# Beef Production Based on Cassava Products and Legume Foliage in Vietnam

Chu Manh Thang
*Faculty of Veterinary Medicine and Animal Science*
*Department of Animal Nutrition and Management*
*Uppsala*

Doctoral Thesis
Swedish University of Agricultural Sciences
Uppsala 2010

Acta Universitatis agriculturae Sueciae
2010:23

Cover: Ohh, straw again !!!! I'd like something else…
(photo: Chu Manh Thang)

ISSN 1652-6880
ISBN 978-91-576-7500-2

Print: SLU Service/Repro, Uppsala 2010

# Beef Production Based on Cassava Products and Legume Foliage in Vietnam


## Abstract

The overall aim of this thesis was to determine the associative effects of supplemental sources of protein, or protein and energy, using cassava products and legume foliage on rumen degradability, *in vivo* digestibility, feed intake and growth rate of crossbred growing cattle.

First it was hypothesized that the substitution of a part of a conventional concentrate with mixed cassava and legume foliage will positively influence rumen degradability and improve the performance of growing cattle. The results showed that there were significantly higher values of rumen degradation with a mixture of cassava and *Phaseolus calcaratus* legume (CA-LE) feed compared to the control feed. With the same dry matter intake (DMI) of concentrate, the level of CA-LE feed did not have any effect on total DMI but live weight gains in CA-LE40 and CA-LE60 were significantly higher (551 and 609 g day$^{-1}$, respectively) than in the control group.

Secondly, it was hypothesized that a combined protein source will result in at least the same feed intake, digestibility and growth rate as in growing cattle fed concentrate. With the same nitrogen supplement, there were no significant differences in DMI, but 30% and 19%, respectively, higher live weight gain in the group fed a mixture of foliages as compared to the groups fed only cassava or stylosanthes.

The third hypothesis was that a synchronisation of energy from cassava meal and protein from the mixture of foliages would improve feed intake and growth rate of cattle. The highest nutrient digestibilities were found in the group fed both high crude protein and metabolisable energy (ME), resulting in a significantly higher daily gain, 558 g day$^{-1}$. In order to overcome the negative effect of hydrogen cyanide, cattle fed high amounts of cassava foliage should be supplied with extra energy in the diet.

The rumen degradation characteristics of some tropical legumes mixed with cassava foliage and cassava root meal were evaluated using the *in vitro* gas production technique. The potential gas production of the mixture samples, indicating the level of solubility and degradability of substrate in rumen, ranking from highest to lowest, were: cassava root meal with *Stylosanthes guianensis*, *Leucaena leucocephala*, *Phaseolus calcaratus*, *Flemingia macrophylla*, and *Trichanthera gigantea,* and cassava foliage with *Stylosanthes guianensis*, *Phaseolus calcaratus*, *Leucaena leucocephala*, *Flemingia macrophylla*, and *Trichanthera gigantea*. The highest estimated values of organic matter digestibility, short chain fatty acids and ME were in cassava root meal mixed with legumes at the level of 75%. There was a negative relationship between ash content and potential gas production in individual feed substrates. No effect of hydrogen cyanide on fermentation kinetics in rumen could be detected in this study.

*Keywords:* Cassava foliage, legume foliage, balanced protein-energy, disappearance, degradability, feed intake, digestibility, growth rate, growing cattle.

*Author's address:* Chu Manh Thang, Department of Science and International cooperation, National Institute of Animal Science – Thuy Phuong, Tu Liem, Hanoi, Vietnam
*E-mail:* cmthang76@gmail.com

# Dedication

*To my parents*
*My wife Pham Thi Bich Huong*
*My daughters Chu Thuy Linh*
*Chu Dieu Huong*

# Contents

# List of Publications

This thesis is based on the following papers, referred to by Roman numerals in the text:

I Thang, C. M., Sanh, M. V. and Wiktorsson, H. (2008). Effects of supplementation of mixed cassava (*Manihot esculenta*) and legume (*Phaseolus calcaratus*) fodder on the rumen degradability and performance of growing cattle. *Asian-Australasian Journal of Animal Science* 21(1), 66-74.

II Thang, C. M., Ledin, I. and Bertilsson, J. (2010). Effect of feeding cassava and/or Stylosanthes foliage on the performance of crossbred growing cattle. *Tropical Animal Health and Production* 42 (1), 1-11.

III Thang, C. M., Ledin, I. and Bertilsson, J.(2010). Effect of using cassava products to vary the level of energy and protein in the diet on growth and digestibility of cattle. *Livestock Science* 128 (1-3), 166-172.

IV Thang, C. M., Ledin, I. and Bertilsson, J. Degradation characteristics and fermentation kinetics of some tropical legumes and cassava foliage/root determined by using the *in vitro* gas production technique (submitted).

Papers I-III are reproduced with the permission of the publishers.

# Abbreviations

| | |
|---|---|
| AA | Amino acids |
| ADF | Acid detergent fibre |
| $BW^{0.75}$ | Metabolic body weight |
| CaF | Cassava foliage |
| CaM | Cassava root meal |
| CA-LE | Mixture of cassava and *Phaseolus calcaratus* foliage (ratio of 3:1) |
| CF | Crude fibre |
| CP | Crude protein |
| CTs | Condensed tannins |
| DM | Dry matter |
| DMI | Dry matter intake |
| DOM | digestible OM |
| EE | Ether extract |
| FCR | Feed conversion ratio |
| Fle | *Flemingia macrophylla* |
| GE | Gross energy |
| HCN | Hydrogen cyanide |
| Leu | *Leucaena leucocephala* |
| LWG | Live weight gain |
| ME | Metabolisable energy |
| NDF | Neutral detergent fibre |
| OM | Organic matter |
| OMD | Organic matter digestibility |
| Pha | *Phaseolus calcaratus* |
| SCFA | Short chain fatty acids |
| SCN | Thiocyanate |
| Sty | *Stylosanthes guianensis* |
| Tri | *Trichanthera gigantea* |

| | |
|---|---|
| URTRS | Urea treated rice straw |

# 1 Introduction

In Vietnam meat and milk production of cattle have been included in development programs put forward by the government with the aim and commitment to increase production and improve living standards in rural areas. The quantity of meat and milk produced as well as the number of slaughtered cattle have increased over the last ten years (FAOSTAT data, 2008). Total beef production in 1998 was 79,000 tonnes from 520,000 head slaughtered. The number of cattle slaughtered in 2008 increased to 1,200,000 head, with a total beef production of 206,145 tonnes. The climate in Vietnam, with 5-6 months of dry season, leads to scarcity of high quality roughage resources. In the suburbs, families who rear cattle have to set aside a certain area of land for growing grasses, as by-products of agricultural crops have been inadequate. The basic fodders available for cattle are low in nutritive value and the seasonal fluctuation in supply makes the farmers much more dependent on concentrate supplements. These factors are limiting the dairy development in Vietnam (Cuong, 1993; Man, 2001).

Locally available foliages are considered to be potentially valuable protein supplements to low quality diets in tropical countries (Preston, 2001; Wanapat, 2001). Promising results have been obtained in the use of cassava foliage (Hong *et al.*, 2003; Khang & Wiktorsson, 2006) and utilisation of legume foliages from e.g. *Sesbania grandiflora* (Luc *et al.*, 2009) as a feed supplement for cattle.

Cassava production in Vietnam has increased steadily during recent years, mainly because of increases in both area planted and yield per hectare. The area cultivated in 2007 was 497,000 ha, with a total tuber yield of about 8 million tonnes. Traditionally, cassava is planted to produce roots for human consumption and small amounts of fresh cassava foliage, including the tender stem, are used directly for animal feeding (Van *et al.*, 2001). If cassava is grown for foliage in the dry season, it can give a DM yield of around 7.9 to

12 tonnes per ha of cassava hay (Hong *et al.*, 2003; Dung *et al.*, 2005; Thang, 2005). Crude protein (CP) content of cassava foliage ranged from 17.7 to 22.6% on dry matter (DM) basis (Khang & Wiktorsson, 2000; Man & Wiktorsson, 2001). However, up to the present time, the potential of cassava foliage as a protein source in ruminant feeds has not yet been fully exploited. The main reason for this is probably the content of hydrogen cyanide (HCN), which may affect animal health. Different kinds of processing, like sun drying or ensiling, will reduce the cyanide content of cassava foliage to a level which is safe for cattle (Khieu Borin *et al.*, 2005; Phengvichith & Ledin, 2007). These processing methods, however, depend on many factors, such as weather, available facilities etc. The combination of cassava foliage and a legume can be a possible solution, not only supplying more nutrients, but also maintaining a moderate level of cyanide in the diet when feeding cassava foliage.

*Objectives of the study:*

- To investigate the effect of supplementation of mixed cassava (*Manihot esculenta*) and legume (*Phaseolus calcaratus*) fodder on the rumen degradability and performance of growing cattle.
- To study the effect of feeding cassava and/or Stylosanthes foliage on the performance of crossbred growing cattle.
- To study the effect of using cassava products to vary the level of energy and protein in the diet on growth and digestibility of cattle.
- To compare the degradability characteristics and fermentation kinetics in the rumen of cassava products and some legume foliages and their mixes.

*Hypothesis*

- Replacing a part of a conventional concentrate with mixed cassava and legume foliage will positively influence rumen degradability and improve the performance of growing cattle.
- The combined protein source will result in at least the same feed intake, digestibility and growth rate as in growing cattle fed concentrate.
- Synchronisation of energy from cassava meal and protein from the mixture of foliages will improve feed intake and growth rate of cattle.

# 2 Background

## 2.1 Smallholder cattle production in Vietnam

Vietnam, a country dominated by agriculture, is situated on the Indo-China Peninsula. The country covers an area of 332,000 km$^2$, and stretches in the shape of the letter S for more than 1,600 km, forming the eastern edge of the Southwest Asian mainland. It shares borders with China to the north, Lao PDR to the northwest and Cambodia to the southwest. The general weather conditions throughout the country are those of a tropical climate. According to GSO (2009), Vietnam has a human population of 86.2 million people, of which over 72% are farmers who live in the rural areas, where agriculture provides the major source of income and livelihood. Livestock production is primarily undertaken on household farms where crop and other agricultural products are also produced. According to a survey (Nin *et al.*, 2003) in Vietnam, more than 92% of the producers utilize only household labour in livestock management (62% in general agriculture), and on average they cultivate 0.77 hectares of land.

### 2.1.1 Cattle breeds and breeding programs

The small Yellow Cattle and the Laisind breeds are the most common breeds of cattle in Vietnam. Yellow Cattle have low average body weights, about 180–200 kg for mature females (with a carcass percentage of 30%), and around 300 kg for bulls (Burns *et al.*, 2002). These cattle are extremely well adapted to the climate and the local feeding conditions and are fertile as well. Some breeding programs have been initiated in order to select beef cattle, including crossbred and native cattle, for sires and dams. Weights and body conformation were the criteria for selection. Since 1920 (during the French rule) the first program started with the crossing of Yellow Cattle with Red Sindh imported from Pakistan (Su & Binh, 2002). The authorities reported that the body weight of this "Laisind" crossbred was 30–35% higher, meat production 5–8% higher and draught power 20% higher compared to Vietnamese Yellow Cattle. The Red Sindh is often used by farmers as a first cross (F1) when attempting to increase the size of their animals. Although fertility rates in these cattle are good the growth rates and profit margins are typically low. Later the breeding program focused on fattening and economic comparisons of beef breeding in different economic zones. The national program under the Beef Cattle Development Project-VIE/86/008 lasted between 1989–1991. Some new crosses were introduced, with *Bos taurus* breeds, such as Brahman or Sahiwal crossed with the "Laisind" breed. In 1996 a program entitled Profitable Beef Cattle Development in Vietnam (AS2/97/18) was started by the Australian Centre for International Agricultural Research (ACIAR). An important goal of this project was to find outcomes which increase the profitability of cattle rearing by smallholder farmers, rather than assessing results only in terms of physical production or productivity measures. This project has focused on developing a crossbreeding program to produce a mid-sized, 'easy care' animal with good growth and good fertility, while remaining well-adapted to the local environmental stresses (Burns *et al.*, 2002).

### 2.1.2 Cattle farms, type and management

Due to the limited land area and fodder availability, the number of cattle per small household in the lowlands is less than five (Huyen *et al.*, 2009). Also Tung *et al.* (2007) reported that the mean herd size for beef cattle in north-western Vietnam is 3.4 animals per farm. They found that nearly 50% of the households kept fewer than three cattle, followed by households that kept three to five (41%) and a few households that kept between six and ten animals (8%). The small farm consisted mainly of arable land, with cattle as a complementary activity and with limited fodder resources for these cattle. In a recent survey, Huyen *et al.* (2009) reported that the availability of feed resources for cattle was a major factor affecting herd size and cattle management on smallholder farms in the northern part of Vietnam.

In small farms, cattle are managed on a low-intensity basis, utilising mixed feed resources of variable quality. Traditionally, cattle are bred chiefly to serve crop production and are kept mainly in the cropping areas. Due to limited arable land, priority is usually given to crops, and there is little grazing land available for ruminants (Trach, 1998). They are usually allowed only strictly controlled access to grazing on roadsides, fallow land and the raised boundaries between the rice fields and other cropping areas. In some mountainous areas, during the winter season when the weather is humid and cold, the cattle are fed almost exclusively on rice straw and also given a little homemade concentrate. The reduced availability and poor quality of these feeds result in reproduction disorders and loss of condition of the animals.

Cattle production has been an integral part of mixed smallholder systems in Vietnam. Cattle keeping in the farms involves the integration of several components, namely livestock, aquaculture, horticulture and rice cultivation (called the VAC system which means garden (crops) – pond (fish) and stable (animals)) (Ogle & Phuc, 1997). These integrated crop-animal systems have considerable advantages compared with specialized cropping or animal production (Trach, 2003). Farmers often integrate crops and animals to maximize the income from their small land area and income per capita (Paris, 2002). Other objectives are to minimize production risks, diversify sources of income, guarantee food security, increase land productivity and improve production sustainability.

### 2.1.3 Social contribution of cattle production

In Vietnam, beef for consumption is mainly from local cattle. Cattle were previously used for working in crop plantations. Presently the draught power from these animals is being replaced by machinery. Consequently, farmers no longer raise cattle for working but for beef production.

