990). The negative effect of tannins on growth rate was caused by a combination of reduced intake and low true digestibility of protein.

*Digestion of fiber fractions.* Tannins may reduce cell wall digestibility by binding bacterial enzymes and(or) forming indigestible complexes with cell wall carbohydrates (Reed *et al.*, 1990). Digestibility of organic matter and fiber fractions was lowest for sheep fed *A. cyanophylla*, the supplement with the highest content of CT and soluble phenolics. Digestibility of ADL was negative for all three acacias foliages in this trial (Reed *et al.*, 1990). At high levels (5-9 %) tannins become highly detrimental (Barry, 1983) as they reduce digestibility of the fiber in the rumen (Reed *et al.*, 1985) by inhibiting the activity of bacteria and anaerobic fungi

(Chesson *et al.*, 1982). High levels also leading to reduced feed intake (Akin and Rigsby, 1985), and above 9 % tannins may become lethal to an animal that has no other feed (Kumar, 1983).

### *Positive Effects*

*Rumen Escape.* Tannins may complex protein at the pH of the rumen and protect protein from microbial enzymes. These complexes are unstable at the acid pH of the abomasum and the proteins become available for digestion ( Barry and Manley, 1984). The evidence for the stability of tannin-protein complexes in the ruminal environment comes from highly simplified *in vitro* systems with purified proteins and tannins in the absence of ruminal microorganisms (Jones and Mangani, 1977). However, a tannin-protein complex that survives the ruminal environment may or may not be digested in the lower tract (Waghorn *et al.*, 1994b).

*Urea Recycling.* Tannins may increase the efficiency of urea recycled to the rumen. Tannins lower the rate of protein degradation and deamination in the rumen and therefore lower ruminal NH<sub>3</sub>-N (Woodward, 1989). Plasma urea nitrogen, ruminal NH<sub>3</sub>-N, and urinary N loss were lower when sheep and goats were fed legumes that contained tannins (Woodward, 1988). Tannins may increase the glycoprotein content and excretion of saliva, which could lead to more N recycled to the rumen (Robbins *et al.*, 1987). Sheep fed pure diets of *L. pedunculatus* had lower plasma urea concentrations, a more rapid plasma urea turnover rate, and a higher irreversible loss than sheep receiving *L. pedunculatus* that was treated with PEG to “deactivate” the tannins (Waghorn *et al.*, 1994b). However, effects of mangosteen peel that contains 16.8% tannins supplementation on NH<sub>3</sub>-N concentrations and urinary N loss were not found in Paper II.

*Microbial Efficiency.* Beever and Siddons ( 1985) and Reed *et al.* (1990) hypothesized that tannins increase microbial yield per unit of organic matter digested. Several researchers have observed increases in non-ammonia nitrogen (NAN) flows to the duodenum that were greater than N intake for forage legumes that contain tannins. Because N is not created in the rumen, part of the increased flow of NAN must be from endogenous sources that have been incorporated into the microbial fraction. Nitrogen flows at the duodenum that are greater than N intake are common for diets low in N (< 1%), but for forage legumes with greater than 2% N, the N flows at the duodenum are normally lower than N intake (Barry and Manley, 1984).

Gas production in paper I were observed to be lowest for the high CT content, which may due to lower digestion by rumen microbes. Moreover, no effect of CT was found on voluntary feed intake, live weight change and digestibility when beef steers were supplemented with mangosteen peel (*Garcinia mangostana*) that contained tannins. However, the efficiency of rumen microbial protein synthesis and P/E ratio were slightly higher in steers fed on mangosteen peel than the control group.

### 3.3.2 Saponins

Saponins are a vast group of glycosides, widely distributed in higher plants. Saponins can be classified into two groups based on the nature of their aglycone skeleton. The first group consists of the steroidal saponins, which are almost exclusively present in the monocotyledonous angiosperms. The second group consists of the triterpenoid saponins, which are the most common and occur mainly in the dicotyledonous angiosperms (Bruneton, 1995). Some authors distinguish a third group called steroidal amines, which are classified by others as steroidal alkaloids (Bruneton, 1995). For the purpose of this review, only steroidal and triterpenoid saponins were considered. Steroidal saponins consist of a steroidal aglycone, a C27 spirostane skeleton, generally comprising of a six-ring structure. In some cases, in fresh plant material, the hydroxyl group in the 26-position is engaged in a glycosidic linkage, and so the aglycone structure remains pentacyclic. This is referred to as a furostane skeleton. Triterpenoid saponins consist of a triterpenoid aglycone, which consists of a C30 skeleton, comprising of a pentacyclic structure.

## *Saponins and ruminants*

Interest in saponins in relation to ruminants has taken two main forms, coming from two different directions. The first involved the assessment of saponins, as chemical modulators, for their effects on rumen fermentation potential feed additives of unknown value while the second started from the objective of finding natural materials which might be used to suppress the growth of rumen ciliate protozoa. What has happened is that the two approaches have merged to a significant extent, because one of the main effects of saponins on rumen fermentation is that they are toxic to protozoa.

