Cassava Foliage for Monogastric Animals

Forage Yield, Digestion, Influence on Gut Development and Nutritive Value

Khieu Borin

Faculty of Veterinary Medicine and Animal Science
Department of Animal Nutrition and Management
Uppsala

Doctoral thesis
Swedish University of Agricultural Sciences
Uppsala 2005

Abstract

The study was aiming to evaluate cassava (*Manihot esculenta* Crantz) managed as a forage crop with respect to biomass yield, and to determine its value as a feed for monogastric animals.

Two common cassava varieties, one short- (ST) and one long-term (LT), grown in Cambodia, were studied with the aim of assessing forage yield, and effects on soil fertility and the utilization of the leaves as a feed for pigs and poultry. The ST variety was intercropped with *Desmanthus virgatus* or *Gliricidia sepium* and the LT variety was fertilised with effluent from biodigesters loaded with either cow (CM) or pig manure (PM). Intercropping of cassava with legumes increased total biomass dry matter (DM) yield and resulted in a higher proportion of cassava leaves, while fertilizing with PM produced the highest cassava total forage and leaf DM yields. The crude protein (CP) content on a DM basis in cassava leaf, and stem plus petiole, was 24-25% and 8-10%, respectively. It was estimated that *D. virgatus* and *G. sepium* fixed 1/3 and 1/2 of the amounts of nitrogen (N) removed by the cassava forage, respectively, while the N uptake was 67 and 27% above the N inputs from PM and CM, respectively.

Both sun-drying and ensiling were efficient in reducing the hydrogen cyanide (HCN) content in cassava leaves, although ensiling after sun wilting was more effective. Pigs utilized the nutrients in cassava leaf silage (CLS) more efficiently than in cassava leaf meal (CLM) and leaves from the ST and LT varieties were utilized equally. Mong Cai (MC) pigs digested dietary fibre components better than Large White x Yorkshire (LxY) pigs, although N utilization was higher in LxY than MC pigs.

Increasing CLM level in poultry diets slightly increased DM intake and decreased the digestibility of DM. Increasing dietary CLM increased the weight and length of most parts of the gastrointestinal tract (GIT) and associated internal organs, except for proventriculus weight and colon length. Broiler chickens and White Pekin ducks had higher GIT and internal organ weights and lengths than local ducks and chickens, respectively, when expressed in absolute units. However, when expressed as kg^{-1} body weight, these values were higher in the local birds, except for small intestine weights, which were similar. Ducks had longer small intestines and higher small intestine, gizzard, pancreas and liver weights as

kg^{-1} body weight than chickens, while weights of caeca and colon, were higher in chickens.

It is concluded that both ST and LT cassava varieties have the potential to produce high protein forage for monogastric animals. Pigs utilized cassava leaf silage more efficiently than sun-dried leaves. Increasing cassava leaf meal in the diet reduced DM digestibility and increased the length and weight of most parts of the gastrointestinal tract and associated organs of chickens and ducks.

Author's address: Khieu Borin, Centre for Livestock and Agriculture Development (CelAgrid), Kandal Village, Rolous commune, Kandal Stung district, Kandal province, P.O. Box 2423, Phnom Penh 3, Cambodia.
E-mail: khieu_borin@forum.org.kh

For
My parents,
My wife, Khlok Peng Thol,
My children, Khieu Mayuty and Mavanneh

Contents

Appendix

The present thesis is based on the following papers, which are referred in the text by their Roman numerals.

I. Khieu Borin and Frankow-Lindberg, B.E. (2005). Effects of legumes-cassava intercropping on cassava forage and biomass production. *Journal of Sustainable Agriculture* (*In press*).

II. Khieu Borin and Frankow-Lindberg, B.E. (2005). Forage yield from cassava grown as a perennial crop fertilised with effluent from biodigesters loaded with pig or cow manure and the effect on soil fertility. *Biological Agriculture and Horticulture* (*Accepted*).

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

IV. Khieu Borin, Lindberg, J.E. and Ogle, R.B. (2005). Effect of cassava leaf meal on gastrointestinal tract and organ development of local and exotic chickens and ducks. *Journal of Animal Physiology and Animal Nutrition.* (*In press*).

List of abbreviations

ADF	Acid detergent fibre
BC	Broiler chicken
CF	Crude fibre
CLM	Cassava leaf meal
CLS	Cassava leaf silage
CM	Cow manure
CP	Crude Protein
CTTAD	Coefficient of Total Tract Apparent Digestibility
DM	Dry Matter
GE	Gross Energy
GIT	Gastrointestinal tract
HCN	Hydrogen Cyanide
LC	Local chicken
LD	Local duck
LT	Long term
MC	Mong Cai
N	Nitrogen
NDF	Neutral Detergent Fibre
OM	Organic Matter
PD	White Pekin duck
PM	Pig manure
ST	Short term

Introduction

Both energy and protein are important components in animal feeds for maintenance, growth and reproduction. The supply of nutrients that are adequate in terms of both quality and quantity allows animals to perform according to the genetic potential of each breed and species. However, to optimize the growth of animals by providing them with diets of optimum quality is not the primary aim of smallholder livestock producers in development countries; the maximum economic return is normally the principal goal. In this respect the feed resources locally available or grown on farm play a vital role in minimizing the production cost, allowing small and medium scale producers to obtain a better economic return.

The quality of a feedstuff is dependent on several biological factors, such as the dry matter and crude protein content in relation to the amino acid balance, and the contents of metabolisable energy, crude fibre and anti-nutritional factors. In general, the factors limiting the optimal use of farm feed resources for monogastric animals include low palatability, often due to high levels of anti-nutritional factors (e.g. in cassava), low dry matter content (e.g. in duckweed) or high fibre content (e.g. in banana leaves and stem) and the low biological value of the protein. These are probably the most common reasons why potentially useful feed resources in the tropics have often not been given much attention by researchers and livestock producers.

Cassava *(Manihot esculenta* Crantz*)* originated in the eastern regions of tropical South America and Western and Southern Mexico (FAO, 1998), and is one of the most potentially valuable tropical crops due to its high tuber yield and simplicity of cultivation. However, it has been shown that the fertility of the soil decreases after the cultivation of cassava for root production without fertilization. In cassava producing countries, better soils are almost always devoted to more profitable crops, leaving those areas with soil problems for cassava (high aluminium content, low exchangeable base content, high P fixation, and various degrees of erosion) (Howeler, 1991). Presently, cassava is widely grown throughout tropical and some sub-tropical areas. Cassava is generally cultivated for root production for both humans and animals. Cassava covered an area of 18.8 million ha in 1993, estimated to increase to 22.9-24.2 million ha in 2020, with a growth rate of 0.94 percent annually (Scott *et al.*, 2000). Of the total world area for cassava cultivation in 1993, Africa accounted for 63.3%, followed by Southeast Asia with 18.6%.

Cassava leaves have been reported to have good amino acid content, comparable to soybean meal, but are deficient in the sulphur-containing amino acids (Eggum, 1970; Phuc *et al.,* 2000a). However, the most

important limitation for the utilization of cassava leaves as feed for monogastric animals is probably the content of anti-nutritional factors (HCN and tannins) (Awoyinka *et al.*, 1995) and fibre (Diaz *et al.*, 1997). Techniques for reducing the level of anti-nutritional factors in both cassava leaves and roots have been developed, such as ensiling, sun-drying, boiling and fermentation, and these have been confirmed as making the products safe for use as human food and animal feed (Gomez and Valdivieso, 1985 and 1988; Panigrahi *et al.*, 1992; Padmaja, 1995; Essers *et al.,* 1995; Diaz *et al.,* 1997).

For several decades, cassava has been given considerable attention by a number of research institutes in developing countries, in particular the International Centre for Tropical Agriculture (CIAT). Research has mainly concentrated on tuber production, and less attention has been given to evaluating cassava for forage production. However, the last few years, with financial support from Sida/SAREC to the MEKARN (Mekong Basin Animal Research Network) programme, researchers in South-East Asia have focused their efforts on cassava with respect to forage production and its utilization as feed.

Cassava is able to produce reasonable root yields (10-15 t ha^{-1}) without inputs on eroded soils (Howeler, 1991) and this could be an explanation for the general conclusion that soil fertility eventually decreases after the cultivation of cassava for root production. Improvement of the nutritional status of cassava through fertilization or intercropping is important, not only to increase the quantity and quality of the cassava produced, but directly and indirectly to improve soil fertility. Cadavid *et al.* (1998) reported that the application of nitrogen (N), phosphorus (P) and potassium (K) fertilizer to cassava significantly increased root and top biomass and reduced root HCN content. Moore (1976) reported high potential yields of foliage when cassava was managed as a semi-perennial crop with repeated harvesting at 2-3 month intervals.

AFRIS (2004) reported a range in CP content of the leaves from 22.8-24.1% and 25.4-29.0% in the rainy and dry seasons, respectively, and also that the CP content depended on the age of the leaves.

Several studies have been undertaken on the use of cassava leaves as feed for cattle (Moore and Cock, 1985; Man and Wiktorsson, 2001), goats (Seng Sokerya and Rodriguez, 2001; Van *et al.,* 2001), pigs (Ravindran *et al,* 1983 and 1987; Phuc *et al.,* 2000b) and poultry (Ravindran *et al.*, 1986, Montillas *et al.*, 1985). However, the leaves evaluated in much of the earlier research were mainly from the plant after harvesting the tubers and these probably have a higher fibre content due to their maturity. Therefore

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in our study it was considered of interest to focus on leaves harvested at a younger stage with appropriate nutrient inputs.