The contributions of cattle to the farm differ between different types of farm according to the major purpose in keeping cattle. In contrast to large farms, that regarded beef production as an income generating activity, the social and cultural value of beef and food security were the major contributions of cattle on small and medium sized farms. The role of cattle as savings and maintaining the farm security by providing emergency finance (i.e. weddings, funerals) have been reported by Huyen *et al.* (2009). These cattle are used as sources of animal power, income and as a form of wealth.

### 2.1.4 Products and marketing

The meat marketing system in the Vietnamese villages is quite extensive. The meat sold for local consumption and to the meat processing factories is normally from the abattoir, where live animals are purchased directly from the villagers. Direct sale of products to consumers is uncommon due to the relatively remote locations of many livestock holdings. The animals are priced per head by individual appearance, and the size of the animal. The fresh meat is sold to the butcher at a certain price per kg carcass weight and then to the local consumers; thus, the price is double that of the original carcass. A lack of an organized livestock market infrastructure means that farmers usually deal with buyers on an individual basis. Only around 2% of producers had ever been involved in livestock supply contracts (Nin *et al.*, 2003). Good quality meat can now be bought in general markets. In terms of livestock sales, only 8% of the income of the farmer sales are from cattle; while pigs and poultry make up 60% and 23%, respectively (Nin *et al.*, 2003).

### 2.1.5 Economic importance of cattle production

According to Phung (2001) a low input system, with the advantage of available natural pasture in mountainous areas of Central Vietnam created favourable conditions for an increase in the number of cattle, resulting in an increase in farm income. Hall *et al.* (2006) concluded that for large ruminants in Vietnam, medium farms with 3 to 10 animals are the most economically efficient and most competitive in the market, provided that land is available. One of the reasons is that in smallholder farms the producer appears to use less processed feed than in large scale farms, and because of this, they are less likely to be disadvantaged by the changes in relative price (Nin *et al.*, 2003). The author explained that the feed costs are still the dominant cost component, even when household labour is valued at full cost. Smallholder farms still use mainly grazing systems with homemade concentrate, while larger farms use much more commercial feed. In contrast, some of the bigger farms, in spite of considerable government subsidy during their establishment, could not cope with the high expenditure for feed necessary because of limitations in available pasture area and the consequent shortage of feed resources, especially during the winter, leading to a stagnation of the output.

### 2.1.6 Implications for national policies and future development

As a result of the rapidly growing economy, the demand for beef is predicted to rise as incomes increase and forecasts indicate a widening gap in the supply of beef from national sources. A need was identified to raise the productivity of beef farming enterprises. Therefore the ACIAR Project, Profitable Beef Cattle Development in Vietnam mentioned earlier which was designed to assist in the profitable development of Vietnam's beef industry through conducting collaborative research in six key areas. Two of the six key areas were: (1) forage production within intensive farming systems and (2) evaluation of agro-industrial by-products as cattle feed. In a case study of livestock production in Vietnam and the effect of impact of trade liberalization, Nin *et al.* (2003) suggested that smallholder farms can exploit these opportunities in market orientation if appropriate productivity – enhancing technologies are available and are accessible.

## 2.2 Feeding of cassava foliage to cattle and its constraints

Cassava (*Manihot esculenta* Crantz), a tropical root crop widely cultivated in South East Asian countries, has a great potential as a starch source for both human and animal consumption. Cassava can survive prolonged water deficit, and is tolerant of acid soils. Cassava foliage can be used for animal feeding and a high edible biomass yield, ranging from 3.8 to 7.9 tonnes DM $ha^{-1}$, has been reported (Tung *et al.*, 2001; Van *et al.*, 2001; Hong *et al.*, 2003; Wanapat, 2003; Khang, 2004; Vongsamphanh, 2004; Thang, 2005; Wanapat, 2008). Cassava can give up to 12 tonnes of foliage DM $ha^{-1}$ if it is grown for foliage only (Dung *et al.*, 2005), depending on the variety, plant population density, harvesting height, cutting interval and rows when intercropped with legumes. The foliage from 6 to 12 month old plants contained 25-30% DM, and as dried foliage 13 to 20% CP and 16 to 20% crude fibre (CF) (Gómez, 1985). Cassava foliage, or hay made from cassava foliage, has been fed to ruminants with good results (Devendra, 1977; Ffoulkes & Preston, 1978; Wanapat *et al.*, 1997; Wanapat, 2008) and increases the ratio of protein to energy, and hence improves the productivity in ruminants (Wanapat, 2003). Many studies have focused on cassava foliage as a feed for dairy cows. In Cuba, it was suggested that the whole cassava plant could be used for dairy cows as a supplement to pasture (Garcia Lopez *et al.*, 1994; Garcia Lopez & Herrera, 1998) and in Thailand, cassava hay was used as a partial replacement of concentrate to dairy cows (Wanapat *et al.*, 2000a; Wanapat *et al.*, 2000b; Wanapat *et al.*, 2000c; Hong *et al.*, 2003; Kiyothong & Wanapat, 2004). Supplementation with malate and cassava foliage improved ruminal fermentation efficiency and increased populations of bacteria, but decreased protozoal populations (Khampa *et al.*, 2009). In Vietnam, an experiment with local beef cattle with the aim of evaluating the effects of supplementation difference levels of fresh (FCF), ensiled (ECF) or pelleted cassava foliage (PCF) on feed intake, growth, liver enzymes and thyroid hormone status in diets based on urea treated fresh rice straw (Khang & Wiktorsson, 2006). The authors concluded that FCF, with its high HCN and condensed tannin content, was slightly unpalatable (6% and 20% residue in ECF and FCF, respectively), and had a negative effect on circulating thyroid hormones at higher supplemental levels and an adverse effect on growth rate, while ECF and PCF supplementation resulted in improved growth rate without adverse effects on thyroid hormones and feed intake when fed to growing heifers.

The negative impact is caused by the HCN and the tannins in cassava foliage. When there is a high tannin content in forage the tannins are probably combined with protein to form indigestible complexes, which result in low feed intake and performance (Barry & McNabb, 1999; Man & Wiktorsson, 2001). The HCN is an anti-nutritional factor in cassava foliage when fed to animals, as reported in many prior studies (Poonam & Hahn, 1984; Dufour, 1988; Ravindran, 1993; Wanapat, 2008). In principle, cyanogen is hydrolysed by enzymes to release HCN, which can take place in the rumen by microbial activity. Then the HCN is absorbed rapidly to the blood and detoxified in the liver by the enzyme rhodanese, which converts CN to thiocyanate (SCN) which is excreted in the urine (Kumar, 1992). However, excess cyanide ions inhibit cytochrome oxidase and consequently stop ATP formation and tissues suffer energy deprivation and death quickly follows.

The processing of sun-drying cassava foliage after chopping into small pieces reduced HCN concentration. Sun-drying cassava leaves reduced HCN content by more than 90% (Gómez, 1985; Wanapat *et al.*, 1997; Wanapat, 2008). These authors found that sun-dried cassava foliage also had a low tannin content. Some studies showed that ensiling markedly reduces the cyanogen content in cassava foliage by HCN volatilization in the ensiling process (Moore & Cock, 1985; Man & Wiktorsson, 2001; Khang, 2004). Supplementation with a mixture of cassava foliage and Stylosanthes legume, which reduced HCN content, resulted in improved milk quality when fed to dairy cattle (Kiyothong & Wanapat, 2004).

## 2.3 Utilization of legume foliage as feeds for cattle

In general, the feed requirements of livestock in the humid countries of Southeast Asia were in excess of the supply (Devendra, 1984). Research has shown that better use of local feed resources is a promising strategy for reducing feed costs and will decrease the dependence on grain-based feedstuffs.

The legume trees have an enormous potential for ruminants in the tropics (Coates, 1995; Poppi & Norton, 1995) and produce large quantities of high protein leaves for animal consumption (Man *et al.*, 1995). Leguminous trees were shown to be potentially valuable sources of biomass and protein in Vietnam. Flemingia DM yields were around 3.9- 4.7 tonnes $ha^{-1}$ and to 11.2-13.4 tonnes $ha^{-1}$ in the first and second year, respectively

(Binh *et al.*, 1998; Tien *et al.*, 2003). Research on some unconventional trees/plants in Central Vietnam showed that the estimated fresh biomass production of *Hibiscus rosa sinensis L, Muntingia calabuara, Morus alba* and *Trichanthera gigantea* was 55-60, 50-60, 40-45 and 80-100 tonnes fresh matter $ha^{-1}$ $year^{-1}$, respectively (Ba & Ngoan, 2003). Yields of some leguminous shrubs and tree foliages (*Gliricidia sepium, Leucaena leucocephala, Flemingia congesta*) in the Southern part of Vietnam were reported by Man *et al.* (1995) to be 3.6-8.7 tonnes DM $ha^{-1}$ after sixteen months planting with three harvestings. According to a previous study reported by Thang (2005), the intercropping system and cutting interval of the *Phaseolus calcaratus* legume with cassava positively influenced dry biomass foliage yield and CP yield. The highest accumulated DM yield (8.8 tonnes $ha^{-1}$) and CP yield (1.98 tonnes $ha^{-1}$, on DM basis) was at one row of cassava intercropped with two rows of legume and 45 days cutting interval.

Many browse and pasture legumes species are endowed with the important attributes of high protein content, palatability and digestibility (Devendra, 1992; Coates, 1995). The message from these earlier studies is that legumes are introduced into diets because they are supposed to increase protein supply to the animal. The relative low solubility of protein fractions in some legumes usually is a further advantage in ruminant feeding as a result of high by-pass protein levels. Another advantage is the component of tannin binding proteins in tropical legumes (Poppi & Norton, 1995). The authors supposed that tropical legumes also have anti-protozoal properties which can be exploited to manipulate rumen microbial ecology. These factors enhance voluntary feed intake of ruminants offered leguminous forage and thus improve animal performance. However, there are data showing that legumes increase protein intake, but they generally do not increase intestinal protein supply per unit of DM intake (Minson, 1990; Mupangwa *et al.*, 2000). This was explained by the fact that tropical legumes are likely to lose significant amounts of protein as ammonia from the rumen due to microbial degradation. Losses of up to 43% protein in the rumen of cattle when fed the legume *Aeschynomene americana* have been found (Higgins *et al.*, 1992). This is especially the case for legumes with low digestibility, that provide insufficient energy for microbes to utilize the degraded protein. If the ratio of CP/kg of digestible OM (DOM) in a legume exceeds the approximate value of 210 g CP/kg of DOM this results in losses of protein or incomplete net transfer (Beever *et al.*, 1986; Ulyatt *et al.*, 1988; AFRC, 1992; Cruickshank *et al.*, 1992). The high rumen ammonia concentrations in the absence of a readily fermentable energy source result in energy limiting microbial growth and a significant loss of legume protein N in net transfer to the small intestine. The main role of condensed tannins (CTs) in tropical legumes has been reported as reducing ruminal degradability, but they have potential anti-nutritional effects in terms of overprotection (Poppi & McLennan, 1995). On the other hand, CTs are reported to reduce voluntary intake through astringency, an unpleasant puckering sensation in the mouth brought about by the complexing of tannins with salivary glycoproteins (Kumar & D'Mello, 1995). Some aspects of tannins in the feed in inhibiting the microbial activity or microbial enzymes resulting in reduction in ruminal turnover and rate of digestion have been recorded (Muhammed *et al.*, 1994; Salawu, 1997). The conditions also place more pressure on the utilization of legumes in supplying protein for ruminants, and this is possible if the negative impacts of anti-nutritional factors in legumes are overcome and the deficiency of a readily fermentable energy source in diet when animals are fed a sole legume feed is dealt with. Mupangwa *et al.* (2000) suggested that adding such energy sources can result in increasing ammonia utilisation and microbial protein production when feeding rations based on forage legumes.

## 2.4 Role of balanced protein - energy supply in diets for growing cattle

The growth rate and milk production of cattle fed poor quality tropical foliage, crop residues and agro-industrial by-products, are usually only about 10% of the animal's genetic potential (Leng, 1997). This may be explained by low feed intake due to poor nutritive value of the diet resulting in low digestibility, prolonged rumen retention time, bulkiness and low palatability (Aitchison *et al.*, 1986). Therefore, growing cattle require additional protein and energy to maintain an efficient rumen ecosystem that will stimulate nutrient intake and improve animal performance (Preston & Leng, 1987).

Live weight gain depends mainly on the supply of amino acids (AA) and energy-yielding substrates delivered to the tissues, up to the genetic limit for protein synthesis (Poppi & McLennan, 1995). The supply of AA depends on the protein content of the diet, its net transfer through the rumen to the intestines as microbial, undegraded and endogenous protein. The accumulation of protein depends on the efficiency of the utilization of the absorbed protein, which is dependent on readily available energy sources. Supplying energy to the rumen can be an effective way to deliver protein in the diet to the animal, either by enabling normally lost ammonia to be captured (Poppi & McLennan, 1995) or by assisting in increasing ammonia utilization and microbial protein synthesis (Mupangwa et al., 2000). On the other hand, under high temperatures and humidity, as in the tropics, there is a linear relationship between heat production and protein synthesis (MacRae & Lobley, 1986). Trying to improve growth rate by increasing protein supply might give an unexpected response, because the upper levels of heat dissipation are being approached. The related principle was highlighted by Leng (1990). Altering the protein/energy ratio of absorbed products through supplementation strategies might be one way of by-passing this limit, either through improving the efficiency of use of metabolisable energy (ME) for energy retention and so reducing heat production, or by stimulating protein synthesis without a major change in heat production. Poppi (1990) showed that the ability to alter body composition of animals at pasture will depend on obtaining high protein/energy ratios of absorbed nutrients. However, if the ratio exceeds about 210 g of CP/kg of digestible organic matter (OM) this is an evidence that losses of protein or incomplete net transfer will happen (AFRC, 1992). Such a value represents a degradable CP per available energy relationship for ruminal microbes, expressed as g of CP/MJ ME, for potential protein degradabilities.

The effects of nutrient imbalances (deficiency and excess) are well documented for protein and energy, but the implication of the balance within the extremes of deficiency and excess is a matter for conjecture. Some unexpected results may indicate a variable contribution of endogenous tissue nucleic acids (Mupangwa et al., 2000). If the energy and/or nitrogen intake was low in the diet, animals would have been mobilising tissue protein or energy to meet their requirement. The availability of these nutrients is also influenced by their interactions at specific sites of digestion and metabolism in different organs and tissues (MacRae & Lobley, 1986).

