DEVELOPMENT OF CASSAVA (*Manihot esculenta* Crantz) LEAF MEAL AS AN ANIMAL FEED

by

Velmurugu Ravindran

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APPROVED:

E. T. Kornegay, Chairman

L. M. Potter

C. P. Fontenot

J. A. Cherry

R. R. Grandhi

L. A. Swiger, Department Head

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Blacksburg, Virginia

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Chapter I

INTRODUCTION

Pig and poultry productions represent one of the quickest means of increasing the supply of animal proteins and alleviating the problem of protein malnutrition in the tropics. The scope for increased production, however, is greatly handicapped by the escalating cost and chronic shortages of traditional feed ingredients, especially those of protein supplements. Nevertheless within the tropical environment, there exists a wide spectrum of agricultural by-products, some of which have substantial nutritive value and are available inexpensively and in large quantities. Since feed cost constitutes the largest single item in all animal production systems, the future of animal agriculture in the tropical regions lies in a better utilization of such lesser-known non-traditional feed sources. Cassava (Manihot esculenta Crantz) leaves, a by-product of cassava root production, is exemplary of this unrealized potential.

Cassava belongs to the family Euphorbiaceae and is an all-season crop of the tropics where it is estimated to provide the staple food for over 500 million people (Lancaster et al., 1982). In addition to its commercially valuable roots, cassava also produces a lush and high protein foliage. The potential yields of cassava leaves as a by-product at root harvesting may amount to as much as 1.85 t dry

matter per hectare (Gomez and Valdivieso, 1984a), corresponding to a crude protein yield of 480 kg per hectare. This valuable feed source is at present returned to the soil as a green manure and, hence, underutilized. The leaves could be easily processed into a protein feed with excellent storage qualities.

Cassava leaves are unique among non-legumes in that the protein content is extremely high. The crude protein content of cassava leaves, depending on variety, stage of maturity, soil fertility and climate, may range from 14.7 to 39.9%. Cassava leaf protein is well balanced with respect to essential amino acids, with the exception of methionine. Although deficient in methionine, it is rich in lysine (Rogers and Milner, 1963). Lysine is the most limiting amino acid in a typical corn-oil meal based swine diet used in the tropics and inclusion of cassava leaf meal offers an added promise in this context. Cassava leaves are also a good source of minerals (Ravindran et al., 1982) and vitamins (Caldwell, 1972; Caldwell and Enoch, 1972; FAO, 1972).

One of the major limitations in utilizing cassava leaf meal as an animal feed is its cyanogenic glucoside content. The cyanide levels in leaves are generally much higher than those in cassava roots. Reported levels of leaf cyanide range from 8.3 to 101.6 mg per 100 g fresh leaves (approximately 415 to 5030 mg per kg dry leaf weight). This fact along with the possibility of their increased utilization as

a novel source of protein underlines the urgent need to determine processing methods that are effective in lowering the cyanide content of cassava leaves.

In common with most root crops, frequent harvesting of cassava leaves would result in adverse effects on root yields. Cassava is cultivated primarily for its roots and it is therefore imperative that leaf harvesting should not greatly reduce root yields. Methods must be developed for harvesting of cassava leaves while maintaining reasonable yields of roots.

The present study seeks to evaluate cassava leaf meal as a potential replacement for coconut oil meal, which is the major protein supplement in swine and poultry diets in many tropical countries. Favorable results could lead to the development of a tropical leaf meal with all the impact of alfalfa meal in temperate climates. The specific objectives of this study were as follows:

1) Evaluation of cassava leaf meal as a source of protein in broiler diets.
2) Evaluation of feeding value, digestibility and protein utilization values of cassava leaf meal for swine.
3) Comparison of different processing methods to lower the cyanide levels in cassava leaf meal, and

4) Study of the effects of different leaf harvesting intervals on the root and leaf yields of a short-age (7-month) cassava variety, MU 22.

Chapter II

REVIEW OF LITERATURE

THE CASSAVA PLANT

Cassava (*Manihot esculenta* Crantz) is the only edible cultivated species in the genus *Manihot* that comprise about 125 species. The genus belongs to the spurge family Euphorbiaceae to which several other economically important species such as para rubber (*Hevea brasiliensis*) and Castor Bean (*Ricinus communis*) also belong.