Another important issue is the potential of local indigenous and improved animal breeds with respect to their utilization of cassava leaves. Fevrier *et al.* (1992) reported that indigenous Chinese Meishan pigs utilized fibrous components of the feed better than exotic breeds, but only in the case of very high levels of dietary ADF and in the presence of highly digestible protein sources. Jamroz *et al.* (2001) fed diets containing 40% of barley to chickens, ducks and geese from 21-42 days of age, and found that chickens digested dietary fibre better than ducks and geese.

Aims of the studies

The general objectives of the present studies were to investigate (1) the effect of intercropping cassava and legume species and of fertilization on cassava foliage production and soil fertility and (2) to evaluate cassava leaves as feed for different species and breeds of monogastric animals.

The specific objectives were:
- To determine the yield of cassava foliage when intercropped with *Desmanthus virgatus* and *Gliricidia sepium*;
- To study the effect of effluents from biodigesters loaded with cow or pig manure as fertiliser sources on cassava foliage production;
- To evaluate preservation techniques and the nutritive value of cassava forage harvested at an age of 60 days;
- To assess the fertility of soils after 18 months of intensive cassava foliage production;
- To determine the digestibility of nutrients and nitrogen utilization by local and improved pigs fed ensiled or dried cassava leaves from long- and short-term varieties with sugar palm juice and crude palm oil as the energy sources; and
- To study the effect of different levels of cassava leaf meal inclusion in the diet on dry matter digestibility and the development of the gastrointestinal tract and associated internal organs of local and exotic chickens and ducks.

Background

Livestock, food security and poverty alleviation in Cambodia

Livestock make significant contributions to food security and income generation of people in the developing world. Livestock contribute up to 20% of the agricultural GDP in the Southeast Asian sub-region, which includes Cambodia, Indonesia, Laos, Malaysia, Myanmar (Burma), the Philippines, Thailand and Vietnam (Devendra and Thomas, 2002).

An analysis by the World Food Programme in Cambodia suggested that low agricultural productivity caused by poor opportunities for using improved technologies, poor marketing infrastructure and declining access to common property resources, such as forests and fisheries, are major causes of food insecurity and malnutrition. Other causes of rural poverty include: (i) high transportation costs impeding efficient marketing and distribution of outputs; (ii) poor basic infrastructure, including the transportation network; (iii) inadequate or lack of access to affordable credit and high level of indebtedness; and (iv) lack of tenure and land title for many poor households.

The small producers in Cambodia hold the majority of livestock (90 ± 5%) and the mean holding of livestock in the rural areas has been reported to be 1.6 large ruminants, 0.96 pigs and 13 head of poultry per household, mostly managed in scavenging or semi-scavenging systems (European Commission, 2003). Farmers keep their animals in traditional scavenging systems as a means of risk management rather than in systems more orientated toward increased production and incomes. Besides large ruminants (cattle and buffalo), mainly used for draught, there is a considerable contribution from pigs and poultry to farmers' livelihoods and food security. The majority of people raise local breeds, including pigs, chickens and ducks, although small and medium scale commercial farms close to the cities and towns keep exotic or improved breeds in order to meet the increasing demand for meat and eggs of the fast growing and increasingly affluent urban populations.

Livestock-based farming systems

Crop-animal systems have played an important role in the mixed, small-farm households. Livestock convert plant materials of low nutritive value to high quality products such as meat and milk and return nutrients to the

soil in the form of manure. This synergistic interaction between livestock and crops improves the sustainability of the farm and maintains or improves soil fertility. In general farmers understand the importance of having animals as the entry point to improve farm productivity. However, in many cases, limited resources, including land area and shortage of capital to purchase, in particular, large ruminants (cattle and buffaloes), prevent them from developing integrated farming systems. In the case of Cambodia, it was observed that farmers keep more pigs after harvesting rice due to the availability of rice by-products such as broken rice and rice bran (Khieu Borin, personal observation). Also many flocks of 200-500 ducks are released to eat the remaining paddy in the field. Those farmers that do not own any large ruminants give rice straw to those who have cattle or buffaloes and in return get manure to fertilize their crops. Thorne and Tanner (2002) reported that farmers claimed that plant growth responses are greater with manure-based compost than with fresh manure or inorganic fertilizers. Therefore appropriate management of the animals within the farming system is important in order to maximize the use of manure for crops, and in return to optimize the utilization of crop residues as animal feed, thus resulting in an efficient 'food-feed system'.

Several plants, including those producing roots and tubers, are potentially valuable dual-purpose crops, producing human food and animal feed. Cassava is also known as manioc, tapioca and yuca, in different parts of the tropics, and is an important subsistence crop for resource-poor farmers due its capacity to produce reasonable yields for home consumption, animal feed and income, while at the same time being simple to cultivate and requiring low inputs. In addition, with adequate nutrient inputs and proper management practices, it is expected that both tuber and leaf yields will increase and the sustainability of the system improve.

In Cambodia, cassava cultivation for mainly tuber production was estimated to increase by around 57% from 1993 (16,000 ha) to 2003 (25,039 ha) (FAO/RAP, 2003). However, the main increase was observed in 2002-2003 due to the introduction of a new variety and a high market demand. The national average tuber production of cassava in 2003 was 13.2 tonnes ha^{-1} (FAO/RAP, 2003) although there was a large variation, with yields from 2.3 to 18.3 tonnes ha^{-1}. This variation was due to differences in soil fertility in the different regions in Cambodia, and to different varieties and cultivation practices. The 6-8 months variety (short-term) is very common in the lowland rain-fed rice region, with tuber yields ranging from 4-6 tonnes ha^{-1}, and is generally cultivated without fertilization. In the upland regions cassava is planted by ethnic minority communities in association with upland rice and sesame, and between rubber trees in the red basaltic soil region. It is also cultivated along the Mekong river, where it is planted after the water recedes in November-

December and harvested before the subsequent floods (July-August). Cultivation of cassava is also common in the backyards and gardens of the lowland, rain-fed rice region. In recent years, a long-term variety has been unofficially introduced, and although it has no formal name its physical characteristics are close to the KS50 variety from Thailand. This variety is presently grown in extensive areas of the red basaltic soil region in Cambodia for commercial root production.

Traditional farming systems rely on the natural recycling of nutrients to maintain soil fertility and consequently the productivity of the crops. Today, due to the intensive use of land and the shortening of the fallow period, nutrient management should be applied, either through the direct application of organic or inorganic fertilizers or by adoption of intercropping systems. However, due to their unavailability, lack of knowledge concerning their application, and the high price of inorganic fertilizers, it would be more beneficial for the small-scale farmers to increase the utilization of existing on-farm resources, such as manure.

Potential and limitations of cassava leaves as animal feed

Cassava, one of the world top calorie producers for human consumption, is generally grown without fertilization on soils with poor fertility where other crops would fail (Howeler and Cadavid, 1990). Cassava can survive prolonged water deficit (Alves and Setter, 2000), and is tolerant to acid soils, but the yield is limited by poor phosphorus (P) supply (Howeler, 1985). However, it is possible to obtain from cassava leaves more than 6 tonnes of crude protein ha^{-1} $year^{-1}$ with proper agronomic practices directed toward foliage harvesting (AFRIS, 2004). In the sandy soils of northern Colombia applying N, P, and K fertilizer in moderate amounts significantly increased root and top biomass and reduced root hydrogen cyanide (HCN) concentration (Cadavid, et al., 1998). In Thailand applying approximately 10 kg N ha^{-1} from cow manure, resulted in DM yields and CP contents of cassava forage of 3.6 to 4.4 tonnes ha^{-1} and 20.6 to 22.0%, respectively (Poungchompu et al., 2001).

The nutritional limitations of cassava leaves include the HCN content, low digestible energy, bulkiness and possibly the high tannin content (Ravindran et al., 1986). The cyanogenetic glycosides, which on hydrolysis yield toxic HCN, may limit its use as a monogastric animal feed. The HCN concentration, produced after the action of hydrolytic enzymes occurring in the plant on the cyanogenetic glycosides, is influenced by the nutritional status and age of the plant (Ravindran and Ravindran, 1988), and higher HCN levels were found in leaves from bitter than in sweeter varieties (Tewe and Iyayi, 1989). The average HCN content found in a Sri Lankan variety (Ravindran et al. 1987) was over

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4,000 mg kg^{-1}. However, the HCN concentration and the bitterness associated with high cyanogenetic glycoside contents in leaves (Sundaresan *et al.,* 1987) decreases with the maturity of the leaves. Ravindran et al. (1987) reported that HCN contents of cassava leaf blades in the stage of the expanding (1-4 leaves), fully expanded (5-7 leaves) and mature (8-11 leaves) were 3,161, 1,962 and 774 mg kg-1 DM respectively. However, the HCN content was also found to be reduced after fertilisation with N, P and K (Cadavid *et al.,* 1998).

Gómez (1991) suggested that 100 mg HCN kg^{-1} feed on dry matter basis, as indicated by the Council of the European Community, could be the permissible maximum level. However, in poultry it was reported that broilers could tolerate diets containing 141 mg total cyanide kg^{-1} without any negative effects on growth performance (Panigrahi *et al.*, 1992). In pigs, the inclusion of 15% fresh cassava leaves in the diet had no adverse effects on the performance of growing-finishing pigs (Sarwat *et al*., 1988). However, Ravindran (1990) reported a linear depression in weight gain and feed efficiency when cassava leaf meals were included at up to 30% in diets for growing-finishing pigs. Khieu Borin *et al.* (2005) reported that a significant improvement of daily weight gain of crossbred pigs fed, *ad libitum*, a mixture of cassava leaves and water spinach as compared with those fed cassava leaves alone, due to increase intake and possibly the better amino acid balance of the mixture.