## 2.5 Evaluation of the feeding value using the *in vitro* gas production technique

For the quantitative description of digestive and metabolic processes, appropriate biological data are required and can be obtained using *in vivo*, *in situ* and *in vitro* methods. Information obtained *in vivo* is the most reliable and should be a reference when evaluating other methods, because it represents the actual animal response to a dietary treatment (López *et al.*, 2000). However, *in vivo* trials to measure digestibility are also expensive and are not ready applicable to screen large numbers of samples or not suitable for developing feed supplementation strategies using various feed constituents (Makkar, 2002). Thus, the prediction of feed digestibility or energy values from *in vitro* information has become a necessity in all feeding systems. Also, a number of *in vitro* methods have been developed to estimate the digestibility and extent of ruminal degradation of feeds and to study their variation in response to changes in rumen conditions. The *in vitro* gas method based on syringes appears to be the most suitable for use in developing countries (Menke *et al*., 1979; Blümmel *et al*., 1997). This method does not require sophisticated equipment or the use of a large number of animals (preferably two fistulated animals are required) and helps selection of feeds or feed constituents based not only on the DM digestibility but also the efficiency of microbial protein synthesis (Makkar, 2002). France *et al.* (2005) derived a general compartmental model to link the gas production technique output to animal performance.

### 2.5.1 Rate and extent of degradation

Rumen function involves complex, dynamic interactions between numerous components and processes. The amount of substrate degraded in the rumen is the result of the competition between digestion and passage. Blaxter *et al.* (1956) showed that kinetic degradation parameters and passage are integrated to estimate the actual extent of degradation of feed in the rumen. The *in vitro* gas production technique measures the rate of production of fermentation gases that can be used to predict the rate of feed degradation, assuming that the amount of gas produced reflects the amount of substrate degraded. The degradation rate of nutrients in the rumen is a key factor in predicting energy supply to the animal from a given feed, because it can have significant effects on both the ruminal microbes and the host. The fractional degradation rate can be considered an intrinsic characteristic of the feed, depending on factors such as the forage chemical composition, the proportion of different plant tissues, as affected by the stage of maturity, surface area and the cell-wall structure. On the other hand, estimates of the extent of degradation of feeds in the rumen are invariably highly sensitive to the passage rate value (Dijkstra & France, 1996). The removal of digesta from the rumen is also one of the major processes controlling the intake. Once feed enters the rumen, the degradation rate may also be affected by factors related to the animal, such as the rate of partical-size reduction by rumination and microbial activity and ruminal conditions (pH, osmotic pressure, mean retention time of digesta), which have a profound effect on microbial degradative activity.

### 2.5.2 Feed evaluation models

To associate gas production curve with digestion in the rumen, several models have been developed based on compartmental schemes and the assumption that a feed component comprises at least two fractions: a potentially degradable fraction S and an undegradable fraction U. Fraction S will be degraded at a fractional rate μ ($h^{-1}$), after a discrete lag time T (h) (López *et al.*, 2007). The most commonly used model (Ørskov & McDonald, 1979) assumes first-order kinetics, implying that substrate degraded at any time is proportional to the amount of potentially degradable matter remaining at that time, with constant fractional rate μ, and that only characteristics of the substrate limit degradation. In Germany, the Hohenheim gas test, in which the total gas volume at a fixed time point (i.e. 24h of incubation) is combined with the feed's chemical composition to predict digestibility and ME content, is widely used (Menke & Steingass, 1988). Several authors have reported good correlations between gas production at fixed incubation time points and *in vivo* OM degradation (Getachew *et al.*, 1998; Makkar, 2002; Dijkstra *et al.*, 2005). The short chain fatty acids (SCFA) production can be predicted from gas value measurements after 24h incubation (Makkar, 2002). The level of SCFA is an indicator of energy availability to the animal. If the molar proportion and amount of SCFA are known, the theoretical amounts of $CH_4$ and $CO_2$ expected from rumen fermentation can be calculated (Getachew *et al.*, 1998).

However, like any *in vitro* system of microbial activities, the partitioning of ruminally available substrate between fermentation (producing gas) and direct incorporation into microbial biomass may vary, depending on the size of the microbial inoculum and the balance of energy and nitrogen in the substrate (Pirt, 1975). A further limitation of the gas production technique is that it merely provides kinetic information on the degradation of OM or of cell contents and cell-wall material, but can not supply detailed information on the flow of aminogenic, glucogenic and lipogenic substrates out of the rumen. Moreover, the measured gas production is affected by the factors that may impact *in vitro* fermentation, such as the source and preparation of inoculum, donor animal, medium composition and preparation of the substrate (Dijkstra *et al.*, 2005). Although, it is clearly not a simulation of rumen or caecal fermentation, the *in vitro* gas production methods can be used as an estimation of the potential rate and extent of fermentation of a feed (Krishnamoorthy *et al.*, 2005) and the gas production values can be used as inputs to models to predict animal response or environment impact (France *et al.*, 2005).

# 3 Summary of materials and methods

## 3.1 Location and climate of study area

The feeding experiments (Paper I and II) were conducted at the BaVi Cattle and Forage Research Centre at Sontay, Hatay province in Northern Vietnam. The centre is located in the buffer zone between the mountainous area and the delta at E105°25 longitude and N21°06 latitude, and is 220 m above sea level. In Paper III, the growth and digestibility experiments were carried out at the YenPhu Dairy and Beef Cattle Breeding Centre in Ninh Binh Province, which is about 120 km south of Hanoi. The centre is located in the foothills of the northern MayBac mountains at E105°29 longitude and N20°14 latitude. The climate in the sites is tropical monsoon with a wet season between April and November and a dry season from December to March. The mean temperature and humidity during the experiments were 13 to 30°C and 73 to 88%, respectively. The chemical analyses and the studies of *in sacco* (Paper I), *in vivo* digestibility (Paper II) and *in vitro* gas production (Paper I, II and IV) were at the laboratory of National Institute of Animal Husbandry, Hanoi city.

*Figure 1.* Map of Northern Vietnam, location of study areas.

## 3.2 Experimental animals, feeds and management

The experimental cattle were crossbred F1 (50% local Yellow cattle and 50% Sindhi) (both *Bos indicus*) (Paper II and III) or F2 (25% Red Sindhi and 75% Holstein Friesian) (Paper I). The animals used were around six months of age and weighed on average 129 kg (SD=6) (Paper I), 114 kg (SD=12.3) (Paper II) and 126 kg (SD=16.6) (Paper III). Eleven (Paper I) or twenty eight (Paper III) female cattle were used, while twenty eight male cattle were selected for the experiments of Paper II. All the cattle in the feeding trials were kept in individual pens with roofing and concrete floor and were individually identified by numbered ear tags. Eight male cattle were penned in individual metal metabolism cages which were designed with an upper and a lower floor allowing separation of feces and urine in the *in vivo* digestibility study (Paper II). The animals had free access to clean drinking water and a mineral lick block, individually. Before the adaptation period, all animals were treated against intestinal parasites and were vaccinated against pasteurellosis and 15 days later for Foot and Mouth Disease. Three rumen fistulated cows (Red Sindhi× Holstein Friesian, 250-300 kg LW) were used for the *in sacco* rumen degradability studies (Paper I). Two of these fistulated animals were used as donors of rumen fluid in the *in vitro* gas production studies (Paper I, II and IV).

The mixture of cassava and legumes was produced from cassava and legume foliage. The cassava foliage was collected at the first harvesting time three months after planting (Paper II and III) or from five harvesting times after planting, with 45 days interval (Paper I). Cassava meal was prepared by cutting un-peeled cassava root into slices, 1-2 mm thick, sun-drying and grinding (Paper III). The stylosanthes legume (Paper II) was harvested 4 months after planting while the *Phaseolus calcaratus* legume (Paper I) was collected separately at the same time as the cassava foliage. At the harvesting time the material was cut 30 cm above ground and chopped in to 3 to 4 cm length by machine for cassava foliage or 7 to 10 cm length by hand for legumes. After cutting the foliages were sun-dried immediately for two to three days to reach a moisture content of less than 12% and were packed in separate plastic bags. Before the adaptation period the separate cassava and legumes foliages were re-sun-dried for a second time to prevent mould. The mixture ratios were either 75:25 of cassava-*Phaseolus calcaratus* foliage (CA-LE feed) (Paper I) or 50-50 of cassava-Stylosanthes foliage (Paper II) (on DM basis) or based on the level of CP and ME in the diets (Paper III).

For gas production, the samples were taken randomly in the field at 5 places of each variety and chopped into pieces of 4-5 cm in length by a small cutter. The samples were dried at 45°C in a forced air oven before grinding. All test feed samples were ground to pass through a 1 mm screen for *in vitro* incubation and chemical analysis (Paper I, II and IV).

The concentrate feed (Control feed) was mixed every fifteen days, and consisted of 40% maize meal, 35% rice bran, and 25% green bean husks (Paper I) or 40% maize meal, 40% cassava meal, and 20% green bean husks (Paper II). All the ingredients were bought in the local market.

For preparation of urea treated rice straw (URTRS), urea solution (4 kg urea plus 0.5 kg of salt dissolved in 80 L of water) was sprinkled onto 100 kg of dried rice straw that was spread out on a plastic sheet placed on the ground. The URTRS was then stored in an airtight plastic bag for three weeks before feeding (Paper I and II).

The re-growth of elephant grass (*Pennisetum purpureum*) at an age of 45 days was harvested daily in the morning. It was chopped into 10-15 cm length before feeding (Paper I and III).

## 3.3 Experimental design and treatments

For the feeding trials, the animals were allocated to a Completely Randomized Design (CRD). In Paper I the amounts of DM of concentrate were given equally while in Paper II the treatments were similar in content of nitrogen and in Paper III the level of energy and protein was varied. In Paper I, the daily rations for the animals in each treatment consisted of a basal ration of 1.47 kg chopped elephant grass, urea treated rice straw (URTRS) fed *ad libitum* (15 percent surplus), and the concentrate supplement supplied as follows: Control group was fed 2.24 kg Control feed, CA-LE40 group 1.34 kg Control feed and 0.9 kg CA-LE feed, and CA-LE60 group 0.9 kg Control feed and 1.34 kg CA-LE feed, all on DM basis. In group CA-LE40 and CA-LE60 40% and 60% of DM, respectively, of Control feed was substituted by the CA-LE feed. In Paper II, the daily rations for the cattle in each treatment consisted of a basal diet of URTRS fed *ad libitum*, 0.87 kg concentrate, 0.22 kg molasses, and one of the following supplements: the control group 0.26 kg soybean meal, the cassava (CA) group 0.95 kg cassava foliage, the stylosanthes (STY) group 1.01 kg stylosanthes foliage and the mixture (CA-STY) group 0.49 kg of stylosanthes foliage and 0.49 kg of cassava foliage all on DM basis. In Paper III, the cattle were randomly allocated to four treatments in a completely randomized 2 x 2 factorial design with seven animals in each group. The four diets consisted of two levels of CP (400 g/day and 540 g/day) and two levels of ME (25 MJ/day and 32 MJ/day). In Paper IV, the test feed samples included foliage (CaF) and root (CaM) from cassava (*Manihot esculenta* Crantz), *Trichanthera gigantea* (Tri), *Leucaena leucocephala* (Leu), *Flemingia macrophylla* (Fle), *Stylosanthes guianensis* (Sty) and *Phaseolus calcaratus* (Pha). The mixtures were done by groups, either CaF or CaM with legumes Tri, Leu, Fle, Sty or Pha. The mixture ratio of CaF or CaM with legumes were at the ratios 25:75, 50:50 or 75:25. The thirty mixed samples (2 cassava x 5 legumes x 3 mix ratio) were done with three replications on DM basis. The experimental period of feeding trials lasted for 60 days (Paper I) and 90 days (Paper II and III).

In the digestibility study, eight male cattle were randomly assigned to four treatments in a repeated Latin square design (4 × 4) (Paper II). Twenty eight female cattle were used in a CRD experiment of digestibility (Paper III). The treatments in the digestibility studies were the same as the diets in the growth trials.

## 3.4 Measurements and chemical analysis

### 3.4.1 Feeding trials

The animals were fed twice per day, in the morning (07.30 h) and afternoon (16.30 h). At each feeding occasion, the concentrate and supplemental feed was supplied first in a separate bucket, and then the roughage including either elephant grass (Paper I and III) or URTRS (Paper I, II) was offered in a feed trough. The intake of the individual feed was measured daily, based on the amount of feeds offered and refused in morning of the next day. The feeds were recorded daily for individual animals and weighed before new feed was added. The total feed intake of each animal was calculated as the sum of the intake of the feed components. The feeds offered and individual feed refusals were sampled daily and the samples were pooled for 2 weeks and analyzed. At the start and the end of the growth trial, all animals were weighed individually for two consecutive days in the morning before feeding, and the mean taken as the initial and final weight. During the experimental period, the LW was recorded every 14 days with the same procedure. Growth rate of each animal was calculated from the average daily live weight gain (LWG) at each 14 days measurement change over the time of experiment. Feed conversion ratio (FCR) was calculated as kg feed consumed $kg^{-1}$ gain, kg CP intake $kg^{-1}$ gain and MJ ME $kg^{-1}$ gain.

### 3.4.2 Digestibility studies

The faeces and urine (Paper II) or only faeces (Paper III) excreted by individual animals were collected and measured daily. During the collection time the faeces were sampled and frozen and stored for future analysis. Urine was collected in a bottle containing 800 to 1000 ml of 10% sulphuric acid to preserve nitrogen and around 1% of the total urine excreted was sampled and stored at 4°C for further analysis. The pH value of urine was tested by litmus paper to ensure that the pH value of the urine in the collection bottle was kept below 3.0. For the sampling, the feeds offered, refused and individual samples of faeces and urine were taken daily and then pooled separately for the whole collection period (Paper II) or five days collection (Paper III).