Van Nevel and Demeyer (1990) in a study of sarsaponin *in vitro* showed no indication of toxic effects or effects on microbial growth or protein breakdown. In contrast, Lu *et al.* (1987) discovered that alfalfa saponins appeared to suppress fermentation in continuous culture. Subsequent *in vivo* investigations (Lu and Jorgensen 1987) confirmed a general decrease in fermentative activity when alfalfa saponins were supplied to the sheep rumen, of which decreased VFA concentrations and decreased cellulose digestion were symptomatic. Significantly, Lu and Jorgensen (1987) also noted large decreases in protozoal numbers in sheep receiving alfalfa saponins. Goetsch and Owens (1985) concluded that the benefits of sarsaponin would be diet dependent, increasing the digestion of sorghum silage and other fibrous feeds but apparently decreasing digestion of cereal and protein meals.

Removal of rumen ciliate protozoa, or defaunation, has been an objective of rumen microbiologists for a generation. There are many consequences for the fermentation, and consequently for nutrition, that result from the removal of protozoa (Williams and Coleman 1992). Anti-protozoal agents, such as surface-active agents, that have been investigated in attempts to apply defaunation at the farm level, have been hampered by problems with toxicity, either to other ruminal microorganisms (Eadie and Shand 1981; Orpin 1977; Bird and Leng 1978; Bird *et al.*, 1979;) or to the host (Lovelock *et al.*, 1982). Lipids are toxic to protozoa (Machmuller *et al.*, 1998; Matsumoto *et al.*, 1991; Newbold and Chamberlain 1988) but also to cellulolytic microbes fibre digestion (Broudiscou *et al.*, 1994). Thus, there is no reliable, safe method available to suppress ruminal protozoa. Recently, some tropical plants were found to have the potential to be used as a safe possible means of suppressing or eliminating protozoa from the rumen (Diaz *et al.*, 1994; Navas-Camacho *et al.*, 1993; Newbold *et al.*, 1997; Odenyo *et al.*, 1997). These plants all had the characteristic that they were rich in saponins.

### *Effects of saponins on ruminal microorganisms*

Because dietary saponins are poorly absorbed, their biological effects occur in the digestive tract (Cheeke, 1996). Although antimicrobial effects of saponins and saponin-containing plants were to be expected from the wealth of information from other biological systems, among the earliest observations in ruminal microorganisms came relatively recently from *in vitro* continuous culture systems. Valdez *et al.* (1986) observed that sarsaponin, from *Yucca schidigera*, decreased protozoal numbers but not bacterial numbers in a 22-d semi-continuous system, and Lu *et al.* (1987) found that the bacterial population was changed in the presence of alfalfa saponins from a morphologically diverse one in controls to one in which fewer morphotypes were present in vessels receiving saponins. As well as the saponins having an effect on ruminal microorganisms, the microorganisms can metabolise the saponins, thus introducing another factor to be considered in the application of saponins to ruminant nutrition.

### *Protozoa*

Numerous studies have now demonstrated that saponins and saponin-containing plants have toxic effects on protozoa. *In vitro*, toxicity of *Y. schidigera* extract towards protozoa has been noted from a fall in numbers in fermenters (Makkar *et al.*, 1998; Wang *et al.* 1998) or in bacteriolytic activity (Wallace *et al.* 1994). Butanol extraction of the *Y. schidigera* extract resulted in all anti-protozoal activity being located in the butanol fraction, consistent with the active component being saponins. Saponins from *Quillaja saponaria* and *Acacia auriculoformis* (Makkar *et al.*, 1998) and foliage from *Sesbania sesban* (Newbold *et al.*, 1997) were also anti-protozoal *in vitro*, the *S. sesban* active component again being extractable in butanol. *In vivo*, supplementastion with powdered *Y. schidigera* decreased rumen protozoal numbers in heifers (Hristov *et al.*, 1999). A decrease in protozoal numbers was reported in the rumen of sheep infused with pure alfalfa saponins (Lu and Jorgensen, 1987) or fed saponin-containing plants, including *S. sesban* (Newbold *et al.*, 1997; Odenyo *et al.*, 1997) and *Enterolobium cyclocarpum* (Navas-Camacho *et al.*, 1993).

The sensitivity of ciliate protozoa towards saponins may be explained by the presence of sterols in protozoal, but not bacterial membranes (Williams and Coleman 1992). Thus, the sterol-binding capability of saponins (Hostettmann and Marston 1995) most likely causes the destruction of protozoal cell membranes.

### *Bacteria and fungi*

In mixed cultures and *in vivo*, saponins have been shown also to affect ruminal bacteria. Newbold *et al.* (1997) found that bacterial numbers increased when foliage from *S. sesban* was introduced into the diet, presumably as a consequence of the suppression of protozoal numbers. Valdez *et al.* (1986) found a similar trend with *Y. schidigera* extract. Steroidal saponins from *Y. schidigera* had no effect on total or cellulolytic bacterial counts in Rusitec; however, inoculating fluid from the fermenter into medium containing saponins decreased the viable count (Wang *et al.* 1998).