Preservation techniques

Two common techniques, drying and ensiling, have been used with the aim of improving the quality of, and/or preserving feeding materials during periods of excess. Drying and ensiling are commonly used to preserve fish and vegetables, especially in Cambodia, Laos and Vietnam, although these techniques are not widely used to preserve feeding materials for animals. In Cambodia farmers boil cassava leaves for human consumption, as well as for their animals. Boiling has also been used for other feedstuffs, such as the water lily (*Nymphaea odorata*), water spinach (*Ipomoea aquatica)*, taro (*Colocasia esculenta*) and various vegetables and leaves before feeding to pigs (Khieu Borin, personal observation).

Sun-drying

Sun-drying is probably the cheapest method in the tropics for preserving feeding materials for animal use. Small-scale farmers in Cambodia preserve cassava tubers by manually chopping and sun-drying them before selling them to middlemen. Sun-drying is a common practice in the dry season, which is the usual harvest time of the tubers and leaves, due to

strong sunlight, high temperatures, low humidity and availability of space. However, when cassava leaves are harvested in the rainy season sun-drying is difficult, due to the lack of sun and high frequency of rain. Prolonged drying due to bad weather creates favourable conditions for bacteria and fungi, producing mouldy hay and mycotoxins (Oke, 1994). Another problem is that in windy conditions major losses of the dried material will occur if not properly handled. In the small-scale farms, drying is commonly done on the roadsides, or directly on the ground in front of the house, which increases exposure to contaminants, such as dirt and organisms that can cause animal diseases through the passage of animals, humans and vehicles. To reduce such problems, drying beds can be constructed 0.5-1.0 m above ground. However, this will increase the cost of processing, which could discourage some farmers from practicing this technique. However, if the forage can be sun-dried for 2-3 days this is sufficient for the leaves to be milled into a meal before storage. To speed up the drying process, the forage material is first chopped into 3-5 cm lengths, allowing quicker evaporation of moisture and subsequently the release of volatile toxic substances such as HCN. The aim of this preservation process is to reduce the moisture content of the green materials to a minimal level, resulting in the inhibition of plant cell enzymes and microbial activities (McDonald *et al.*, 1995). The moisture content of green biomass usually ranges from 65 to 85%, except in water plants such as duckweed and water spinach, which have higher moisture contents (90-95%). Studies have reported that for a good quality hay, the moisture content should be between 13-14% on dry matter basis. In moist hays (> 200 g H_2O kg^{-1}), microbial respiration, primarily via oxidation of non-structural carbohydrates, can cause negative changes via spontaneous heating within the hay mass (Maillard reactions), increased concentration of field and storage fungi (Roberts, 1995) and potentially also toxic metabolites (Cole and Cox, 1981).

Ensiling

In contrast to sun-drying, plant materials can be ensiled at any time of the year and ensiling is the more practical alternative during the rainy season, when weather conditions preclude sun-drying. However, there are prerequisites for ensiling, for example the availability of materials such as molasses and plastic bags, skilled labour for mixing the chopped forage with molasses, and techniques for excluding air when pressing the material to be ensiled into plastic bags, which ensures anaerobic conditions that prevent the activities of undesirable micro-organisms. Sprague (1974) reported that 90% of the oxygen in the plant material was consumed within 15 minutes and that less than 0.5% remained 30 minutes after sealing the silage container. However, when anaerobic conditions are established anaerobic and facultative organisms start an exponential growth and

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compete for available nutrients, leading to a favourable ensiling process (Pettersson, 1988). A source of readily fermentable carbohydrates is necessary, and in the case of Cambodia 5% sugar palm syrup has been used in the preparation of silage from cassava foliage silage, which is low in fermentable carbohydrates (Chhay Ty *et al.*, 2001). Sugar palm syrup is a product that farmers in palm-growing regions produce during the period of December-May (Khieu Borin *et al.,* 1996; Khieu Borin, 1998). Sugar palm syrup is also available in markets in other areas of the country. Chhay Ty *et al.* (2001) reported that after 56 days, the ensiled cassava leaves were safe for feeding to monogastric animals. Fermentation ensures not only increased shelf life and microbiological safety of a food, but also makes some foods more digestible and in case of cassava fermentation reduces the toxicity of the substrate (Caplice and Fitzgerald, 1999). The overall pH value of cassava foliage decreased from 6.1 to 3.7 between 0 and 14 days of ensiling (Chhay Ty *et al.*, 2001), and a pH of about 4 would normally have preserved the silage materials satisfactorily (Phuc *et al.,* 2000a; An *et al.,* 2004).

The creation of anaerobic conditions discourages the activity of undesirable micro-organisms such as clostridia and enterobacteria. The growth of these organisms produces butyric acid and degrades amino acids, resulting in poor nutritive value of the silage. In order to inhibit the development of these undesirable micro-organisms lactic acid fermentation should be promoted. Lactic acid bacteria are also present on harvested crops, and grow under both aerobic and anaerobic conditions, fermenting sugars in the crop to a mixture of acids, but predominantly lactic acid.

Cassava foliage as feed for livestock

Cassava leaf was reported to have higher concentrations of most essential amino acids compared with soybean meal (Table 1), but is deficient in the sulphur-containing amino acids (Eggum, 1970). There is a significant variation in CP content between cassava varieties, and it also changes with age. Rogers and Milner (1963) reported a range of 17.8 to 34.8% crude protein in 20 cassava cultivars. Ravindran and Ravindran (1988) found a decrease of CP content from 38.1% in very young leaves to 19.7% in mature leaves, and a similar trend for most amino acids, while crude fibre, hemicellulose and cellulose contents increased. Cassava is also a rich source of most minerals, especially Ca and micro-minerals (Ravindran and Ravindran, 1988). In addition, it is important to know the chemical composition of the different parts of the cassava plant, which allows the determination of the parts most suited for different species of animal. Cassava leaves have been reported as having higher protein content than stems and petioles (Khieu Borin *et al.,* 2005), and therefore cassava leaves

will be suitable for monogastric animals such pigs and poultry, while stems plus petioles or whole plant are more suitable for ruminants.

Studies on the use of cassava leaf meal (CLM) for poultry are not well documented, although a few studies suggested a maximum level of 10% of cassava leaf inclusion to avoid negative effects on growth performance (Ravindran et al., 1986; Eruvbetine et al., 2003). Considerably more work has been done on pigs and a study by Phuc et al. (2000a) reported that increasing levels of CLM inclusion in cassava root meal based diets for growing pigs resulted in a linear decrease in the total tract digestibility of OM and CP. Agunbiade et al. (2004) also found a decrease in the total tract digestibility of DM, OM and GE of CLM diets compared to diets including full-fat soybean meal, fish meal and extracted soybean. The digestibility of CP decreased by about 22% when 50% of the dietary CP was supplied by CLM (Phuc et al., 2000b). The CF ranged from 48-290 g kg^{-1} in 28 different sources of CLM, as reported by Ravindran (1993). However, a fibre-free energy source, such as sugar palm or cane juice improved the utilization of fibrous protein feeds (Elliott and Kloren, 1987; Beech et al., 1990), and including oils at between 20 to 80 g kg^{-1} dietary DM increased energy density, improved palatability and supplied vitamins (carotene and tocopherols), and also improved the texture of the rations and DM feed conversion (Ocampo and Lean, 1999). Meanwhile, a study with growing-fattening crossbred pigs given broken rice as a source of energy showed that the pigs had higher intakes of a mixture of cassava leaves and water spinach than cassava leaves alone, resulting in better weight gains (Khieu Borin et al., 2005). The explanation could be that broken rice is palatable and energy-rich and has a low fibre content (AFRIS, 2004), allowing higher intake of the cassava leaves-water spinach mixture. The mixing of cassava leaves with other leaves from leguminous and water plants and providing low fibre sources high in energy such as oil, broken rice, and sugar palm or cane juice, improves the utilization of cassava leaves in animal production, although mixing the leaves with sugar palm or cane juice seems to be difficult in practice in the case of chickens (Naren Toung et al., 1994).