Samples of feeds, refusals and faeces were analysed for DM, ash, CP, ether extract (EE), neutral detergent fibre (NDF) and acid detergent fibre (ADF). The urine samples were analysed for DM, CP and ash. The DM (ID 930.15), CP (ID 976.05), and ash (ID 942.05) were analysed according to the standard methods of (AOAC, 1990). The EE was analysed by ISO (6492:1999) and NDF and ADF concentrations were determined according to the procedure of Van Soest *et al.* (1991). Total tannins (ID 30.018) were analysed according to (AOAC, 1975) and HCN content of the cassava foliage by the method of Ikediobi *et al.* (1980). The ME values of the feeds were calculated based on either gas production data at 24h of incubation according to the equation by Menke & Steingass (1988) (Paper I, II and IV) or on gross energy (GE). The digestibility of OM (OMD) was estimated from gas volume at 24h (Paper I and II) or measured in the *in vivo* study (Paper III).

The indoor temperature and humidity of the cattle shed were measured three times per day at 07.00 h, 14.00 h and 21.00 h to investigate the effect of the environment on feed intake and performance of the animals (Paper I, II, and III).

### 3.4.3 *In sacco* degradation

DM degradation was determined by incubating about five g of dry sample of CA-LE feed and Control feed in nylon bags in three rumen fistulated cattle according to the method of Ørskov *et al.* (1980) (Paper I). The degradation was determined at 4, 8, 12, 24 and 48 h of incubation. The mean of three results for each time were used when calculating the rate of degradation. The diet of the animals was similar to the substrates being tested. At the time of withdrawal of the bags from rumen, the sample bags were immediately placed in bucket of cold water to prevent further fermentation and to wash off the feed particles adhering to the outside of the bags. All the bags were washed under running cold water in the laboratory until the water became clear. Then the bags were dried to constant weight at 65°C before recording the weight of bags plus incubated samples. The course of degradation of the feed was described by fitting DM loss values to the exponential equation of Orskov and McDonald (1979): $P = a+b(1-e^{-ct})$. The degradation characteristics of the samples are defined as: A = washing loss (representing the soluble fraction of the feed); B = (a+b)-A (representing the insoluble but fermentable materials); c = the rate of degradation of B (Ørskov *et al.*, 1980).

### 3.4.4 *In vitro* degradation

The feed samples were incubated with rumen fluid in calibrated glass syringes as proposed by (Menke & Steingass, 1988) (Paper I, II and IV). Sample (200 mg) were weighed into 100 ml calibrated glass syringes with pistons lubricated with vaseline. Buffered mineral solution was prepared and placed in a water bath at 38.5°C under continuous flushing $CO_2$ until the blue colour turned to pink and then clear. Rumen fluid was collected after the morning feeding from two ruminally fistulated cattles fed on a diet consisting of 50% medium hay and 50% green grass at maintenance level. The rumen liquor was filtered through three layers of gauze and flushed with $CO_2$. Rumen fluid was mixed into the buffer solution at a ratio of 1:2 in a water bath at 39°C under the activities of magnetic stirrer. All handling was under continuous flushing with $CO_2$. About 30ml of buffered rumen fluid was dispensed into syringes (numbered) containing the feeds. After closing the clips on the silicon tube at the syringes tip, the syringes were gently shaken and tubes opened to remove gas by pushing the piston upwards to achieve complete removal of gas. The clip was closed, this initial volume recorded, and syringes placed vertically in incubator kept at 39°C. Three blank syringes containing 30 ml of medium only were incubated at the beginning, in the middle and at the end of the process. All the syringes were gently shaken 30 minute after the start of the incubation and four times daily later. Gas production was read at 3, 6, 12, 24, 48, and 72 hours of incubation.

To describe rate and extent of the fermentation of feed samples, the results of the gas volume reading (means of triplicates) at different times of incubation were fitted to the exponential equation $P = a + b(1 - e^{-ct})$, where P represents gas production at time t, a the intercept, (a+b) the potential gas production, c the rate of gas production. The Neway Excel program (Chen, 1997) was used for the computation. The gas produced on incubation of 200 mg DM after 24 h of incubation together with the levels of other chemical constituents were used to predicted digestibility of organic matter OMD and ME. The values of the feeds were calculated based on gas production data according to the equation by Menke and Steingass (1988):

OMD(%) = 16.49 + 0.9042GP + 0.0492CP + 0.0387Ash

ME (MJ $kg^{-1}$ DM) = 2.2 + 0.1357GP + 0.0057CP + $0.0002859CP^2$

GP is expressed in ml per 200 mg DM after 24 h of incubation, CP, ash as g $kg^{-1}$ DM.

## 3.5 Statistical analysis

All the data were analysed statistically by ANOVA using the general linear model (GLM) procedure of Minitab software version 14.0 (Minitab, 2003). The treatment least square means showing significant differences at the probability level of $P<0.05$ were compared using Tukey's pairwise comparison procedure.

For the growth trial, the statistical model was $Y_{ij} = \mu + \alpha_i + \varepsilon_{ij}$ where $Y_{ij}$ is the dependent variable, $\mu$ is the overall mean, $\alpha_i$ is effect of treatment i, and $\varepsilon_{ij}$ is a random error (Paper I and II) or $Yij = \mu + \alpha i + \beta j + (\alpha\beta)ij + \varepsilon ij$ where: Yij is the dependent variable, $\mu$ is the overall mean, $\alpha i$ is the effect of protein level i, $\beta j$ is the effect of energy level j, $(\alpha\beta)ij$ is the effect of interaction protein*energy, $\varepsilon ij$ is the random error (Paper III) or $Y_{ijk} = \mu + S_i + L_j + R_k + SL_{ij} + LR_{jk} + SR_{ik} + SLR_{ijk} + \varepsilon_{ij}$ where: $Y_{ijk}$ is the dependent variable, $\mu$ is the overall mean, $S_i$ the effect of substrate (CaF and CaM), $L_j$ the effect of mixed legume (Sty, Tri, Leu, Fle, Pha) and $R_k$ the effect of mixed ratio (25-75, 50-50, and 75-25). $SL_{ij}$, $LR_{jk}$, $SR_{ik}$, and $SLR_{ijk}$ are substrate x legume, legume x rate, substrate x rate and substrate x legume x rate interactions, respectively; $\varepsilon_{ijk}$ is the random error effect (Paper IV). Nutrient intake, LWG and FCR were tested with the initial weight, ambient temperature and humidity as covariates in the statistical model, but since the covariates were not significant ($P>0.05$) they were omitted from the final model (Paper I, II, and III).

For the digestibility study, the statistical model was $Y_{ijkl} = \mu + S_i + A_{j(i)} + P_k + T_l + \varepsilon_{ijkl}$ where $Y_{ijkl}$ is the dependent variable, $\mu$ is the overall mean, $S_i$ is the random effect of square, $A_{j(i)}$ is random effect of animal within square, $P_k$ is the fixed effect of period, $T_l$ is the fixed effect of treatment and $\varepsilon_{ijkl}$ is random residual error (Paper II).

The data obtained from the *in sacco* and the *in vitro* gas production tests were subjected to ANOVA using the GLM of Minitab software version 14.0 (Minitab, 2003).

# 4 Summary of results

## 4.1 Chemical composition of the feeds

The CP, NDF, ADF, total tannins and HCN contents of the CaF varied from 138 to 168 g $kg^{-1}$, 386 to 460 g $kg^{-1}$, 265 to 310 g $kg^{-1}$, 12 to 20 g $kg^{-1}$ and 184 to 225 mg $kg^{-1}$, on DM basis, respectively. The CaF and Tri had a low content of OM (Paper II and IV). The CP content of Sty foliage ranged from 130 g $kg^{-1}$ DM (Paper II) to 158 g $kg^{-1}$ DM (Paper IV). Total tannins of legume ranked from highest to lowest as Fle, Leu, Pha, Sty, and Tri and varied from 24 to 47 g $kg^{-1}$ DM.

The CP content of urea treated rice straw used in Paper I and II varied from 95 to 151 g $kg^{-1}$ DM. The re-growth of elephant grass (*Pennisetum purpureum*) had about the same CP content, around 88 g $kg^{-1}$ DM (Paper I and III). The CP contents of the home-made concentrate varied from 98 to 104 g $kg^{-1}$ DM (Paper I and III).

The estimated ME value in MJ $kg^{-1}$ DM ranged from 6.5 to 7.9 in CaF (Paper II and III), 7.0 to 7.6 in elephant grass (Paper I and III), 5.2 to 5.6 in URTRS (Paper II and III) and 7.3 to 8.4 in the mixture of CaF and Pha (ratio of 3:1)(Paper I and IV).

## 4.2 Feed characteristics by *in sacco* and gas production

*In sacco* degradation of feeds was studied and reported in Paper I. More than 72% of the DM and 83% of the CP of the CA-LE feed had disappeared after 48 h incubation. Compared to the Control feed, in the CA-LE feed the degradability of DM after 24 h and 48 h incubation increased by 33.8% and 25.4%, respectively.

During the first 12 hours, the increase in gas production of the CA-LE mixture was significantly higher than control feed (Paper I), while the gas volume did not differ among the sole and mixed foliages (Paper II). The gas volume produced after 24h incubation of cassava foliage ranged from 93 to 95 ml $g^{-1}$ DM (Paper II and IV), of individual legumes varied from 41 to 157 ml $g^{-1}$ DM while these value were from 58 to 197 ml $g^{-1}$ and 89 to 272 ml $g^{-1}$, on DM basis in mixed of legumes with cassava foliage and with cassava meal, respectively (Paper IV). The lowest gas volume produce was at the mixed ratio of 1:3 in cassava foliage: Fle or cassava meal: Tri, while the highest value was observed in mixture of cassava products with Pha at the rate of 3:1 recording at 24 h of incubation. The potential gas production of the mixture samples ranking from the highest to the lowest were: CaM with Sty, Leu, Pha, Fle, and Tri and for CaF with Sty, Pha, Leu, Fle and Tri. The highest estimated values of OMD, SCFA and ME were in CaM mixed with legumes at the level of 75%. There was a negative relationship between ash content and potential gas production in individual feed substrates. There was no effect of HCN in the mixture of CaM/CaF with legumes on gas production profiles (Paper IV).

The regression of DM degradability and the gas production yield at 4, 8, 12, 24 and 48 h incubation showed that there was a high correlation between *in sacco* DM lost and gas accumulation ($R^2$ = 0.9; Paper I).

## 4.3 Feed intake

With the same DMI of concentrate, increasing the CP intake by feeding a combination of cassava and Pha (CA-LE) foliages had no effect on the DM intake of URTRS (Paper I). There was, however, a significantly higher intake of URTRS in the group fed soybean meal than in the groups fed mixtures or sole foliage (Paper II). The total DMI ranged from 80 to 98 g $kg^{-1}$ $W^{0.75}$ (Paper II and III) and from 111 to 118 g $kg^{-1}$ $W^{0.75}$ (Paper I). The lowest OM and NDF intake was observed in the group offered cassava foliage as a sole feed (Paper II). When increasing the levels of CP and ME by supplying cassava products to the diet, the OM intake was significantly higher at high ME and/or CP than at low ME and CP (Paper III). There were interactions between level of protein and energy in the diets in relation to intake of DM $kg^{-1}$ $W^{0.75}$, EE $g^{-1}$ day and HCN mg $kg^{-1}$ BW and no significant differences in DMI among groups fed high ME and/or CP (Paper III).

Animals fed cassava foliage and legumes had a higher intake of tannins and HCN (Paper I, II and III). The HCN intake in the group fed a mixture of cassava and Stylosanthes was less than half the HCN intake in the group offered only cassava foliage (Paper II).

## 4.4 Digestibility

The lowest OM, NDF and ADF digestibility values was observed in the animals offered cassava foliage as a sole feed compared to the groups fed soybean, stylosanthes or mixtures of cassava foliage and legumes. The fecal N excretion was higher for cattle fed foliages whereas the highest N excretion in urine occurred in the group fed soybean meal (Paper II). The highest nutrient digestibility was in the group fed both high ME and high CP. Animals fed high energy had significantly higher digestibility of OM and GE than those fed low energy. The lowest digestibility of OM and CP values were observed in the animals fed the low ME-high CP and high ME-low CP, respectively (Paper III).

There was a significant linear regression between OM digestibility and intake of HCN and tannins intake in the group offered the low energy level (Paper III).

## 4.5 Growth performance

No animal health problems were observed among the cattle fed cassava foliage during all the experiments. The live weight gain increased with increasing CP level of substitution with combination of CA-LE (Paper I), or with mixture of cassava and stylosanthes as compared to the foliage fed as a sole feed with the same N supplement (Paper II) or with offering both high levels of CP and ME (Paper III). The LWG were 281, 551 and 609 g day$^{-1}$ in Control, CA-LE40 and CA-LE60, respectively (Paper I). Among groups fed foliages, the LWG was 337, 408 and 477 g day$^{-1}$ for the groups offered cassava, stylosanthes and the mixture of foliages, respectively (Paper II). The value of 577 g day$^{-1}$ were obtained in animals fed the diet with 32MJ ME and 540g CP (Paper III).

Increasing CP the level by including CA-LE mixture in the diet had a positive influence on feed conversion ratio (FCR) of DM and CP compared to the Control group (Paper I). The lowest efficiency of feed utilisation was observed in animals offered only cassava foliage (Paper II) or in the group fed the low ME and high CP level (Paper III).

The feed cost in CA-LE40 and CA-LE60 corresponded to 42.7% and 34.7%, respectively, of the feed cost of the Control group (Paper I).

# 5 General discussion

## 5.1 Effect of combining cassava foliage and some legume foliages

Feeding combinations of feeds could shift the site of N digestion (Axe *et al.*, 1987; Streeter *et al.*, 1989) or affect the efficiency of ruminal microbial protein synthesis (Streeter *et al.*, 1989), because the feeds differ in their AA composition and ruminal degradability (NRC, 1996). Cassava foliage is characterized by a marked deficiency of sulphur containing AA (i.e. methionine and cystine) and has a high concentration of cyanide (Rogers & Milner, 1963; Gómez, 1985; Gómez & Noma, 1986; Ravindran, 1993). The tropical legumes can supply soluble carbohydrates and fermentable N for the rumen (Smith & Van Houtert, 1987; Devendra, 1995) and provide a valuable source of protein and soluble nitrogen as well as minerals for post-rumen digestion (Smith & Van Houtert, 1987; Leng *et al.*, 1992; Devendra, 1995). By combining cassava foliage and legumes the complementing factors can be taken advantage of and give a positive associative effect. The combination of feeds might not only give a high concentration of peptides, AA and $NH_3$ to the microbes in the rumen but also ensure a balanced access of AA for the animal through by-pass protein in tannin-rich foliages. The presence of a higher concentration of peptides and AA in the rumen fluid is often shown to stimulate growth of rumen bacteria (Soto *et al.*, 1994) which can possibly change the ruminal N metabolism (Huck *et al.*, 1998). The solubility of the legume plays an important role in increasing the rumen degradation. When dietary protein degradation is rapid, the rumen microbes are unable to utilize all the peptides, the AA and $NH_3$ produced and some soluble dietary N escapes intact into the duodenum (Udén, 2000). It is clear that a markedly lower cyanide concentration was observed in the group fed a mixture of cassava foliage and legumes compared to those fed cassava foliage as sole feed (Paper II). In all the studies, the effects of supplementation with a combination of cassava foliage and legumes were positive on feed intake, digestibility and live weight gain of the animals.