Pure cultures also indicate possible antibacterial effects of saponins. *Y. schidigera* extract abolished growth of the fibre digester, *Butyrivibrio fibrisolvens*, and prolonged the lag phase of *Streptococcus bovis* (Wallace *et al.* 1994). Similar sensitivity of *S. bovis* to *Y. schidigera* extract was found by Wang *et al.* (2000), who additionally found that cellulose digestion by *Ruminococcus* spp. and *Fibrobacter succinogenes* was inhibited.

A potentially very significant observation is that the anaerobic ruminal fungi, *Neocallimastix frontalis* and *Piromyces rhizinflata*, were highly sensitive to *Y. schidigera* saponins (Wang *et al.*, 2000). Ruminal fungi appear to fill an important niche in the digestion of recalcitrant plant fibres, because they cause physical as well as enzymatic disruption of plant cell walls (Orpin and Joblin, 1997). The results from this experiment (paper II) provided similar effects. The mangosteen peel (*Garcinia mangostana*) supplement, that contains tannins, increased the bacterial population. Protozoal population were decreased in all treatments with supplementation of mangosteen peel when compared with the control group, while fungal zoospore populations were not affected. However, increasing the level of mangosteen peel supplementation tended to change rumen the microbial population.

### *Saponins and ruminant production*

There is no doubt, therefore, that saponins have selective effects on ruminal microorganisms that might be useful in livestock production. A safe, persistent suppression of ciliate protozoa may have the widest application. Ciliate protozoa are primarily responsible for the substantial turnover of bacterial protein which occurs during rumen fermentation (Ushida *et al.*, 1991; Wallace and McPherson 1987; Williams and Coleman 1992). As a consequence, nitrogen retention is improved by defaunation, which has been amply demonstrated in many

studies where the protozoa were removed by chemical or physical means, or where the animals had been isolated from birth and thus had not been colonised by protozoa (Williams and Coleman 1997). The argument in favour of defaunation depends on other factors as well, however. As some species of protozoa are cellulolytic, there are implications for fiber breakdown of removing protozoa (Demeyer and Van Soest 1986; Kayouli *et al.*, 1984). Also, the protozoa are proteolytic, so there would be consequences there too (Ushida *et al.*, 1991). However, it is generally agreed that removing or suppressing protozoa would make the best use of nitrogenous resources, particularly on low-protein diets.

Effects of saponins on the bacterial population merit further examination. Wang *et al.* (2000) suggested that *Y. schidigera* extract would be best used with high grain diets, because of its suppressive effect on *S. bovis* is a starch-digesting, lactate-producing Gram-positive species which is a major cause of rumen fermentation lapsing into lactic acidosis (Stewart *et al.*, 1997). Caution may be required in more fibrous diets, however. The suppression of those bacteria involved in fiber digestion, as described earlier, could have serious consequences to overall digestion (Preston and Leng, 1987).

In animal feeding trials, there have been mixed observations concerning fermentation and productivity. Lu and Jorgensen (1987) found that alfalfa saponins caused a decrease in the efficiency of microbial protein synthesis in sheep, because the growth of bacteria as well as protozoa was depressed. A 36% fall in the efficiency of protein synthesis occurred in cattle receiving *Y. schidigera* extract (Goetsch and Owens, 1985). In contrast, inclusion of *E. cyclocarpum* foliage increased the rate of body weight gain in sheep by 24% (Leng *et al.*, 1992) and 44% (Navas-Camacho *et al.*, 1993) and wool growth by 27% (Leng *et al.*, 1992), which was attributed to an increased flow of metabolisable protein to the intestines when there is a decrease in protozoal numbers. These differences therefore imply that the effects of saponins on ruminant nutrition are complex, and depend on diet, and the saponins involved. General observations with saponins, where changes in ruminal fermentation characteristics occur, that saponins administration decreases  $\text{NH}_3\text{-N}$  concentration (Lu and Jorgensen 1987; Lu *et al.*, 1987; Makkar *et al.*, 1998) and, where VFA are affected, increases propionate concentration (Lu *et al.*, 1987; Hristov *et al.*, 1999) are typical effects of decreased protozoal numbers (Williams and Coleman 1992). According to the results in paper II, supplementation of mangosteen peel (*Garcinia mangostana*) that contains saponins in steers diets improved rumen ecology by reducing protozoal and increasing bacterial populations. However, ruminal  $\text{NH}_3\text{-N}$ , BUN, TVFAs, digestibility and voluntary feed intake were not affected.

#### **4. Conclusions**

Based on this experiment, it can be concluded that local feed resources which have high levels of CT and/or CS contents can be used as alternative dietary strategic supplements to improve rumen ecology and act as a defaunating source in ruminants. They would be particularly useful for small-holder farmers to improve feed efficiency and production, as well as being positive for the environment.

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