Table 1. Crude protein (g kg^{-1} DM) and indispensable and dispensable amino acid (g 16g^{-1} N) contents of soybean meal and selected forages

	Soybean meal[a]	Leucaena leaves[a]	Dried[b] SPL[‡]	Ensiled[b] SPL	Dried[a] CL[†]	Ensiled[a] CL
Crude protein	482	283	269	234	264	245
Indispensable amino acids						
Arginine	7.5	5.7	5.2	5.0	5.9	5.6
Histidine	2.5	2.0	2.0	1.9	1.9	1.7
Isoleucine	4.2	4.1	4.2	3.6	4.4	4.2
Leucine	7.6	7.9	8.8	9.0	8.0	8.3
Lysine	5.3	5.8	4.1	3.9	5.6	5.4
Methionine	1.1	1.2	1.6	1.2	1.5	1.2
Phenylalanine	5.6	5.6	6.9	7.1	5.7	5.6
Threonine	3.5	4.0	5.2	5.2	4.0	3.9
Tyrosine	3.7	4.3	4.0	3.7	4.0	4.4
Valine	4.7	5.3	5.7	5.4	5.3	5.3
Dispensable amino acids						
Alanine	4.3	5.5	5.4	5.0	5.7	6.4
Aspartic acid	11.2	9.0	11.0	11.2	9.7	9.3
Glutamic acid	17.5	10.0	9.9	10.0	11.2	9.6
Glycine	3.6	4.3	3.5	2.7	4.1	4.1
Proline	4.4	4.0	3.4	3.4	3.6	4.3
Serine	5.3	4.4	4.1	4.7	4.7	3.8
Total amino acids	92.0	83.1	85.0	83.0	85.3	83.1

[†] Sweet potato leaves, [‡] cassava leaves

[a] Phuc *et al.*, 2000a

[b] An *et al.*, 2004

Effect of breed and diet composition on nutrient utilization of fibrous feeds

During evolution, all animal species have adapted to their specific environment and available feeds, in particular with respect to energy and nutrient contents (Jozefiak *et al.*, 2004). It is logical to assume that this evolutionary process had a major influence on the type of digestive system of animals, and on the nutrients that are digested and absorbed. However, several wild animal species with the potential for lean growth have been domesticated and more recently selected to optimize the economic return (Jozefiak *et al.*, 2004), although it is unclear whether improved breeds continue to utilize nutrients from plant materials with similar efficiency to their ancestors or non-improved breeds. Generally plant materials used as feed are available on farm and require lower production inputs. Therefore, studies on the advantages and disadvantages of improved exotic and non-improved local breeds with respect to nutrient utilization under different

environmental conditions are important for small-scale livestock producers in the tropics.

In the case of pigs in Cambodia, crossbreeding, either formal or informal, has had a broad impact in most provinces, although crossbreds are more common close to provincial and district towns. The preference for crossbred pigs is due to market requirements, but at the same time farmers prefer pigs which can still utilize on farm resources effectively.

In poultry, White Pekin ducks (*Anas domesticus*), bred from the wild Mallard (*Anas platyrhynchos*), and broiler chickens (*Gallus domesticus*), developed from the Red Jungle Fowl (*Gallus bankiva* or *Gallus gallus*), have about 3-4 times the adult body mass compared to their ancestors. This body mass is reached within 5-6 weeks under feeding regimes based on high quality concentrates, while the corresponding indigenous breeds require 16-24 weeks under scavenging systems, depending on season and the amount of supplementary feed provided. In the rainy season, there are more insects, worms and green materials available, which are good sources of energy and protein for scavenging chickens and ducks, and therefore growth rates are higher. Indigenous breeds, particularly chickens, are kept by the majority of farmers in Cambodia, with a mean of 13 head of poultry per household, mostly managed under scavenging or semi-scavenging systems. This preference for local breeds is due to market demand, low input and management requirements, ease of raising and their appropriate size for family consumption.

Effect of breed and species on nutrient utilization

Pig breeds in South-East Asia that are considered indigenous or non-improved breeds include the Mong Cai of Vietnam, the Chinese Meishan and the so-called Elephant, Kampot and Kandol pigs found in Cambodia. These three Cambodian indigenous breeds are generally kept by farmers in remote rural areas, as piglets for fattening are readily available and rural consumers still prefer a fatty carcass. Generally they grow slowly, taking 10-12 months to reach maturity, but survive under harsh local conditions, particularly in the dry season, during which time they are released to look for their own feed. The sows have reasonably good reproductive characteristics in terms of litter size and the number of piglets that survive after weaning. However, the present Cambodian urban market requirements and demands in terms of meat quality have encouraged farmers to look for sound, fast growers with low fat content in the carcass, resulting in increasing numbers of crossbred pigs.

Several studies have been carried out to evaluate the capacity of improved and non-improved breeds with respect to protein and nutrient utilization,

although based on these it is not possible to draw any firm conclusions with respect to differences in feed utilization, growth and feed conversion between improved and non-improved breeds. Fevrier *at al.*, (1992) reported that a high level of fibre depressed the apparent digestibility of all dietary components in both Large White and Meishan pigs, while Kemp *et al.* (1991) reported a slight improvement in the digestibility of crude fibre from oat and lucerne meal in Meishan pigs compared to Landrace. According to Ndindana *et al.* (2002), Mukota pigs of Zimbabwe have adapted anatomically to eating poor quality fibrous diets by increasing residence time in the small intestines and fermentative capacity in the hindgut compared to exotic breeds. Another non-improved breed, the Iberian pig, seems to have a lower capacity for protein accretion at feeding intakes close to *ad libitum* than the Chinese Meishan pig (Keryazakis *et al.*, 1993).

There have been a number of investigations of between – species differences in poultry. For example, Jamroz *et al.* (2001) fed diets containing 40% barley to chickens, ducks and geese from 21-42 days of age, and found that chickens digested dietary fibre better than ducks and geese.

Effect of diet on gastrointestinal tract development in poultry

The gastrointestinal tract (GIT) constitutes the first barrier to nutrient metabolism in animals (Cant *et al.*, 1996) and different feed structures may affect the digestive tract of birds and consequently nutrient digestibility (Gabriel *et al.,* 2003). Dietary fibre (DF) influences the transit time, with a reduction in the upper part and an increase in the lower digestive tract, and decreases the digestibility of almost all nutrients and energy (Wenk, 2001). Bach Knudsen (2001) reported that the degradation of DF (dietary fibre) in the large intestine depends on the degree of lignification, solubility and structure of polysaccharides. Peristaltic movements throughout the intestine are affected by gizzard activity, and contraction of gizzard, proventriculus and duodenum are totally coordinated through the electrical potential in the muscular cells (Duke, 1986).

The caeca were reported to play an important role with respect to the microbial degradation of some carbohydrates (Jørgensen *et al.,* 1996), absorption of water (McNab, 1973), microbial synthesis of vitamins (Coates *et al.,* 1968) and degradation of nitrogenous compounds (Goldstein, 1989). Heavier caeca and colon would increase digestive capacity toward dietary fibre and there is evidence that the caeca of chickens contain a higher number of bacteria compared to ducks (Barnes *et al.*, 1972). Studies carried out with ducks, chickens and geese demonstrated that the amount and composition of dietary fibre may influence its

digestibility and the utilization of nutrients, and might even affect the development of the gastrointestinal tract (Jamroz *et al.,* 1992; Jørgensen *et al.,* 1996).

Effect of cassava leaf meal on growth performance

There are few studies comparing improved and non-improved breeds and species, particularly of poultry, with respect to the utilization of CLM and effects on their performance, and the maximum recommended levels of inclusion. Available studies suggest a range of 10-40% of cassava leaf meal inclusion, or 60% of a mixture of cassava products (flour, peel and leaves plus stem) that could be included in poultry and pig diets without having any adverse effect on animal performance. However, the level of inclusion or supplementation of CLM depends on the other dietary ingredients and/or preparation techniques. Montilla (1977) reported that CLM could be included up to 20% in pelleted broiler rations, while Ravindran *et al.* (1986) recommended that up to 15% could substitute for coconut meal. The inclusion of 10% cassava product (50:50 of cassava root and leaf meal) in the diet (Eruvbetine *et al.*, 2003) had no effect on broiler chicken performance in terms of growth, feed conversion, and carcass characteristics. Akinfala and Tewe (2001) included up to 60% of cassava products (40% flour, 10% peels and 10% leaves plus tender stems) in diets for pigs and this resulted in comparable daily weight gain to a maize-based diet. Ravindran *et al.* (1987) reported that replacement of up to 66% of coconut meal by CLM did not have any adverse effects on the performance of growing pigs.

Determination of nutritive value

Several factors, such as growth performance and relative economic advantages, encourage researchers and producers to carefully study potential ingredients in ration formulation. The appropriate quantity and quality of a formulated feed that results in an optimum economic return is the main objective of those involved in animal feed-related businesses. All dietary components, such as protein, amino acids and vitamins and minerals, are important when formulating feeds, particularly for monogastric animals. However the cheapest dietary ingredients are normally those coming from plant materials and agro-byproducts, which generally contain high levels of fibre.

The potential value of a food for supplying a particular nutrient can be determined by chemical analysis. However, the actual value can be arrived at only after making allowances to animals through the process of

digestion, absorption and metabolism (McDonald *et al.*, 1995). Scientists have developed methods to estimate the availability and utilization of nutrients by animals. Two methods, *in vitro,* and indirect and direct *in vivo* methods, have been used to estimate the digestibility of feed materials. Most research workers have used the direct *in vivo* method. Other techniques such as the growth assay and cannulation techniques are also used to assess the nutritive value of feeds.

Total tract digestibility

Total tract digestibility determination is still the most common technique used by researchers due to its simplicity. In pigs, the male is preferred to the female because it is easy to separate faeces and urine. However, it is more complicated with poultry due to the fact that urine and faeces are voided from a single orifice, the cloaca, as a single excretum, and therefore the term used for this mixture is 'excreta'. The separation can be done chemically, considering the fact that most urine nitrogen is in the form of uric acid or that faecal nitrogen is present as true protein (McDonald *et al.,* 1995).

The estimation of the total tract digestibility of nutrients is the value of the difference between the nutrients in the feed consumed and those present in the faeces. Those nutrients which do not appear in the faeces are assumed to be absorbed by the animal and this value is the digestible nutrients, which can either be expressed as a percentage or coefficient. Another technique used to determine digestibility is the addition to the diet of a constituent, which is known to be indigestible. Chromic oxide is the most common indigestible marker used for digestibility studies (McDonald *et al.*, 1995). The quantity used is between 3-5 g of chromic oxide kg^{-1} of feed (Kadim and Moughan, 1997; Jamroz *et al*. 2002).