### 5.1.1 Effect on feed intake

The results in the present studies showed that there was no significant effect on URTRS intake or total DM intake with increasing CP level when supplementing a mixture of cassava and Pha foliage (Paper I) or among the groups fed cassava and Sty foliage as a mixture or sole supplemental feed when offered at the same level of N (Paper II). In contrast, Khang & Wiktorsson (2006) showed that the use of cassava foliage, which contains tannins, resulted in significantly lower feed intake from fresh URTRS with increasing levels of cassava foliage in the diet. The intakes of tannins were significantly higher in the animals fed the mixture of cassava and legume foliage as compared to the sole feed (Paper II) or increased with increasing of level of CA-LE feeding (Paper I). The tannins tend to affect the nutritive value of ruminant feeds by reducing the voluntary feed intake (Barry & McNabb, 1999). At high levels of tannins, (50 to 90 g/kg DM) the digestibility of the fibre in the rumen is reduced (Reed *et al.*, 1985) through inhibition of the activity of bacteria and anaerobic fungi (Chesson *et al.*, 1982). There are several possible explanations for the differences in results. First, the tannin intake was low, ranging from 17 to 33 g $kg^{-1}$ DM (Paper I and II) in the animals fed diets containing mixtures of cassava and legume. Secondly, only a half of the amount of HCN was consumed by the animals fed a mixture of cassava and legume compared to the animals fed only cassava foliage. Thirdly, it is probable that tannins are interacting with HCN. Generally, anti-nutritive factors do not exist in isolation (Makkar, 2003). The simultaneous presence of tannins and HCN might alleviate the adverse effects of each other (Goldstein & Spencer, 1985), and a process of inhibiting the enzyme release of HCN by tannins could occur. In certain ecological situations, tannins may interfere with the release of HCN from the hydrolysis of cyanogenic glycosides, thus reducing their toxic effects (Makkar, 2003). This may have resulted in that the negative effects of HCN and tannins on the total DM intake could not be observed in the present studies. Similar results were reported by Kiyothong & Wanapat (2003), who concluded that supplementation of cassava hay in combination with Stylo hay did not increase total DM and forage intake.

### 5.1.2 Effect on nutrient digestibility

The digestibility of the dietary components depends on several factors, such as fibre source, the ingredients used in the feed and their preparation, and the digestive capacity of the animals. One concern when feeding only cassava foliage to cattle, besides the content of HCN, is the high ash content, which has been found to reduce nutrient digestibility. A diet with low OM will result in a lower NDF digestibility, leading to an unbalanced nitrogen/energy ratio and more nitrogen excreted in the faeces (Paper II). On the other hand, the significantly higher apparent OM and CP digestibility in the groups fed stylosanthes foliage or soybean meal compared to those fed only cassava foliage indicates that there was probably an adverse effect of HCN in our study. Feeding a combination of cassava foliage and stylosanthes resulted in slightly higher nutrient digestibility and N retention than when feeding the foliages as a sole feed, but the difference was not significant. The concentration of HCN was lower in the diets with a mixture of cassava and legume feed, but the tannin concentration in the mixed diet was high when compared to the diets with foliages as the sole feed (Paper II). Feeding tannin-rich combined foliages limited ruminal fermentation due to adverse effects on ruminal fibre degradation (Reed, 1995; Theodorou *et al.*, 2000). This was mainly the result of a reduced digestibility of the ADF (cellulose and lignin) fraction of the fibre (Fässler & Lascano, 1995; Tiemann *et al.*, 2008). In contrast, no significant differences of fibre (NDF and ADF) digestibility were observed in animals fed cassava foliage, Stylosanthes and their mixture (Paper II). It can be suggested that the increase in tannin concentration caused by a combination of foliages did not affect the fibre digestibility in the diet. Min *et al.* (2003) proposed that depending on the type and concentration, tannins could be either advantageous or disadvantageous in terms of digestibility and metabolisable protein supply to the animal.

### 5.1.3 Effect on growth rate

Although, in theory, N-rich legume-cassava foliage could enhance protein metabolism in the rumen, the protein degradation in the rumen was affected by the tannin content in the combination of foliages. The effects of tannin were reflected in lower ruminal ammonia concentration (Carulla *et al.*, 2005; Tiemann *et al.*, 2008). It is well documented that tannins reduce ruminal protein degradation by precipitation of proteins, making them less degradable by ruminal microbes (Reed, 1995; Mueller-Harvey, 2006). The N degradation in the rumen was not investigated in this study but when supplementing tanniferous foliages the flux of dietary protein to the abomasum is increasing and the apparent absorption of essential AA in the intestine may increase (Ben Salem *et al.*, 2005; Ramírez-Restrepo & Barry, 2005; Hess *et al.*, 2008). In this context, more balanced AA are probably absorbed from the intestines of the cattle when fed the combination of cassava and legume foliage, and this could have improved animal performance. With the same N supplement, 30% and 19%, respectively, there was a higher LWG in the group fed a mixture of foliages as compared to the groups fed only cassava or stylosanthes (Paper II). Increasing the feeding level of the mixture tended to increase the growth rate of the animals. By replacing part of the Control feed with the CA-LE feed, the daily total CP intake increased by 100 g and 130 g in CA-LE40 and CA-LE60, respectively, which resulted in a significant increase in LWG (Paper I). The positive associative effect of feeding a mixture could be partly due to the complementation of AA deficient in cassava foliage. If a certain amount of the protein which by-passes ruminal degradation is available for digestion and absorption, such mixtures could have a protein-sparing effect and increase the supply of metabolisable protein to the animal (Hess *et al.*, 2008). Feeding tannin-rich foliages would be possible when a high ruminal ammonia concentration is maintained by utilisation the soluble N from urea in the diet. This is consistent with urea supplementation of diets to cattle fed tannin-rich foliages, as suggested by Norton (2000). The URTRS used in the experiments in this thesis as basal diets was made by ensiling rice straw with 4% of urea (Paper I and II). The moderate concentration of tannins in the mixtures probably affected the protein precipitation properties of the diet in a positive way (Terrill *et al.*, 1992). However, if the effect of a rapid degradable energy source (like starch) in the rumen in combination with the mixture of foliages was to enhance the efficiency of nutrient utilization, adding starch could improve growth rate in cattle fed foliages and a basal diet of URTRS. A detailed discussion of the effect of balanced CP/ME on feed utilisation is addressed in the following section. The balance between digestible protein (DP) and digestible energy (DE) is a key factor (Preston & Leng, 1987). Decreased dietary DP/DE ratio could increase protein conservation. The essential AA in this case are not catabolized for energy, but are conserved in the body. Higher LWG of animals were obtained when they were given both high levels of protein and energy in the diet with cassava products (Paper III).

## 5.2 Effect of a balanced protein and energy ratio supply in diets when feeding cassava products

The primary target of the ruminant nutritionist is to achieve maximum output of protein in products such as milk, meat and wool with a minimum of dietary CP input. Increasing the efficiency of protein N utilisation by livestock, leading to lower N excretion, is becoming an environmental imperative in many countries (Castillo *et al.*, 2001). In theory, efficient use of N in the diet of ruminants depends on the supply of N to the rumen, related to amounts and appropriate forms, and the efficiency of protein deposition in the animal's tissues. Factors such as restricted ME intake or mineral or vitamin deficiencies lead to sub-maximal protein deposition in the animal, and N requirements are reduced accordingly (Oldham *et al.*, 1977; Nolan & Dobos, 2005). Cattle, when grazing in a tropical situation, require additional protein and energy to maintain an efficient rumen ecosystem that stimulates nutrient intake and improves animal performance (Preston & Leng, 1987).

Protein sources from the mixture of tannin-rich foliages have the ability to by-pass ruminal degradation, as discussed above. But the question is whether all the proteins protected from ruminal degradation were digested and absorbed in the small intestine. In fact, as compared to the control group fed soybean meal, with the same N offered, there were still much higher faecal N losses in animals fed the mixture of cassava and legume foliage as compared to those fed soybean meal (Paper II). The issue of faecal losses and how to increase N absorption in the lower gut need to be further looked into. The amount of N absorbed depends on other factors, such as energy intake from diet (Poppi & McLennan, 1995). Therefore, in the context of our studies, the effects of supply of both protein and energy using cassava products as a nutrient combination on the feed intake, growth and feed utilisation were addressed (Paper III). Higher dietary energy values can be obtained for cassava foliage by taking advantage of the positive effects that occur when cassava foliage is fed in combination with cassava meal. A significantly higher gas volume produced *in vitro*, indicating higher degradability, was observed when a mixture of cassava root meal and legumes was incubated (Paper IV). Feeding a diet with high energy containing cassava foliage (32 MJ day$^{-1}$) resulted in an improvement of apparent digestibility coefficients of OM, GE, NDF and ADF as compared to the low level (25 MJ day$^{-1}$) (Paper III). Increasing LWG of cattle, with increasing level of CP and ME in the diet, was achieved when using cassava products and low quality grasses (Paper III). The daily gain, feed efficiency and protein utilisation were not improved when the animals were fed only ensiled cassava foliage (Khang & Wiktorsson, 2006), or low level of energy (Paper III). According to Moore & Cock (1985) the addition of dried cassava chips as an energy source to an ensiled cassava foliage diet improved growth rate by 59% and feed efficiency by 34% compared to a diet containing only ensiled cassava foliage, while the addition of cottonseed meal to the diet did not further improve animal performance. The authors suggested that energy was more limiting than protein in the diet with only cassava foliage. Although cassava foliage is known to have the potential to increase intestinal protein supply (Wanapat, 2008), increasing the levels of cassava in the diet is limited by protein loss in the rumen degradation, resulting in reduced intestinal protein supply per unit of DM intake. The supply of an energy source such as cassava meal (starch) in a diet of cassava foliage overcomes this limitation (Paper III). The supplementation of energy with protein to ensure an appropriate protein to energy (P/E) ratio in the nutrients absorbed is important for optimum efficiency of feed utilisation (Leng *et al.*, 1992).

A beneficial effect of extra energy supply in diets containing cassava foliage, besides improved digestibility and growth rate, on the detoxification may be possible. An interaction of energy level with anti-nutritive factors (i.e. HCN) on digestibility was also found in our study. With a low dietary energy supply, the higher HCN and tannin intake achieved by increasing cassava foliage supply in the diet might be one of the reasons for the lower OM digestibility (Paper III). It is questionable whether there is an effect of energy level in the diet on metabolic detoxification in cattle or not. But it is obvious that the ingestion of HCN resulted in no negative effect on digestibility and growth rate in the group fed high energy (Paper III). When feeding a sub-lethal dose of HCN, the detoxification process can occur via several pathways, but the most important is the reaction of cyanide with thiosulphate to form thiocyanate and sulphite. The thiocyanate inhibits the intra-thyroidal uptake of iodine, leading to an increase in secretion of thyroid stimulating hormone, and causes a reduction in thyroxine level, which is necessary for growth (Tewe, 1992). The use of vegetable oil improved the growth rate of local Yellow cattle when supplementing cassava foliage to a basal diet of rice straw (Seng, 2001). Fomunyam *et al.* (1980) also reported that the toxic effect of HCN in cassava was greatly reduced in the presence of palm oil. To supply vegetable oil or palm oil would increase the dietary energy. These supplements delay the breakdown, and therefore prevent the absorption of the cyanogenic glucosides (Fomunyam *et al.*, 1980; Tewe, 1992). If both the level of protein and energy are increased (simultaneously) the diet could play a role in HCN detoxification. The improved protein quality in the diet could be used to overcome HCN toxicity. It has been shown that high quality protein provides methionine and cystine (from lysine) to detoxify HCN to a harmless compound – ammonium formate (Oke, 1979; Osuntokun, 1981).

## 5.3 Characteristics of rumen degradation and fermentation kinetics of cassava products and some legume foliages

To evaluate feedstuffs or diets of ruminants, the feed or diet should ideally be fed to the ruminant of interest and production responses determined. Testing all possible feeds in all possible situations *in vivo* would clearly provide the most accurate ranking of feedstuffs in terms of nutritive quality, but it is neither practicable nor cost effective (Dijkstra *et al.*, 2005). The potential gas production of the mixture samples, indicating the level of solubility and degradability of substrate in rumen, ranking from the highest to the lowest, was: CaM with Sty, Leu, Pha, Fle, and Tri and for CaF with Sty, Pha, Leu, Fle and Tri (Paper IV). Among these mixtures of CaF with legumes, the results showed that both the legumes, Sty and Pha, had a tendency to increase gas volume produced in the first 12 h of incubation at all mixture ratios. This is consistent with early results of an increase in gas production of the mixture of CaF and Pha (ratio of 3:1) and significantly higher than the control feed (Paper I). Although legumes are characterised by high degradation in rumen, there is a difference in potential gas production which might depend on their chemical composition (Getachew *et al.*, 2004) and concentration of anti-nutrient factors in the foliages (Getachew *et al.*, 1998; Silanikove *et al.*, 2001; Makkar, 2002). The lower gas volume of mixtures measured at 24h is probably due to the high ash content in Tri or high tannin in Fle and Leu, which was found in Paper IV. The kinetics of gas production is dependent on the relative proportions of soluble to insoluble but degraded substrates (Getachew *et al.*, 1998). The high NDF content of Sty resulted in the highest gas volume when mixed with cassava products. Deaville & Givens (2001) reported that NDF degradability possibly had an effect on gas volume produced. The previous *in sacco* study showed that 73% of DM and 83% of CP in the mixture of CaF and Pha had disappeared in the rumen after 48 h of incubation (Paper I). Another study in tropical Africa showed that the DM degradability of Leucaena and Gliricidia was around 68% after 48 h incubation (Smith *et al.*, 1989). The combination of CaF with either Sty or Pha improved the characteristic of ruminal degradation compared to the other legumes. The positive interaction of cassava and legume foliage seems to be due to the structure, the high CP content and the fragility of cell walls, which resulted in a high degradation rate during the first time period of incubation in the rumen. However, the gas volume did not differ among the CaF, Sty and the mixed foliages (Paper II). It may be that there were other factors affecting the gas production characteristics in the substrate of CaF and Sty. Rooke & Armstrong (2007) suggested that the supply of both energy and nitrogen to the rumen is  important to improve the efficiency of rumen fermentation. This supports the results in this study. Higher gas volume produced occurred in the mixtures of legumes with CaM compared to those with CaF (Paper IV). The available source of energy and nitrogen lead to  differences in nutrient availability and could have resulted in increased microbial growth at the beginning of incubation, which resulted in more fermentation and increased gas production (Calabrò *et al.*, 2005). According to the results of Sutton (1985) and Krishnamoorthy *et al.* (1991) energy supplements differ in their rate and extent of ruminal fermentation. It was suggested that one needs sufficient data on rates of protein degradation and carbohydrate fermentation by rumen microbes in ruminant feed formulations (Williams, 2000).

The importance of methodological considerations should therefore be assessed by addressing the crucial question of the objective of the *in vitro* gas production. It is clearly not a simulation of rumen fermentation, but perhaps it can be used as an estimation of the potential rate and extent of fermentation of a feed (Krishnamoorthy *et al.*, 2005). These data could then be used as inputs to other models to predict animal response. The database of our study is not powerful enough to be used to simulate a model, but there were consistent results in the feeding trials and the data obtained from *in vitro* gas production. The high gas volume produced by the combination of CaF and Pha/Sty, coincided with the improved growth rate of animals fed the diet with these mixtures (Paper I, II, IV).

# 6 General conclusions and implications

## 6.1 Conclusions

- Replacing a conventional concentrate with a mixture of cassava and *Phaseolus calcaratus* legume as a protein source in the ration for growing heifers resulted in improved rumen environment, increased growth rate, reduced feed cost per kg live weight gain, and gave a higher economic return.
- Feeding stylosanthes and a mixture of cassava and stylosanthes foliage improved dry matter intake, digestibility and live weight gain of animals fed a basal diet of urea treated rice straw. The low organic matter and high level of hydrogen cyanide in the diet when feeding only cassava foliage explains the negative effect on intake, neutral detergent fibre digestibility and nitrogen retention, and resulted in lower growth rates.
- Increasing the level of crude protein and metabolisable energy in the diet using cassava products improved the digestibility and growth rate of cattle fed low quality grasses. Supplying extra energy in the diet of cattle fed a high amount of cassava foliage might be necessary to overcome the negative effect of hydrogen cyanide.
- Increasing the level of cassava root meal improved the degradation and fermentation of mixtures with legumes. There was a negative relationship between ash content and potential gas production in individual feed substrates. No effect of hydrogen cyanide on fermentation kinetics in rumen could be detected in this study.

## 6.2 Implications and further research

Ruminants, particularly cattle, play an important role as assets and source of income for smallholders in Vietnam, where livestock systems are frequently based on roughages which are deficient in crude protein and high in structural carbohydrates. Farmers often cultivate the cassava crop and legumes in their field as an integral part of a mixed smallholder system. Planting cassava in mono-culture leads, after several years, to erosion of the soil and the fertility of the soil is also affected negatively. Legumes are known to have a good regeneration ability and improve the soil fertility by root nitrogen fixation. Cropping of both cassava and legumes together improves the benefits for the farmers in terms of crop yields and stabilized soil fertility, and thus is more sustainable. They are also an available feed resource for ruminants, resulting in increased net income of the farmers. If possible, cassava and legume foliage should be processed and sun-dried to ensure high amounts of edible biomass, but also low levels of HCN. Cattle should be fed the cassava and legume foliage as a combined feed, and the ratios should be 50:50 or 25:75 cassava:legume foliage. Further research is needed to explore the potential of cassava foliage and tropical legumes as feeds for cattle. Alternative feeding strategies which involve HCN-rich cassava foliage in combination with legumes free of cyanide and with high protein content could have the potential of diluting cyanide content while still retaining the benefits of cassava foliage, e.g. high edible biomass and N content, on the feed efficiency and metabolisable protein supply.

In practice, feeding a certain amount of cassava foliage improved DMI, digestibility and LWG of animals fed a basal diet of urea treated rice straw, but more benefit will be obtained if cassava root meal is offered simultaneously, since probably more N was absorbed from the intestine and the growth rate of the cattle was further improved. The effects of feeding a combination of foliages (CA-LE) together with a highly fermentable energy source such as cassava meal should be investigated.

From an environmental point of view, feeding tanniferous foliage from a combination of cassava and legumes could be of interest for research into strategies for reducing methane emissions from cattle. A mixture as a supplemental feed could be prepared to balance the nutrient absorption and to decrease ruminal degradation as a result of inhibition of methane emission.

# References


AFRC (1992). Technical Committee on Responses to Nutrients. Report No. 9. Nutritive requirements of ruminant animal. *Nutrition Abstracts Reviews, series B* 62(12), 787-835.

Aitchison, E.M., Gill, M., Dhanoa, M.S. & Osbourn, D.F. (1986). The effect of digestibility and forage species on the removal of digesta from the rumen and the voluntary intake of hay by sheep. *British Journal of Nutrition* 56(02), 463-476.

AOAC (1975). *Official Methods of Analysis*. Washington, DC: 12th ed. Association of Analytical Chemists.

AOAC (1990). *Official Methods of Analysis*. Washington, DC: 15th ed. Association of Analytical Chemists.

Axe, D.E., Bolsen, K.K., Harmon, D.L., Lee, R.W., Milliken, G.A. & Avery, T.B. (1987). Effect of wheat and high-moisture sorghum grain fed singly and in combination on ruminal fermentation, solid and liquid flow, site and extent of digestion and feeding performance of cattle. *Journal of Animal Science* 64, 897-906.

Ba, N.X. & Ngoan, L.D. (2003). Evaluation of some unconventional tree/plants as ruminant feeds In: Preston, T.R. & Ogle, B. (Eds.) *Proceedings of Final National Seminar-Workshop on Sustainable Livestock Production on Local Feed Resources, 25 - 28 March*. HUAF-SAREC, Hue City, Vietnam. pp. 21-26.

Barry, T.T.N. & McNabb, W.W.C. (1999). The implications of condensed tannins on the nutritive value of temperate forages fed to ruminants. *British Journal of Nutrition* 81(4), 263-272.

Beever, D.E., Dhanoa, M.S., Losada, H.R., Evans, R.T., Cammell, S.B. & France, J. (1986). The effect of forage species and stage of harvest on the processes of digestion occurring in the rumen of cattle. *British Journal of Nutrition* 56(02), 439-454.

Ben Salem, H., Makkar, H.P.S., Nefzaoui, A., Hassayoun, L. & Abidi, S. (2005). Benefit from the association of small amounts of tannin-rich shrub foliage (*Acacia cyanophylla* Lindl.) with soya bean meal given as supplements to Barbarine sheep fed on oaten hay. *Animal Feed Science and Technology* 122(1-2), 173-186.

Binh, D.V., Tien, N.P. & Mui, N.T. (1998). Study on biomass yield and quality of *Flemingia macrophylla* and on the performance and on soil fertility. In: *Proceedings of workshop of Animal Nutrition Science*. Hanoi: Ministry of Agriculture and Rural Development, Hanoi, Vietnam.

Blaxter, K.L., Graham, N.M. & Wainman, F.W. (1956). Some observations on the digestibility of food by sheep, and on related problems. *British Journal of Nutrition* 10(02), 69-91.

Blümmel, M., Makkar, H.P.S. & Becker, K. (1997). *In vitro* gas production: a technique revisited. *Journal of Animal Physiology and Animal Nutrition* 77, 24-34.

Burns, B.M., Binh, D.V. & Su, V.V. (2002). Beef Cattle Genetic and Breeding Projects in Vietnam and the Future Direction. In: Jack Allen & Ancharlie Na-Chiangmai (Eds.) *Development Strategies for Genetic Evaluation for Beef Production in Developing Countries, ACIAR Proceedings 108*. Khon Kaen Province, Thailand, July 23–28 2001: Australian Centre for International Agricultural Research (ACIAR). pp. 148-153.

Calabrò, S., Cutrignelli, M.I., Piccolo, G., Bovera, F., Zicarelli, F., Gazaneo, M.P. & Infascelli, F. (2005). *In vitro* fermentation kinetics of fresh and dried silage. *Animal Feed Science and Technology* 123-124(Part 1), 129-137.

Carulla, J.E., Kreuzer, M., Machmuller, A. & Hess, H.D. (2005). Supplementation of *Acacia mearnsii* tannins decreases methanogenesis and urinary nitrogen in forage-fed sheep. *Australian Journal of Agricultural Research* 56(9), 961-970.

Castillo, A.R., Kebreab, E., Beever, D.E., Barbi, J.H., Sutton, J.D., Kirbi, H.C. & France, J. (2001). The effect of energy supplementation on nitrogen utilisation in lactating dairy cows fed grass silage diets. *Journal of Animal Science* 79, 240-246.

Chen, X.B. (1997). Neway Excel–A utility for processing data of feed degradability and in vitro gas production. *XBC laboratory. Rowett Research Institute. Aberdeen, AB2 9SB. UK.*

Chesson, A., Stewart, C.S. & Wallace, R.J. (1982). Influence of plant phenolic acids on growth and cellulolytic activity of rumen bacteria. *Applied and Environmental Microbiology* 44(3), 597-603.

Coates, D.B. (1995). Tropical legumes for large ruminants. In: D'Mello, J.P.F. & Devendra, C. (Eds.) *Tropical legumes in animal nutrition* CAB International. ISBN 0 85198 926 8, pp. 191-231.

Cruickshank, G.J., Poppi, D.P. & Sykes, A.R. (1992). The intake, digestion and protein degradation of grazed herbage by early-weaned lambs. *British Journal of Nutrition* 68(02), 349-364.

Cuong, L.X. (1993). *Dairy cattle production in Ho Chi Minh city. IAS-IDRC survey report 1993* [online]

Deaville, E.R. & Givens, D.I. (2001). Use of the automated gas production technique to determine the fermentation kinetics of carbohydrate fractions in maize silage. *Animal Feed Science and Technology* 93(3-4), 205-215.

Devendra, C. (1977). Cassava as a feed source for ruminants. In: Nestel, B. & Graham, M. (Eds.) *Proceedings of Workshop on Cassava as Animal Feed*. University of Guelph: International Development Research Centre (IDRC). pp. 107-119.

Devendra, C. (1984). Prospects for processing and treatments of crop residues and agro-industrial by-products for ruminants in the Asian region. In: Hutagalung, R.I., Hew, V.F., Devendra, C. & Vijuchulata, P. (Eds.) *Proceedings of conference on Feeds and feeding systems for livestock, 8th Annual conference of the Malaysian Society of Animal Production*. Malaysia. pp. 25-36.

Devendra, C. (1992). Nutritional potential of fodder trees and shrubs as protein sources in ruminant nutrition. In: Speedy, A., Pugliese, P.L. (Ed.) *Legume trees and other fodder trees as protein sources for livestock*. 100 Rome, Italy: FAO of the United Nations, 1992; 102). ISBN 92-5-103203-3, pp. 95-113.

Devendra, C. (1995). Composition and nutritive value of browse legumes. In: D'Mello, J.P.F. & Devendra, C. (Eds.) *Tropical legumes in animal nutrition* CAB International. ISBN 0 85198 926 8, pp. 49-66.

Dijkstra, J. & France, J. (1996). A comparative evaluation of models of whole rumen function. *Annales de Zootechnie* 45 (Suppl.), 175-192.

Dijkstra, J., Kebreab, E., Bannink, A., France, J. & López, S. (2005). Application of the gas production technique to feed evaluation systems for ruminants. *Animal Feed Science and Technology* 123-124(Part 1), 561-578.

Dufour, D. (1988). Cyanide content of cassava ( *Manihot esculenta* , Euphorbiaceae) cultivars used by Tukanoan Indians in Northwest Amazonia. *Economic Botany* 42(2), 255-266.

Dung, N.T., Ledin, I. & Mui, N.T. (2005). Intercropping cassava (*Manihot esculenta* Crantz) with Flemingia (*Flemingia macrophylla*); effect on biomass yield and soil fertility. *Livestock Research for Rural Development* 17(1), 1-13.

FAOSTAT data (2008). FAO Statistical Databases. *Food and Agriculture Organization of the United Nations* [online]Available from: http://faostat.fao.org/default.aspx.

Fässler, O.M. & Lascano, C.E. (1995). The effects of mixtures of sun-dried tropical shrub legumes on intake and nitrogen balance by sheep. *Tropical Grasslands* 29(2), 92-96.

Ffoulkes, D. & Preston, T.R. (1978). Cassava or sweet potato forage as combined sources of protein and roughage in molasses based diets: effect of supplementation with soybean meal. *Tropical Animal Production* 3(3), 186-192.

Fomunyam, R.T., Adegbola, A.A. & Oke, O.L. (1980). The role of palm oil in cassava-based rations. In: Terry, E.R., Oduro, K.A. & Caveness, F. (Eds.) *Proceedings of Tropical root crops, research strategies for 1980's.* Ottawa, Canada: International Development Research Centre, IDRC. pp. 152-153.

France, J., Lopez, S., Kebreab, E., Bannink, A., Dhanoa, M.S. & Dijkstra, J. (2005). A general compartmental model for interpreting gas production profiles. *Animal Feed Science and Technology* 123-124(Part 1), 473-485.

Garcia Lopez, R. & Herrera, J. (1998). Milk production from pastures and cassava (*Manihot esculenta*) or sweet potato (*Ipomea batata*) integral forage plant supplementation. *Cuban Journal of Agricultural Science* 32, 29-31.

Garcia Lopez, R., Mejias, R. & Herrera, J. (1994). Preliminary results of the combination of cassava and sugar cane foliar area as a supplement for dairy cows under grazing conditions. *Cuban Journal of Agricultural Science* 28, 41-43.

Getachew, G., Blümmel, M., Makkar, H.P.S. & Becker, K. (1998). *In vitro* gas measuring techniques for assessment of nutritional quality of feeds: a review. *Animal Feed Science and Technology* 72(3-4), 261-281.