Summary of materials and methods

Experimental site

All experiments were carried out at the Ecological Farm of the University of Tropical Agriculture Foundation (UTAF), about 15 km south of Phnom Penh City (11.6^{o}N, 104.8^{o}E) in Cambodia, from February 2001 to January 2003. The climate is tropical monsoonal, with two seasons; a dry season (November-April) and a rainy season (May-October).

Forage yields and soil fertility (Paper I, II)

In Paper I, a short-term (ST) variety was investigated while in Paper II a long-term variety (LT) was studied. The defining characteristics of the ST

variety are the light green leaves and red petiole, with the tubers ready for harvest after 6-8 months of growth. The LT variety has a green stem and petiole and purple top leaves, and the tubers are ready for harvest about 12 months after planting. The two varieties are currently popular for tuber production in Cambodia, although the long-term variety was only introduced into the country fairly recently. The middle parts of the stems (20-25 cm long) were planted in a slanted position, two per drill, with a distance between drills of 50 cm. In total 84 stems were planted in each experimental plot of 30 m^2.

In Paper I, cassava was intercropped with *Desmanthus virgatus* (*Dv*) or *Gliricidia sepium* (*Gs*) and no fertilization was applied to any plots, including the control plots. In Paper II, a total of 350 kg N ha^{-1} year^{-1} was applied in weekly dressings of effluent from biodigesters loaded with either pig (PM) or cow (CM) manure. A control treatment (C) received no nitrogen (N). The plants in both experiments were irrigated once daily during establishment (<30 cm height); whereafter the frequency of irrigation was regulated according to rainfall. The first harvest was done when the cassava reached a height of about 150 cm (three months after planting) when all plant material 60 cm above-ground was cut. Thereafter, the plants were harvested every 60 days. Cassava forage was sampled at every harvesting time by manually partitioning the harvested materials into leaves, and stem plus petiole. Only five samples of legumes (leaves and stems) were taken during the experimental period. Soil samples were taken every six months during the experimental period for fertility assessment (biological test with maize) and twice for chemical analysis.

Ensiling and drying (Paper III, IV)

Cassava leaves of ST and LT varieties harvested at 60 days of age were sun-dried for 2-3 days on plastic sheets before being ground in a hammer mill to pass through a 5 mm screen. The cassava leaf meal (CLM) produced was stored in sealed plastic containers prior to the digestibility trial. Cassava leaves of both varieties, also at 60 days of age, were sun-wilted for 1-2 hours before being chopped into 3-5 cm lengths prior to ensiling. The chopped and wilted leaves were well mixed with 5 g sugar palm syrup (75% brix value) diluted in 1:1 water (fresh basis) kg^{-1} wilted leaves. The mixture was packed tightly in polyethylene bags and compacted to exclude air, and then the bags were inserted into polypropylene containers to prevent damage. The ensiling period was 56 days and the cassava leaf silage was used in the animal trials immediately thereafter.

Balance experiment (Paper III)

Four indigenous Mong Cai (MC) (17.2 ± 2.61 kg) and four crossbred (Landrace x Yorkshire) (LxY) (41.5 ± 2.12 kg) castrated male pigs, aged 136 days, were randomly allocated to individual metabolism cages to study the total tract digestibility of dietary components and nitrogen (N) utilization. The experiment had a 2 x 2 x 2 factorial arrangement, with two breeds of pig (MC and LxY), two cassava varieties (ST and LT) and two processing methods (sun-drying and grinding to produce a meal [CLM] and ensiling [CLS]). The two breeds of pig were each assigned to a 4 x 4 Latin Square. The experimental periods were 10 days, comprising 5 days of adaptation to each diet and 5 days of quantitative data and sample collection. The experiment lasted for 40 days (10 days for each period). The cassava leaves, preserved by either sun-drying or ensiling, were fed in amounts corresponding to 30 and 20 g dry matter (DM) kg^{-1} body weight, respectively, with energy supplied by sugar palm syrup and crude palm oil. The diets and residues were weighed twice daily and fresh water was available at all times.

Dry matter intake and digestibility and gastrointestinal tract and organ development (Paper IV)

A total of 192 male birds at 90 days of age was used, of which 48 each were local chickens (LC), broiler chickens (BC), local ducks (LD) and White Pekin ducks (PD). They were randomly allocated to the metabolism cages for the determination of dry matter (DM) digestibility and development of the gastrointestinal tract and associated organs, and given diets in which increasing proportions of the dietary dry matter were replaced by cassava leaf meal (CLM). The experiment was carried out as a 4 x 2 x 2 factorial arrangement, with four treatments (CLM0, CLM7, CLM14 and CLM20, corresponding to 0, 7, 14 and 20% CLM), two breeds (local and exotic) and two species (chickens and ducks). Each treatment was replicated three times with four birds in each replicate. The feed was given *ad libitum* and residues were collected twice daily at 8:00 and 17:00h and fresh water was available all times. Excreta were collected twice daily at 6:00 and 16:00h. Birds were killed four hours after feeding as described by Kadim and Moughan (1997). Two birds from each breed and species within respective treatments were killed by cervical dislocation for the measurements of gastrointestinal tract (GIT) and associated organs.

Chemical analyses (Paper I, II, III, IV)

Samples were dried for 24h at a temperature of 60°C in a locally made forced-air oven. Dried samples were ground to pass through a 1 mm sieve and representative samples were taken and kept in a freezer at temperatures between 4-9°C below zero for subsequent analysis.

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Fresh, dried and ensiled cassava leaves of the ST and LT varieties were analysed for hydrogen cyanide (HCN) content according to AOAC (1990). The DM was determined using microwave radiation according to Undersander *et al.* (1993). Crude protein (CP; N x 6.25; method 988.05), crude fibre (CF; method 978.10), and ash (method 942.05) were analysed by standard procedures (AOAC, 1990). Neutral detergent fibre (NDF) was analysed according to Van Soest *et al.* (1991), with addition of sodium sulphite and EDTA (ethylenediaminetetraacetic acid). Acid detergent fibre (ADF) was analysed according to Goering and Van Soest (1970). The gross energy (GE) content of crude palm oil and sugar palm syrup was determined using an automatic adiabatic bomb calorimeter (CB 100; Gallenkamp & Co. Ltd., England) to calculate the energy supply to meet NRC recommendations (1998). A hand refractometer (Atago N1, Japan) was used to determine daily the soluble sugars content of the sugar palm syrup used.

Statistical analyses

The data were analysed by analysis of variance (ANOVA) using the General Linear Models (GLM) procedures of Minitab version 13.31 statistical software (Minitab, 2000). When the F test was significant at $P < 0.05$, Tukey pair-wise comparisons were performed to determine differences between treatment means.

Summary of results

Forage yield, chemical composition and soil fertility

Cassava-legume intercropping

Cassava-legume intercropping did not affect the total cassava forage or leaf DM yields; however, the highest stems plus petiole DM yields ($P< 0.05$) were obtained when cassava was planted alone (Paper I). The highest total biomass DM yield ($P< 0.001$) and leaf proportion ($P< 0.05$) were obtained when cassava-legume intercropping was practiced. The CP content of the cassava leaves decreased with time ($P< 0.001$), but this was not the case for stems plus petioles. The highest total CP yield was obtained from the cassava-legume intercropping treatment ($P< 0.001$), while there was no effect of intercropping on cassava forage, cassava leaf or cassava stem plus petiole total CP yields. There was a significant harvest occasion effect on total CP yield ($P< 0.01$), and on cassava leaves CP yield ($P< 0.001$). There was a significant increase in carbon, organic matter and Mg ($P<0.01$) contents and also the C/N ratio and pH from the beginning until the end of

the experiment, while N (P<0.05), Ca and K (P<0.001) contents were reduced. All these changes were general and not due to the different treatments.

Cassava fertilization

The highest total forage (P< 0.05) and leaf (P< 0.01) DM yields were obtained in plots using the effluent from the biodigester loaded with pig manure (PM), while stem plus petiole DM yield and leaf proportion were not affected by treatment (Paper II). All plots followed a general yearly pattern of DM production, with peaks around June and a depression beginning in October, with the PM treatment recovering more quickly from this, while it lasted into April in plots fertilized with effluent of the biodigester loaded with cow manure (CM) and the control plots (no fertilizer) (P< 0.05). There was no effluent treatment effect on the CP content of either the leaves or the stems plus petioles. However, there was a significant harvest occasion effect on CP content in the leaves (P< 0.01) but not in the stems plus petioles. Treatment PM gave the highest total forage CP (P< 0.05) and leaf CP (P< 0.01) yields. The soil N balance was negative in all treatments but least so in the CM treatment. A trend of decreasing N, K and Ca and an increase in C/N and pH was observed in all plots from the beginning until the end of the experiment, while the content of Mg increased in all plots.

Ensiling and drying of two cassava leaf varieties

The HCN content of the ST and LT fresh leaves was 545 and 408 mg kg^{-1} DM, respectively, and these values were reduced by 63% and 33% in the ST and LT varieties, respectively, after sun-drying and by 78% and 77%, respectively, after ensiling (Paper III). The pH of the ST and LT leaves after 56 days of ensiling was 4.2 and 4.5, respectively.