Getachew, G., Robinson, P.H., DePeters, E.J. & Taylor, S.J. (2004). Relationships between chemical composition, dry matter degradation and *in vitro* gas production of several ruminant feeds. *Animal Feed Science and Technology* 111(1-4), 57-71.

Goldstein, W.S. & Spencer, K.C. (1985). Inhibition of cyanogenesis by tannins. *Journal of Chemical Ecology* 11(7), 847-858.

Gómez, G. (1985). Cassava foliage: chemical composition, cyanide content and effect of drying on cyanide elimination. *Journal of the Science of Food and Agriculture* 36(6), 433-441.

Gómez, G. & Noma, A.T. (1986). The amino acid composition of cassava leaves, foliage, root tissue and whole-root chips. *Nutrition Report International* 33, 595-601.

GSO (2009). Statistical Yearbook of Vietnam 2008. In: General Statistics Office (Ed.). Hanoi: Statistical Publishing House.

Hall, D.C., Thao, T.M., Minh, D.V. & Lien, L.V. (2006). *Competitiveness of the livestock sector in Vietnam. A study conducted by the FAO of the United Nations for the World Bank*: The World Bank, March 2006.

Hess, H.D., Mera, M.L., Tiemann, T.T., Lascano, C.E. & Kreuzer, M. (2008). *In vitro* assessment of the suitability of replacing the low-tannin legume *Vigna unguiculata* with the tanniniferous legumes *Leucaena leucocephala*, *Flemingia macrophylla* or *Calliandra calothyrsus* in a tropical grass diet. *Animal Feed Science and Technology* 147(1-3), 105-115.

Higgins, S.J., Tabrett, S.J., Poppi, D.P. & Norton, B.W. (1992). Duodenal protein supply in cattle grazing signal grass (*Brachiaria decumbens*) and glenn joint vetch (*Aeschynomene americana*). In: *Proceedings of the Australian Society of Animal Production* p. 52.

Hong, N.T.T., Wanapat, M., Wachirapakorn, C., Pakdee, P. & Rowlinson, P. (2003). Effects of timing of initial cutting and subsequent cutting on yields and chemical compositions of cassava hay and its supplementation on lactating dairy cows. *Asian-Australasian Journal of Animal Sciences* 16(12), 1763-1769.

Huck, G.L., Kreikemeier, K.K., Kuhl, G.L., Eck, T.P. & Bolsen, K.K. (1998). Effects of feeding combinations of steam-flaked grain sorghum and steam-flaked, high-moisture, or dry-rolled corn on growth performance and carcass characteristics in feedlot cattle. *Journal of Animal Science* 76(12), 2984-2990.

Huyen, L.T.T., Herold, P. & Valle Zárate, A. (2009). Farm types for beef production and their economic success in a mountainous province of northern Vietnam. *Agricultural Systems* 103(3), 137-145.

Ikediobi, C.O., Onylia, G.O.C. & Eluwah, C.E. (1980). A rapid and inexpensive enzymatic assay for total cyanide in cassava (*Manihot esculenta Crantz*) and cassava products. *Agricultural and Biological Chemistry* 44(12), 2803-2809.

Khampa, S., Chumpawadee, S. & Wanapat, M. (2009). Supplementation of malate level and cassava hay in high-quality feed block on ruminal fermentation efficiency and digestibility of nutrients in lactating dairy cows. *Pakistan Journal of Nutrition* 8(4), 441-446.

Khang, D.N. (2004). *Better use of local forages for dairy cattle in Vietnam: Improving grasses, rice straw and protein rich forages.* Diss.:Swedish

University of Agricultural Science, Uppsala, Sweden. ISBN 91-576-6752-7.

Khang, D.N. & Wiktorsson, H. (2000). Effect of cassava leaf meal on the rumen environment of local yellow cattle fed urea-treated paddy straw. *Asian-Australian Journal of Animal Science* 13(8), 1102-1108.

Khang, D.N. & Wiktorsson, H. (2006). Performance of growing heifers fed urea treated fresh rice straw supplemented with fresh, ensiled or pelleted cassava foliage. *Livestock Science* 102(1-2), 130-139.

Khieu Borin, Lindberg, J.E. & Ogle, R.B. (2005). Effect of variety and preservation method of cassava leaves on diet digestibility by indigenous and improved pigs. *Animal Science* 80(3), 319-324.

Kiyothong, K. & Wanapat, M. (2003). Cassava hay and stylo 184 hay to replace concentrates in diets for lactating dairy cows. *Livestock Research for Rural Development* 15(11), 1-8.

Kiyothong, K. & Wanapat, M. (2004). Supplementation of cassava hay and stylo 184 hay to replace concentrate for lactating dairy cows. *Asian-Australasian Journal of Animal Sciences* 17(5), 670-677.

Krishnamoorthy, U., Rymer, C. & Robinson, P.H. (2005). The *in vitro* gas production technique: Limitations and opportunities. *Animal Feed Science and Technology* 123-124(Part 1), 1-7.

Krishnamoorthy, U., Soller, H., Steingass, H. & Menke, K.H. (1991). A comparative study on rumen fermentation of energy supplements in vitro. *Journal of Animal Physiology and Animal Nutrition* 65, 28-35.

Kumar, R. (1992). Anti-nutritional factors, the potential risks of toxicity and methods to alleviate them. In: Speedy, A. & Pugliese, P. (Eds.) *Proceedings of Legume Trees and Other Fodder Trees as Protein Source for Livestock, 14-18 October.* Kuala Lumpur, Malaysia. : FAO, Rome. pp. 145-160.

Kumar, R. & D'Mello, J.P.F. (1995). Anti-nutritional factors in forage legumes. In: D'Mello, J.P.F. & Devendra, C. (Eds.) *Tropical legumes in animal nutrition* CAB International. ISBN 0 85198 926 8, pp. 95-135.

Leng, R.A. (1990). Factors affecting the utilization of 'poor-quality' forages by ruminants particularly under tropical conditions. *Nutrition Research Reviews* 3(01), 277-303.

Leng, R.A. (1997). *Tree foliage in ruminant nutrition*: FAO Animal Production and Health Papers 139, FAO, Rome, Italy ISBN 9251040869.

Leng, R.A., Choo, B.S. & Arreaza, C. (1992). Practical technologies to optimise feed utilisation by ruminants. In: Speedy, A. & Pugliese, P. (Eds.) *Proceedings of Legume trees and other fodder trees as protein sources for livestock*. Kuala Lumpur, Malaysia. : FAO, Rome. pp. 75-93.

López, S., Dhanoa, M.S., Dijkstra, J., Bannink, A., Kebreab, E. & France, J. (2007). Some methodological and analytical considerations regarding application of the gas production technique. *Animal Feed Science and Technology* 135(1-2), 139-156.

López, S., Dijkstra, J. & France, J. (2000). Prediction of energy supply in ruminants, with emphasis on forages. In: Givens, D.I., Owen, E., Axford, R.F.E. & Omed, H.M. (Eds.) *Forage Evaluation in Ruminant Nutrition* CABI Puplishing. ISBN 0 85199 344 3, pp. 63-94.

Luc, D.H., Thu, N.V. & Preston, T.R. (2009). Feed intake, rumen fermentation, microbial protein synthesis and nitrogen retention in growing cattle given maize or molasses with two levels of crude protein as supplements to a basal diet of rice straw and grass. *Livestock Research for Rural Development* 21(7), 1-9.

MacRae, J.C. & Lobley, G.E. (1986). Interactions between energy and protein. In: Milligan, L.P., Grovum, W.L. & Dobson, A. (Eds.) *Control of digestion and metabolism in ruminants*. Toronto, Canada: Prentice-Hall. ISBN 9780835910149, p. 365.

Makkar, H.P.S. (2002). Applications of the *in vitro* gas method in the evaluation of feed resources, and enhancement of nutritional value of tannin-rich tree/browse leaves and agro-industrial by-products. In: *International Atomic Energy Agency Technical Documents (IAEA-TECDOCs)*. pp. 23-40.

Makkar, H.P.S. (2003). Effects and fate of tannins in ruminant animals, adaptation to tannins, and strategies to overcome detrimental effects of feeding tannin-rich feeds. *Small Ruminant Research* 49(3), 241-256.

Man, N.V. (2001). *Better use of local forages for dairy cattle in Vietnam: Improving grasses, rice straw and protein rich forages*. Diss. Uppsala:Swedish University of Agricultural Science, Uppsala, Sweden. ISBN 91-576-5817-X.

Man, N.V., Hao, N.V. & Tri, V.M. (1995). Biomass production of some leguminous shrubs and tree in Vietnam. *Livestock Research for Rural Development* 7(2), 1-6.

Man, N.V. & Wiktorsson, H. (2001). Cassava tops ensiled with or without molasses as additive effects on quality, feed intake and digestibility by heifers. *Asian-Australasian Journal of Animal Sciences* 14(5), 624-630.

Menke, K.H., Raab, L., Salewski, A., Steingass, H. & Fritz, D. (1979). The estimation of digestibility and metabolizable energy content of ruminant feedstuffs from the gas production when they are incubated with rumen liquor *in vitro*. *Journal of Agricultural Science (Cambridge)* 92, 217-222.

Menke, K.H. & Steingass, H. (1988). Estimation of the energetic feed value from chemical analysis and *in vitro* gas production using rumen fluid *Animal Research and Development* 28, 7-55.

Min, B.R., Barry, T.N., Attwood, G.T. & McNabb, W.C. (2003). The effect of condensed tannins on the nutrition and health of ruminants fed fresh temperate forages: a review. *Animal Feed Science and Technology* 106(1-4), 3-19.

Minitab (2003). *Minitab Statistical Software, Release 14 for Window*. Version: 14. Minitab Inc., State College, PA.

Minson, D.J. (1990). *Forage in ruminant nutrition*. New York: Academic Press. ISBN 0-12-498310-3.

Moore, C.P. & Cock, J.H. (1985). Cassava foliage silage as a feed source for Zebu calves in the tropics. *Tropical Agriculture* 62(2), 142-144.

Mueller-Harvey, I. (2006). Unravelling the conundrum of tannins in animal nutrition and health. *Journal of the Science of Food and Agriculture* 86(13), 2010-2037.

Muhammed, S., Stewart, C.S. & Acamovic, T. (1994). Effects of tannic acid on cellulose degradation, adhesion and enzymatic activity of rumen microorganisms. In: *Proceedings of the Society of Nutritional Physiology*. pp. 174-176.

Mupangwa, J.F., Ngongoni, N.T., Topps, J.H., Acamovic, T., Hamudikuwanda, H. & Ndlovu, L.R. (2000). Dry matter intake, apparent digestibility and excretion of purine derivatives in sheep fed tropical legume hay. *Small Ruminant Research* 36(3), 261-268.

Nin, A., Lapar, M.L. & Ehui, S. (2003). Globalization, trade liberalization and poverty alleviation in Southeast Asia: the case of the livestock sector in Vietnam. In: *The 6th annual conference on global economic analysis, June 12-14, 2003*. Scheveningen, The Hague, The Netherlands. pp. 1–38.

Nolan, J.V. & Dobos, R.C. (2005). Nitrogen transactions in ruminants. In: Dijkstra, J., Forbes, J.M. & France, J. (Eds.) *Quantitative Aspects of Ruminant Digestion and Metabolism* CABI Puplishing, Wallingford, UK. ISBN 0 85199 8143, pp. 177–206.

Norton, B.W. (2000). The significance of tannins in tropical animal production. In: Brooker, J.D. (Ed.) *Tannins in Livestock and Human Nutrition: Proceedings of an International Workshop*. ACIAR Proceedings No.92. pp. 14-23.

NRC (1996). *Nutrient Requirements of Beef Cattle (7th Ed.)*: National Academic Press, Washington, DC.

Ogle, B. & Phuc, B.H.N. (1997). Sustainable intensive livestock-based systems in Vietnam. *IRD Currents* 13(14), 16-22.

Oke, O.L. (1979). Some aspect s of the role of cyanogenic glycosides in nutrition. *World Review of Nutrition and Dietetics* 33, 70-103.
Oldham, J.D., Buttery, P.J., Swan, H. & Lewis, D. (1977). Interactions between dietary carbohydrate and nitrogen and digestion in sheep. *Journal of Agricultural Science (Cambridge)* 89, 467-479.
Ørskov, E.R., Hovell, F.D. & Mould, F. (1980). The use of the nylon bag technique for the evaluation of feedstuffs. *Tropical Animal Production* 5(3), 195-213.
Ørskov, E.R. & McDonald, I. (1979). The estimation of protein degradability in the rumen from incubation measurements weight according to the rate of passage. *Journal of Agricultural Science (Cambridge)* 92, 499-503.
Osuntokun, B.O. (1981). Cassava diet, chronic cyanide intoxication and neuropathy in the Nigerian Africans. *World Review of Nutrition and Dietetics* 36, 141-173.
Paris, T.R. (2002). Crop-animal systems in Asia: socio-economic benefits and impacts on rural livelihoods. *Agricultural Systems* 71(1-2), 147-168.
Phengvichith, V. & Ledin, I. (2007). Effects of supplementing gamba grass(*Andropogon gayanus*) with cassava (*Manihot esculenta* Crantz) hay and cassava root chips on feed intake, digestibility and growth in goats. *Asian-Australasian Journal of Animal Sciences* 20(5), 725-732.
Phung, L.D. (2001). *A system analysis of the use of local Yellow and F1 (red Sindhi x Yellow local) cattle at the hoseholds in the mountainous and lowland zones in QuangNgai, Vietnam*. Diss.:Wageningen University, Netherlands.
Pirt, S.J. (1975). *Principles of microbe and cell cultivation*. Oxford: Blackwell Scientific Publications. ISBN 0470690380.
Poonam, V.R. & Hahn, S.K. (1984). An automated enzymic assay for determining the cyanide content of cassava (*Manihot esculenta* Crantz) and cassava products. *Journal of the Science of Food and Agriculture* 35(4), 426-436.
Poppi, D. (1990). Manipulation of nutrient supply to animals at pasture, opportunities and consequences. In: *Proceedings of Fifth Australasian Association for Animal Production*. Miaoli, Taiwan. pp. 42-79.
Poppi, D.P. & McLennan, S.R. (1995). Protein and energy utilization by ruminants at pasture. *Journal of Animal Science* 73(1), 278-290.
Poppi, D.P. & Norton, B.W. (1995). Intake of tropical legumes. In: D'Mello, J.P.F. & Devendra, C. (Eds.) *Tropical legumes in animal nutrition* CAB International. ISBN 0 85198 926 8, pp. 173-190.
Preston, T.R. (2001). Potential of cassava in integrated farming systems. In: *Proceedings of International workshop on current research and development*

*on use of cassava as animal feed*. Khon Kaen University, Thailand, July 23-24, 2001. pp. 3-12.

Preston, T.R. & Leng, R.A. (1987). Matching ruminant production systems with available resources in the tropics and sub-tropics. In. Armidale Australia: Penambul Book. ISBN 0958829012, p. 245.

Ramírez-Restrepo, C.A. & Barry, T.N. (2005). Alternative temperate forages containing secondary compounds for improving sustainable productivity in grazing ruminants. *Animal Feed Science and Technology* 120(3-4), 179-201.

Ravindran, V. (1993). Cassava leaves as animal feed: potential and limitations. *Journal of the Science of Food and Agriculture* 61(2), 141-150.

Reed, J.D. (1995). Nutritional toxicology of tannins and related polyphenols in forage legumes. *Journal of Animal Science* 73(5), 1516-1528.

Reed, J.D., Horvath, P.J., Allen, M.S. & Van Soest, P.J. (1985). Gravimetric determination of soluble phenolics including tannins from leaves by precipitation with trivalent ytterbium. *Journal of the Science of Food and Agriculture* 36, 255-261.

Rogers, D. & Milner, M. (1963). Amino acid profile of manioc leaf protein in relation to nutritive value. *Economic Botany* 17(3), 211-216.

Rooke, J.A. & Armstrong, D.G. (2007). The importance of the form of nitrogen on microbial protein synthesis in the rumen of cattle receiving grass silage and continuous intrarumen infusions of sucrose. *British Journal of Nutrition* 61(01), 113-121.

Salawu, M.B. (1997). *The nutritive value of the leguminous browse Calliandra calothyrsus and the role of condensed tannins in ruminant feeds*. Diss.:University of Aberdeen, UK.

Seng, M. (2001). Effect of a single drench of cooking oil on the rumen ecosystem and performance of young local" yellow cattle" fed rice straw and cassava foliage. *Livestock Research for Rural Development* 13(4), 1-9.

Silanikove, N., Perevolotsky, A. & Provenza, F.D. (2001). Use of tannin-binding chemicals to assay for tannins and their negative postingestive effects in ruminants. *Animal Feed Science and Technology* 91(1-2), 69-81.

Smith, O.B., Idowu, O.A., Asaolu, V.O. & Odunlami, O. (1989). Comparative rumen degradability of forages, browse, crop residues and agricultural by-products. In: Wilson, R.T. & Melaku, A. (Eds.) *Proceedings of African Small Ruminant Research and Development, 18-25 January, 1989*. Bamenda, Cameroon: ILCA/IFS/IAR. pp. 204-218.

Smith, O.B. & Van Houtert, M.F.J. (1987). The feeding value of *Gliricidia sepium*. A review. *World Animal Review (FAO)* 62, 57-68.

Soto, R.C., Muhammed, S.A., Newbold, C.J., Stewart, C.S. & Wallace, R.J. (1994). Influence of peptides, amino acids and urea on microbial activity in the rumen of sheep receiving grass hay and on the growth of rumen bacteria *in vitro*. *Animal Feed Science and Technology* 49(1-2), 151-161.

Streeter, M.N., Wagner, D.G., Owens, F.N. & Hibberd, C.A. (1989). Combinations of high-moisture harvested sorghum grain and dry-rolled corn: Effects on site and extent of digestion in beef heifers. *Journal of Animal Science* 67(6), 1623-1633.

Su, V.V. & Binh, D.V. (2002). Cattle Breeding in Vietnam. In: Jack Allen & Ancharlie Na-Chiangmai (Eds.) *Development Strategies for Genetic Evaluation for Beef Production in Developing Countries, ACIAR Proceedings 108*. Khon Kaen Province, Thailand, July 23–28 2001: Australian Centre for International Agricultural Research (ACIAR). pp. 98-101.

Sutton, J.D. (1985). Digestion and absorption of energy substrates in the lactating cow. *Journal of Dairy Science* 68(12), 3376-3393.

Terrill, T.H., Rowan, A.M., Douglas, G.B. & Barry, T.N. (1992). Determination of extractable and bound condensed tannin concentrations in forage plants, protein concentrate meals and cereal grains. *Journal of the Science of Food and Agriculture* 58(3), 321-329.

Tewe, O.O. (1992). Detoxification of cassava products and effects of residual toxins on consuming animals. In: Machin, D. & Nyvold, S. (Eds.) *Roots, tubers, plantains and bananas in animal feeding*. FAO Animal Production and Health Paper 95, Rome, Italy. pp. 81-98.

Thang, C.M. (2005). *Effects of intergrated production of cassava and Phaseolus calcaratus legume and use as a protein source for growing cattle (MSc. thesis).* Diss.:Swedish University of Agricultural Science, Uppsala, Sweden.

Theodorou, M.K., Barahona, R., Kingston-Smith, A., Sanchez, S., Lascano, C., Owen, E. & Morris, P. (2000). New perspectives on the degradation of plant biomass in the rumen in the absence and presence of condensed tannins. In: Brooker, J.D. (Ed.) *Tannins in livestock and human nutrition. Proceedings of an International Workshop*. Australian Centre for International Agricultural Reseach (ACIAR), Canberra, Australia. pp. 44-51.

Tiemann, T.T., Lascano, C.E., Wettstein, H.R., Mayer, A.C., Kreuzer, M. & Hess, H.D. (2008). Effect of the tropical tannin-rich shrub legumes *Calliandra calothyrsus* and *Flemingia macrophylla* on methane emission and nitrogen and energy balance in growing lambs. *Animal* 2(05), 790-799.

Tien, N.P., Dung, N.T., Mui, N.T., Binh, D.V. & Preston, T.R. (2003). Improving biomass yield and soil fertility by associations of Flemingia (*Flemingia macrophylla*) with Mulbery (*Morus alba*) and cassava (*Manihot esculenta*) on the slopping land in Bavi area. In: Preston, T.R. & Ogle, B. (Eds.) *Sustainable Livestock Production on local Feed Resources - Proceedings of a National Workshop - Seminar, 25-27 March*. Hue, Vietnam: Agriculture Publishing House, HCM city, Vietnam. pp. 27-35.

Trach, N.X. (1998). The need for improved utilisation of rice straw as feed for ruminants in Vietnam: An overview. *Livestock Research for Rural Development* 10(2), 1-14.

Trach, N.X. (2003). Possible consequences of inappropriate intensification of animal production in Vietnam and approaches to a void. *Livestock Research for Rural Development* 15(1), 1-7.

Tung, C.M., Liang, J.B., Tan, S.L., Ong, H.K. & Jelan, Z.A. (2001). Foliage productivity and growth persistency of three local cassava varieties. *Asian-Australasian Journal of Animal Sciences* 14, 1253-1259.

Tung, D.X., Thanh, N.D., Duc, D.V., Quoc, N.V., Quy, M.T., Thuy, T.P.T. & Loan, N.T. (2007). Identifying factors affecting to the technical economic efficiency of beef production in four different ecological zones in the North Vietnam. In: *Annual Science Report, 2006*. Hanoi, 1-2 Aug 2007: National Institute of Animal Husbandry, Ministry of Agriculture and Rural Development.

Udén, P. (2000). Ruminal metabolism of buffer-soluble proteins, peptides and amino acids *in vitro*. In: Mcnamara, J.P., France, J. & Beever, D.E. (Eds.) *Modelling nutrient utilization in farm animals*. Wallingford, UK: CABI Publishing. ISBN 0 85199 449 0, pp. 63-72.

Ulyatt, M.J., Thomson, D.J., Beever, D.E., Evans, R.T. & Haines, M.J. (1988). The digestion of perennial ryegrass (*Lolium perenne* cv. Melle) and white clover (*Trifolium repens* cv. Blanca) by grazing cattle. *British Journal of Nutrition* 60(01), 137-149.

Van, D.T.T., Mui, N.T. & Binh, D.V. (2001). Cassava as small ruminant feed in the hilly and mountainous area of Bavi district in Northern Vietnam. In: Preston, T.R., Ogle, B. & Wanapat, M. (Eds.) *Proceedings of an international workshop on current research and development on use of cassava as animal Feed, July 23-24*. Khon Kaen University, Thailand. pp. 145-154.

Van Soest, P.J., Robertson, J.B. & Lewis, B.A. (1991). Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. *Journal of Dairy Science* 74(10), 3583-3597.

Vongsamphanh, P. (2004). Comparison of cassava hay yield and chemical composition of local and introduced varieties and effects of levels of cassava hay supplementation in native beef cattle fed on rice straw. *Livestock Research for Rural Development* 16(8), 1-11.

Wanapat, M. (2001). Role of cassava hay as animal feed in the tropic. In: Preston, T.R., Ogle, B. & Wanapat, M. (Eds.) *Proceedings of International workshop on current research and development on use of cassava as animal feed, July 23-25*. KhonKaen University, Thailand. pp. 13-20.

Wanapat, M. (2003). Manipulation of cassava cultivation and utilization to improve protein to energy biomass for livestock feeding in the tropics. *Asian-Australasian Journal of Animal Sciences* 16(3), 463-472.

Wanapat, M. (2008). Potential uses of local feed resources for ruminants. *Tropical Animal Health and Production* 41(7), 1035-1049.

Wanapat, M., Petlum, A. & Pimpa, O. (2000a). Supplementation of cassava hay to replace concentrate use in lactating Holstein Friesian crossbreds. *Asian-Australasian Journal of Animal Sciences* 13(5), 600-604.

Wanapat, M., Pimpa, O., Petlum, A. & Boontao, U. (1997). Cassava hay: A new strategic feed for ruminants during the dry season. *Livestock Research for Rural Development* 9(2), 1-5.

Wanapat, M., Pimpa, O., Petlum, A., Wachirapakorn, C. & Yuanklang, C. (2000b). Participation scheme of smallholder dairy farmers in the Northeast Thailand on improving feeding systems. *Asian-Australasian Journal of Animal Sciences* 13(6), 830-836.

Wanapat, M., Puramongkon, T. & Siphuak, W. (2000c). Feeding of cassava hay for lactating dairy cows. *Asian-Australasian Journal of Animal Sciences* 13(4), 478-482.

Williams, B.A. (2000). Cumulative gas-production techniques for forage evaluation. In: Givens, D.I., Owen, E., Axford, R.F.E. & Omed, H.M. (Eds.) *Forage Evaluation in Ruminant Nutrition* CABI puplishing. ISBN 0 85199 344 3, pp. 189-213.

# Acknowledgements

The studies presented in this thesis were conducted at Bavi Cattle and Forage Research Centre belonging to the National Institute of Animal Science (NIAS), Hanoi, Vietnam, and at the Yenphu Dairy and Beef Cattle Breeding Centre in Ninhbinh province, and at the Department of Animal Nutrition and Management of the Swedish University of Agricultural Science (SLU). The work was supported by the Swedish International Development Agency (SIDA), Department for Research Cooperation with Developing Countries (SAREC). I am grateful for the support from all these institutions.

I would like to express my sincerely gratitude to:

Assoc.prof. Jan Bertilsson, my main supervisor for valuable and constructive advice and useful suggestions during my study, particularly with respect to editing the manuscripts and publishing the papers and in the writing of the thesis as well.

Prof. Inger Ledin, my co-supervisor, and my grand mother as well, for your comments, suggestions and criticisms helping me to complete this work. Special thanks for the ways you taught me to find solutions and improve by myself.

Prof. Dr. Brian Ogle, Director of "Vietnam-SAREC sustainable Livestock Production Systems" project, for valuable support, especially for reading through all my papers for English correction. Thanks for welcoming me warmly in my first time in Sweden.

Professors, lecturers and assistant lecturers in the courses from the Swedish University of Agricultural Sciences, Sweden, which I have attended during my studies for sharing their great knowledge and experiences.

Dr. Ngo Van Man, Regional Coordinator, Dr. Vu Chi Cuong, Vietnamese Coordinator; Mr. Nguyen Van Cong for the monitoring and for caring about me with all their hearts.

Ass. prof. Hoang Van Tieu, the director of NIAS for permission to study, and facilitation and encouragement of the work.
My department of Science, Planning and International Cooperation - NIAS, Dr. Nguyen Huu Tao, Dr. Pham Sy Tiep, Ass. Prof. Le Thi Thuy, Mrs. Tran Thi Thanh, Mr. Nguyen Manh Dzung, Mrs.Trinh Thi Thanh, Mrs. Do Viet Hang, Mrs. Nguyen Thi Thanh Huong, Mrs. Tran Thi Kim Anh for facilitation and taking care of the work when I studied in abroad.

My friends, Mr. Vo Tong Anh for your kind help with computer skills; Mr. Tran Ho Quang for sharing the great pictures and a pleasant dinner together in a kebab restaurant in Uppsala; Dr. Nguyen Manh Khai for great skills in EndNote program; Mr. Vo Lam, Mrs. Nguyen Thi Thuy and Mrs. Nguyen Duy Quynh Tram for teaching me how to cook and preparing the meals; Mrs. Hoang Huong Giang for keeping a warm atmosphere; Dr. Le Thi Thanh Van, Dr. Tran Thi Thu Hong, Dr. Nguyen Van Thieu, for your sharing, help me during the time away from home; Ms. Hao yu Liu, Mrs Lampheuy, and Mr. Keo Sath for your friendship and the great talks with nonsense topics for relaxing; Mrs. Daovy Kongmanila for sharing the funny stories. And with whom I share the special memory of a spicy salmon fish salad that I never will forget.

To my family, my respectable mother and father, my relatives, for all their support and encouragement during these years. Thank you sincerely for believing in me.
My wife, Pham Thi Bich Huong, who encouraged me to embark in my PhD studies while she was all alone during my regular absence for conducting experiments and study in Sweden, and took care of our two lovely daughters, Chu Thuy Linh and Chu Dieu Huong, for all your love, and inspiration.

Lastly, my gratitude to all those anonymous peoples who helped me one way or another and whose names can not appear in here.