Digestibility and N balance experiment

Pigs

There were no cassava variety effects on the DM and nutrient intakes (Paper III). Daily DM, CP and OM (P< 0.001) and NDF, ADF and CF (P< 0.01) intakes were higher in CLM than CLS. There was no effect of cassava variety on the coefficient of total tract apparent digestibility (CTTAD) of DM or dietary components. CLS was higher with respect to CTTAD of DM, CP, OM and CF (P< 0.001) and NDF and ADF (P< 0.01) compared to CLM. The CTTAD of ADF (P< 0.01) and CF (P< 0.001) was higher in Mong Cai (MC) than Landrace x Yorkshire (LxY) pigs. Daily N intake and faecal N (P< 0.001) were higher in CLM than CLS. There was no effect of cassava variety on N utilization. Urinary N was lower in LxY than MC pigs (P< 0.001) and in CLS than in CLM (P< 0.05). The N utilized per unit intake was higher in CLS than in CLM (P< 0.001) and the N retained and N utilized per unit intake and per unit digested were higher in LxY than MC (P< 0.001).

Poultry

Dietary CLM content affected the DM intake, digestible DM intake and CTTAD (Paper IV). There was no significant difference in DM intake between CLM0 and CLM20, although DM intake of CLM20 was higher than for CLM7 (P< 0.05). However, digestible DM intake and CTTAD of DM decreased with increasing dietary CLM inclusion (P< 0.001). DM intake and digestible DM intake expressed in absolute units were higher (P< 0.001) for the local breeds than for the corresponding exotic breeds, and for ducks compared to chickens.

Gastrointestinal tract and organ development of poultry

The dietary CLM content had an effect on most parts of the GIT and associated internal organs (Paper IV). Expressed in absolute units, the weight of caeca, gizzard (P< 0.001) and small intestine (P< 0.05) and the lengths of small intestine and caeca (P< 0.001) increased with increased level of CLM inclusion in the diet. When expressed as kg^{-1} body weight, the length and weight of small intestine and caeca (P< 0.001) and the weight of gizzard (P< 0.01) and colon (P< 0.05) also increased. However liver weight (P< 0.001) decreased with increasing inclusion of CLM, although the effect was not consistent.

The weight and length of the GIT and weight of organs in absolute units were greater (P< 0.001) in the exotic breeds than in the local breeds. However, when expressed as kg^{-1} body weight, the weight of proventriculus, gizzard and pancreas (P< 0.001), colon and liver (P< 0.01) and caeca (P< 0.05) and the length of the small intestine, caeca and colon (P< 0.001) were greater in local than exotic breeds. Similar weights of the small intestine kg^{-1} body weight were recorded in both local and exotic breeds.

Weight of gizzard, pancreas and liver and small intestine length (P< 0.001) in both absolute units and kg^{-1} body weight were greater in ducks than chickens. Small intestine weight kg^{-1} body weight (P< 0.001) and in absolute units (P< 0.01) and proventriculus weight (P< 0.05) kg^{-1} body weight were also higher in ducks than in chickens. The weights of caeca were greater (P< 0.001) in chickens than in ducks, and the length of caeca and colon were similar in both species.

General discussion

Cassava forage yield

The results from these experiments show clearly that the cassava-legumes intercropping increased total biomass DM yield, while cassava forage DM yield was unaffected by this intercropping (Paper I). The effluent from pig manure (PM) loaded into a biodigester produced a higher cassava forage yield than from cow manure (CM) (Paper II) (Table 2).

Intercropping has played an important role in traditional farming systems in many tropical countries. There is a number of possibilities for intercropping practices depending on economic circumstances, for example between cassava and non-legumes, such as cassava/maize, between cassava and legume food crops such as soybean or cowpea/cassava and between cassava and legume fodder trees. The purpose of intercropping cassava and legumes is to benefit from the effect of the legume N_2-fixing systems on cassava production. It is assumed that N_2-fixing systems will satisfy a large proportion of their own N requirements from atmospheric N_2, and that additional fixed N would contribute to soil reserves for the benefit of other crop or forage species (Peoples and Craswell, 1992). A number of studies reported that legumes fix at least 60% of their N requirement from the atmosphere (Nygren *et al.*, 2000; Peoples *et al.*, 1995). Ennin *et al.* (2001) concluded that the land use efficiency increased by 30-52% for cassava/cowpea. However, some studies have reported that the tuber yield of cassava/soybean intercropping is likely to be small under low

availability of soil N and the application of N at planting stage produced higher harvest index (Tsay *et al.,* 1989).

It is uncommon that fertilizer is applied in cassava cultivation, even though it has been observed that tuber yield declines after several consecutive years of cassava cultivation. Putthacharoen *et al.* (1998) reported that cassava removed less N and P but similar amounts of K in the harvested plant parts as compared to maize, sorghum, peanut, mungbean, pineapple and sugarcane. However, it is important to return nutrients when cassava is managed as a perennial crop for forage production in order to produce a high quantity and quality of cassava foliage throughout the year, without soil fertility deterioration. Long-term fertility trials indicate that without adequate K fertilizer, in this case referring to tuber production, cassava yields eventually decline due to K depletion, except in those soils containing large amounts of K-bearing minerals (Howeler, 1991). When cassava is grown continuously with adequate fertilization, and managed in a way that leads to erosion, soil fertility generally declines and cassava yields decrease (Howeler, 1991). The application of N, P and K fertilizer increased root and top biomass yield and reduced root HCN of cassava (Cadavid *et al.*, 1998). Further, fertilization induced production of more vigorous plants, greater soil coverage and protection, increased nutrient recycling from fallen leaves and improved the quality of the planting material (Molina and El-Sharkawy, 1995). Le Ha Chau (1998) reported that the biomass yield per ha was 21% higher when applying effluent from biodigester as compared with raw manure. However, similar yields were found between cow and pig manure, or effluent from a biodigester loaded with cow or pig manure, although a higher protein content in the foliage was found with fertilizer originating from pig manure. Generally, pigs are fed diets with a better nutrient quality, including mineral supplementation, while cattle in SE Asia are mainly fed on fibre-rich, nutrient poor rice straw.

The variation of the protein content of the cassava leaves depends on the stage of maturity, season and fertilization (Ravindran and Ravindran, 1988). In the dry season leaves seem to have a higher protein content, while the lowest CP content is in the rainy season (AFRIS, 2004). The results from our study on cassava-legumes intercropping (Paper I) are in contrast to the study of Nguyen Phuc Tien *et al.* (2003), in which the forage yield of cassava was reduced by 24% when intercropped with *F. macrophylla* as compared with a cassava mono-crop culture. In the current study (Paper I) it was found that *D. virgatus* appeared to be more competitive to cassava in the long term, which is in agreement with

Schroth and Lehmann (1995). The average cassava leaf CP content was about 24%, both after applying effluent and when intercropped with legumes, which is in agreement with Le Ha Chau (1998). A lower value (16.7% CP) was reported by Nguyen Phuc Tien *et al.* (2003) when cassava was intercropped with *F. macrophylla*.

Table 2. Effect of effluent from a biodigester loaded with two kinds of animal manure and intercropping with legumes on cassava forage yield (DM ton ha^{-1} 18 months^{-1}) and CP content (% in DM)

	Short-term variety (ST)			Long-term variety (LT)		
	C	C+Dv[†]	C+Gs[‡]	C	ECM[¥]	EPM[§]
Total biomass	18.0	25.8	25.0	-	-	-
Total cassava forage	18.0	14.7	17.4	27.1	27.8	34.2
Leaf	8.3	8.0	8.9	13.1	13.1	17.0
Stem plus petiole	8.6	6.1	7.8	13.3	13.7	16.1
Leaf CP content	23.7	24.9	24.1	23.8	23.8	25.1
Stem plus petiole CP content	8.4	9.4	9.6	7.4	7.6	8.2
Leaf proportion	49.4	56.7	52.0	50.0	49.5	51.2

[†]Cassava+*Desmanthus vigartus*, [‡]cassava+*Gliricidia sepium*, [¥]effluent of cow manure, [§]effluent of pig manure

Preservation method and its effect on HCN

Besides preserving feeding materials, ensiling and sun-drying were reported in several studies to reduce the HCN content in cassava leaves to a level considered safe for animal feeding (Phuc *et al.*, 2000a; Chhay Ty *et al.*, 2001). In our study, ensiling was more effective in reducing the HCN content in cassava leaves of both ST and LT varieties than sun-drying (Paper III), which is in contrast to the report of Phuc *et al.* (2000a), who found sun-drying more effective. This difference could be due to the preparation technique in our study, in which cassava leaves were chopped and sun-wilted before ensiling. It is known that HCN is released from a cyanogenetic plant after the cellular structure of the plant is disrupted (Conn, 1994). An average of 1,436 mg HCN kg^{-1} DM of fresh cassava leaves eliminated almost 90% with sun-drying and a combination of chopping and 3-day wilting before drying was the most effective, lowering the cyanide content of the final product to about 55 mg kg^{-1} DM (Ravindran *et al.,* 1987). During fermentation, the pH, linamarase activity and total cyanide levels are reduced (Ikediobi and Onyike, 1982), while acid (predominantly lactic acid) levels increase. The pH of the ST and LT

leaves after 56 days of ensiling was 4.2 and 4.5, respectively, which is similar to the value reported by Phuc *et al.* (2000a). A pH of about 4 would normally preserve a crop satisfactorily.

The additive used to prepare the silage in our study was sugar palm syrup, which is a seasonal (November-May) product made by rural farmers in Cambodia, and therefore is readily available and inexpensive for preserving cassava leaves for latter use for their animals. Sugar palm syrup is also available all year round in most local markets. Adding 50 g of sugar palm syrup kg^{-1} cassava leaves after sun-wilting was found to result in a successful fermentation, giving a product of good quality, which is in agreement with Phuc (2000), who added 50 g sugar cane molasses kg^{-1} cassava leaves. However other additives, such as cassava root meal and sweet potato root meal included at a level of 60 g kg^{-1} were also found to produce good quality silage (An and Lindberg, 2004).

Total tract digestibility

Pigs

Effect of processing method on digestibility of nutrients

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. Although plant materials generally have high fibre contents they are commonly used as feeds for monogastrics in the tropics, particularly by small-scale producers, who often rely on forages and water plants to feed their animals. The main concern of using cassava leaves for pigs, besides the HCN, is the high fibre content, which has been reported to reduce nutrient digestibility (Stanogias and Pearce, 1985) in part due to an increase of passage rate of the digesta (Sandoval *et al.,* 1987) resulting in a decrease in the available time for microbial digestion of the fibrous component (Kanengoni *et al.*, 2002). The main site for non-starch polysachharides (fibre) digestion is the large intestine in which the digesta is retained for prolonged periods of time (generally 20-40 hours), allowing prolific bacterial growth (Bach Knudsen and Jørgensen, 2000). However, Lindberg and Andersson (1998) reported that the depression in total tract digestibility of energy was less pronounced with forage fibre inclusion in the diet than with cereal fibre, indicating that pigs have the capacity to utilize forage fibre to a greater extent.

In the present study, the CTTAD was found to be higher for all dietary components in the CLS than the CLM (Paper III) (Table 3). A possible explanation is that some of the fibrous components were degraded by

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hydrolysis during the ensiling process (McDonald *et al.*, 1995). Phuc et al. (1996) substituted soybean meal by cassava leaves (0, 15, 30 and 45% of the dietary protein) and found significant decreases in apparent digestibility of all nutrients when cassava leaf protein replaced 45% of the soybean meal. However, the decline appeared to be less marked for the ensiled compared with the sun-dried leaves and the same authors reported a negative linear relationship between dry matter digestibility and percent cassava leaf protein in the diet, and predicted a digestibility of the leaf dry matter of 76.5% for ensiled leaves and 72.8 for sun-dried leaves. In addition, the utilization of digested N was more efficient in CLS than in CLM, suggesting a minimal degradation of amino acids by microbial activities during the ensiling process (McDonald *et al.,* 1991).

Table 3. Digestibility coefficients of dietary components in foliages in growing pigs

	Dried SPL[†a]	Ensiled SPL[a]	Dried CL[‡b]	Ensiled CL[b]	Dried CL[c]	Ensiled CL[c]
Ileal						
OM	0.84	0.82	0.41	0.42	(-)	(-)
CP	0.74	0.74	0.37	0.37	(-)	(-)
NDF	0.24	0.25	0.26	0.23	(-)	(-)
Total tract						
OM	0.85	0.88	0.54	0.59	0.80	0.84
CP	0.75	0.77	0.45	0.46	0.48	0.71
NDF	0.55	0.56	0.23	0.31	0.70	0.77
ADF	0.32	0.36	0.20	0.21	0.43	0.55
CF	0.61	0.61	0.50	0.59	0.42	0.54

[†] Sweet potato leaves, [‡] cassava leaves, (-) not determined

[a] An *et al.*, 2004

[b] Phuc *et al.*, 2000a

[c] Paper III

Effect of breed on digestibility of nutrients

In Paper III, Mong Cai (MC) pigs, commonly kept by farmers in the rural areas of Vietnam, digested the fibrous components (ADF and CF) of the cassava leaves better than Landrace x Yorkshire (LxY) pigs. This was in agreement with the study by Ndindana *et al.* (2002), who reported that Mukota pigs, a Zimbabwe native breed, digested all nutrients better than the Large White when fed diets based on high maize cob meal, although there was a linear reduction in apparent digestibility of DE, CP, NDF and ADF as maize cob inclusion level increased in both breeds. They suggested that this might be due to a longer and larger caecum-colon in the Mukota

and therefore a higher fermentative capacity to digest relatively large quantities of fibrous materials. In contrast Ly *et al.* (1998) reported that improved pigs (CC21) digested most of the diet components better than Cuban Creole pigs when fed diets high in fibre from plantain foliage meal. The activity of the microbial population present in the hindgut is important for the degradation of dietary fibre (Bach Knudsen and Hansen, 1991). Other studies reported that the inclusion of wheat bran in the diet reduced the total tract apparent digestibility (CTTAD) of all dietary components except nitrogen, in both Alentejano and Large White piglets (Freire *et al.,* 1998). On high fibre diets with the inclusion of alfalfa, oat husk and straw meal, the digestibility of crude fibre was similar between Dutch Landrace and Meishan pigs (Kemp *et al.,* 1991). In a study in Zimbabwe with Large White (LW), Mukota and crossbred (LW x M) pigs, given diets with three levels of inclusion (100-300 g kg^{-1}) of maize cob meal, the Mukota and the crossbred were better able to digest the fibrous components than LW, and in addition, they displayed an ability to retain the protein to the same extent as the LW (Kanengoni *et al.*, 2002).

Poultry

The digestibility and utilization of carbohydrates in poultry is highly dependent on the proportion of starch and dietary fibre in the diet. Yu *et al.* (1998) reported that the dietary fibre source significantly influenced food intake, body weight gain, food conversion, hindgut weight and the activities of }–amylase and cellulose hydrolases in the caeca of geese. The carbohydrate fractions of the dietary fibre cannot be degraded by endogenous enzymes of the gastrointestinal tract, but may be fermented by the microbes of the caeca and colon (Jamroz *et al.,* 2001). However, Svihus and Hetland (2001) reported that 10% of cellulose added to broiler diets resulted in a less-then-expected decrease in feed conversion efficiency however, Hetland *et al.* (2003) concluded that including cellulose in broilers and layers diets improved nutrient utilization. Although there were no significant differences in DM intake (Paper IV) between the CLM0 and CLM20 diets the DM intake of CLM7 and CLM14 was 15 and 12%, respectively, lower than of CLM20 expressed in kg^{-1} body weight. However, the digestible DM intake was about 30% lower in both absolute units and kg^{-1} body weight (Figure 1a) when birds were given the CLM20 diet, and CTTAD was lower on CLM20 compared with the control diet (CLM0). Chen *et al.* (1992) reported that increased fibre content in the diet would probably stimulate food intake and Andersson and Lindberg (1997) reported that high fibre diets could improve the well being of pigs by providing bulk in the diet, thereby increasing satiety. An increase in the quantity of a feed eaten increases rates of passage and therefore results in a reduction in digestibility (McDonald, 1995). Increasing dietary fibre level linearly increased the amount of gut-fill by 0.17 g kg^{-1} body weight for

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each gram of non-starch-polysaccharide (NSP) given as pea fibre, and by 0.10 and 0.09 g kg^{-1} body weight when given as wheat bran, and oat bran, respectively (Jørgensen *et al.*, 1996). It was observed in the present study that the faeces of chickens and ducks became more watery with increasing cassava leaf meal in the diets, particularly on CLM20.

The local breeds had higher feed intake and higher digestible DM intake than exotics, and ducks had higher DM and digestible DM intakes than chickens (Figure 1b). CTTAD of DM was not different between breeds and species, even though the local breeds had greater dimensions of most parts of the GIT than exotics, and ducks have greater small intestine weights than chickens. The results of the present study are in agreement with Jamroz *et al.* (2001), who found no differences in digestibility of DM between chickens and ducks fed diets with high amounts of fibre from barley. However, Barnes *et al.* (1972) reported that the largest concentration of bacteria was found in the caeca of chickens, and the caecal microbes are able to utilize a variety of substrates commonly found in feedstuffs (Jozefiak *et al.*, 2004).
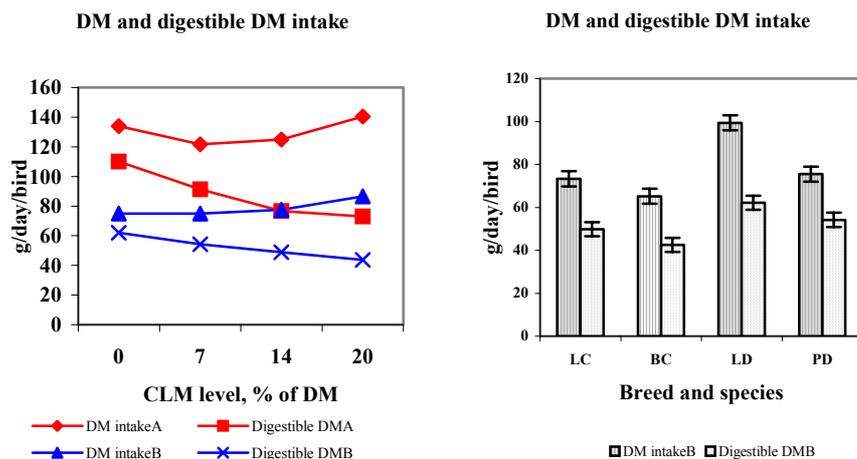
Figure 1a: DM intake and digestible DM of birds given diets with different CLM levels. DM intakeA and digestible DMA expressed in absolute units and DM intakeB and digestible DMB expressed as kg^{-1} bodyweight

Figure 1b: DM intake and digestible DM by breed and species expressed as kg-1 bodyweight. LC [local chicken], BC [broiler chicken], LD [local duck] and PD [White Pekin Duck]

Nutritive value of cassava leaves

The HCN contents in the DM of non-processed leaves of the ST cassava variety were about 34% higher than in the LT variety (Paper III). In contrast, Chhay Ty *et al.* (2001) reported that a long-term 'bitter' variety

had a higher HCN content than a short-term 'sweet' variety. However, the LT variety used in our study (Paper II) was fertilised with effluent from cow or pig manure, which may have reduced the HCN content. Cadavid *et al.* (1998) reported that the HCN concentration decreased and N yield ha^{-1} improved when cassava was fertilized with N, P and K. In this case, nutrient inputs would play an important role in cassava forage production, improving both yields and the nutritive value. However, the chemical composition (DM and N) was similar for the ST and LT varieties used in our studies but was significantly influenced by processing method (Table 4), which resulted in significantly different nutritive values (Paper III). As the CP content of the leaves on a DM basis was twice as high compared to the stem plus petiole, it is important to separate leaves from stem plus petiole when feeding cassava foliage, especially to monogastric animals. The nutritive value not only depends on the bioavailability of nutrients in the dietary ingredients but also depends on the genetic potential of the animals to utilize them. Even on high fibre diets, the LxY pigs were still able to utilize N better than the MC pigs (Paper III), which is in agreement with the study of Kemp *et al.* (1991), who reported that the highly selected Dutch Landrace breed had higher N retention than the indigenous Meishan pig from China.

Table 4. Chemical composition of short and long-term varieties of cassava leaves preserved by sun-drying (CLM) or ensiling (CLS), DM basis

	Variety not stated[†]		Short-term variety[‡]		Long-term variety[‡]	
	CLM	CLS	CLM	CLS	CLM	CLS
DM	-	-	884	495	889	535
CP	264	245	214	208	229	207
OM	913	895	906	894	926	943
Ash	-	-	94	106	74	57
NDF	321	326	320	349	356	353
ADF	221	202	229	241	264	235
CF	164	167	125	144	136	147
HCN	-	-	203	122	273	94

[†] Leaves harvested after root harvest (Phuc *et al.*, 2000a), [‡] leaves harvested at 60 days under fertilization or intercropped with legumes (Paper III).

Several have reported that, due to its bulkiness and high fibre content, the maximum level of inclusion of cassava leaf in diets is between 10-20% for monogastrics (Eruvbetine *et al.*, 2003; Phuc *et al.*, 2000a; Ravindran *et al.*, 1986). However, the maximum level of inclusion also depends on the composition of the other ingredients used in the diet. Adding 3% soybean oil or supplementing with methionine improved broiler performance when given diets containing 20% CLM (Ravindran *et al.*, 1986), and supplementation of DL-methionine in diets containing cassava leaf silage

for Mong Cai pigs appeared to result in an increase in the digestibility of dry matter and organic matter (Ly, 2002).

Effect of cassava leaf meal on gastrointestinal tract and associated organs

High CLM levels in the diet increased the fibre content and had considerable influence on the length and weight of most parts of the GIT and associated organs (Paper IV). The physicochemical properties of the fibre, such as water-binding capacity and viscosity, exert diverse physiological actions along the gastrointestinal tract (Bach Knudsen and Jørgensen, 2000). An increased inclusion of solvent-extracted coconut meal or soybean hulls in diets fed to growing pigs increased the empty weights of the stomach and the colon (Rijnen *et al.,* 2000). At the 20 % level of inclusion of CLM, the increase in the weight of the small intestine was 24.6 and 42.8 % and the increase in length was 17.0 and 28.6 % in absolute units and kg^{-1} body weight, respectively, compared to the control diet in the present study. Jørgensen *et al.* (1996) reported that high intakes of dietary fibre caused a significant expansion of the GIT as well as an increase of the length. According to Jamroz *et al.* (2001) dietary fibre might be expected to influence the peristaltic activity and thereby the length and weight of the intestine. The length of the intestine, and particularly the length and weight of caeca of poultry, increased with increasing fibre level in the diet (Jamroz *et al.*, 1992; Jørgensen *et al.,* 1996) and diets with a high fibre concentration considerably increased gizzard weight (Hetland *et al.*, 2003). In our study (Paper IV), the caeca and gizzard weights in both breeds and species increased when fed diets with increasing CLM levels (Figures 2a, 2b); the caeca weight increased by more than 100% and length by about 35% in birds on the CLM20 diet compared to the CLM0 diet. In most gallinaceous species, the caeca is the major site of fibre digestion, and the digestion proceeds by microbial fermentation and by residual enzymes from the small intestine. The major sources of substrates for caecal fermentation are undigested starch and non-starch-polysaccharides (Yu *et al.*, 1998). However there is a great variation with respect to both dietary composition and animal species (Jamroz *et al.*, 2002). The weight of the gizzard increased by 44 and 57% in absolute units and kg^{-1} body weight, respectively, at the 20% level of inclusion of CLM compared to the control diet. However, these values were similar to those of birds given the CLM7 and CLM14 diets, which implies that only small increases in the level of dietary fibre are needed to stimulate gizzard development. When whole grain was fed to broiler chickens, a higher gizzard weight was found than when they were fed a ground-pelleted diet (Gabriel *et at.*, 2003). This greater development was attributed to an increased frequency of gizzard contractions to cope with the extra grinding

needed to process the large particle size for further digestion in the distal parts of the intestine (Roche, 1981).
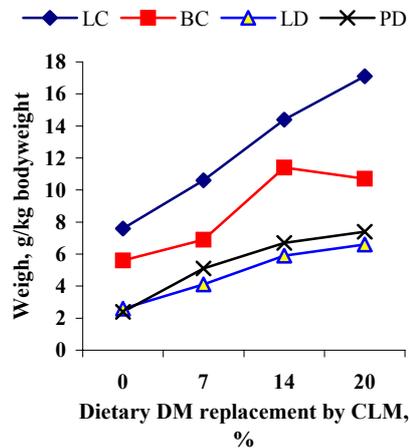

Figure 2a: Caeca development of local and exotic birds with increasing dietary DM replaced by CLM.
LC [local chicken], BC [broiler chicken], LD [local duck] and PD [White Pekin Duck]
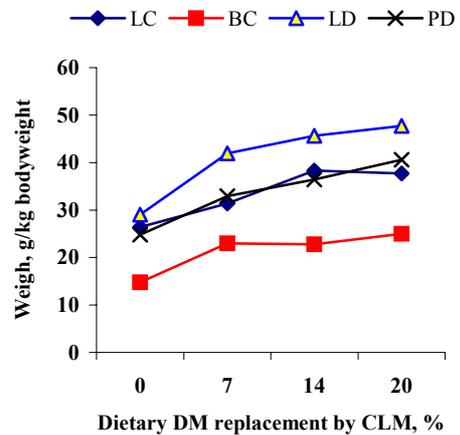
Figure 2b: Gizzard development of local and exotic birds with increasing dietary DM replaced by CLM.

The local breeds had greater GIT and associated organ weights than the exotics expressed as kg^{-1} body weight, although the exotics had almost twice the values of local breeds in absolute units. Watkin *et al.* (2004) reported that the Mallard, an indigenous duck and ancestor of the Pekin duck (PD), has a higher intestinal mass than the PD. Results from our study are in agreement with Jamroz *et al.,* (2001), who reported that ducks had greater gizzard, colon, proventriculus, pancreas and liver weights than chickens, while chickens had greater caeca length.

Conclusions

- Both short- and long-term cassava varieties can be managed for perennial forage production, provided that nutrient inputs are adequate and proper management, including irrigation, is practiced.

- Effluent from a biodigester loaded with pig manure resulted in higher forage yields than when loaded with cow manure, and intercropping

cassava with *Desmanthus virgatus* and *Gliricidia sepium* produced a higher total forage biomass yield.

- Fertilization with 350 kg N ha^{-1} year^{-1} in the form of effluent from biodigesters loaded with pig or cow manure or intercropping with *D. virgatus* or *G. sepium* did not provide sufficient nutrients, particularly N, to maintain soil fertility.

- Ensiling after sun-wilting was more effective in reducing the HCN level in both short- and long-term varieties of cassava. Both short- and long-term varieties were equally well utilized by pigs.

- Mong Cai digested the dietary fibre in cassava leaf based diets better than Large White x Yorkshire, but utilized of dietary nitrogen less efficiently.

- Increasing levels of cassava leaf meal in the diet slightly increased dry matter intake in poultry and each percent unit increase in the CLM content in the diet reduced the dry matter digestibility by around 1.6 percentage units.

- Increasing dietary cassava leaf meal level increased the weight and length of most parts of the gastrointestinal tract and organs of chickens and ducks. Expressed in kg^{-1} body weight, local breeds had higher small intestine, caeca and colon weights than exotic breeds. Ducks had higher weight of organs and most parts of the gastrointestinal tract than chickens. The exception was for caeca and colon weights, which were higher in chickens.

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Acknowledgements

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and who, through many discussions allowed me to learn about and understand their cultures, preparing me for the future.

Last but not least, my wife Khlok Peng Thol, who encouraged me to embark on my PhD studies while she was all-alone during my regular visits to Sweden, and took care of our two lovely children, Khieu Mayuty and Khieu Mavanneh.