The cassava plant is a shrubby, woody, short-lived perennial growing to a height of 3 m or more, with erect stems and varying degrees of branching. In some cultivars, branches are produced only from the base of the stem giving an erect bunch growth habit. In others, the branching pattern and branch growth produce widely spreading plants. The stems are slender with leaves clustered towards the apex and with prominent leaf scars lower down. The large, palmate leaves are arranged spirally on the stems and have long petioles. The leaf blade is deeply divided into 5 to 7 obovate-lanceolate lobes of up to 20 cm long. The leaves are usually dark green in color, but various shades of red, yellow and purple pigmentation may also occur in the foliage (Purseglove, 1968; Cobley, 1976).

NUTRIENT COMPOSITION

The protein content of cassava leaves is extremely high for a non-leguminous plant. A wide range of protein contents has been reported (Table 1). The variability is probably related to differences in cultivars (Rogers, 1959; Ramos-Ledon and Popenoe, 1970; Yeoh and Chew, 1976), sampling procedures (Lutaladio *et al.*, 1984), stage of maturity, soil fertility and climate (Moore, 1976).

Variations in leaf protein content between cultivars have been studied by several researchers. Rogers (1959), who analyzed 60 cultivars, found a range of 20.6 to 30.4% crude protein on a dry basis. In a subsequent study involving 20 cultivars, Rogers and Milner (1963) reported an even greater variability of 17.8 to 34.8%. Recent analyses have shown that crude protein content in some cultivars may be as high as 39.4% (Yeoh and Chew, 1976). Almost 85% of the crude protein fraction is true protein (Eggum, 1970).

The possibility of increasing the leaf protein content by breeding has been explored (Nobre *et al.*, 1973). Crossing cassava cultivars with other *Manihot* species resulted in hybrids with leaf protein content well above that of the superior parent. The protein content of the roots was also increased, but this was accompanied by significant increases in the HCN content of the roots. The leaves were not analyzed for HCN levels in this study. However, it is noteworthy that Rogers (1959), analyzing about 60 cultivars,

Table 1. Reported values for crude protein content of cassava leaves (dry basis)

Crude protein content, %	Reference
32.9 - 37.4	Eggum, 1970
15.9 - 21.9	Figueiredo and Rego, 1973
23.9 - 29.8	Gomez and Valdivieso, 1984a
23.2 - 36.0	Lutaladio *et al.*, 1984
14.7	Oyenuga, 1968
19.8 - 31.5	Ramos-Ledon and Popenoe, 1970
20.6 - 30.4	Rogers, 1959
17.8 - 34.8	Rogers and Milner, 1963
16.7 - 19.2	Ross and Enriquez, 1969
26.7 - 39.9	Tupynamba and Vieira, 1979
29.3 - 39.4	Yeoh and Chew, 1976

failed to find any correlation between the HCN and protein contents of cassava leaves.

Little or no attempt has been made so far to select for high leaf protein content, as the primary aim was to produce low cyanide cultivars with high root yields (CIAT, 1973; Sadik et al., 1974). The wide genetic variability that exists between cultivars in leaf protein content is suggestive of the potential response to selection and this appears to be a fruitful area for further research.

Optimization of cultural practices such as fertilizer application may offer another means of increasing the protein content of cassava leaves. Evidence is available to show that leaf protein content is influenced by water availability and soil fertility (Moore, 1976).

Although cassava leaves are rich in protein, other factors such as high crude fiber may limit its nutritive value for non-ruminant animals. Rogers and Milner (1963), analyzing 20 cultivars, reported a range of 4.0 to 15.2%. Immature cassava leaves were evidently used in the above analyses, since values as high as 29.0% have been reported in mature leaves (Table 2). Stage of maturity is the major factor contributing to the variability in fiber content, but environmental and cultivar factors are also implicated (Rogers and Milner, 1963).

Table 2. Proximate composition of alfalfa leaf meal and cassava leaf meal (dry basis)

Nutrient	Alfalfa leaf meal[a]	Cassava leaf meal 1[b].....2[c]
Crude protein	20.0	27.3..20.2
Ether extract	3.5	10.5...6.2
Crude fiber	20.0	4.8..29.0
Ash	10.5	5.7...7.8
Nitrogen-free extract	46.0	51.9..36.8

[a]Allen, 1984.

[b]Rogers and Milner, 1963 (immature leaves).

[c]Ravindran et al., 1982 (mature leaves).

Table 3. Mineral composition of alfalfa leaf meal and cassava leaf meal (dry basis)

Mineral	Alfalfa leaf meal[a]	Cassava leaf meal[b]
Calcium, %	1.50	1.35
Magnesium, %	.32	.42
Iron, ppm	281	859
Copper, ppm	9	12
Manganese, ppm	34	252
Zinc, ppm	19	249
Sodium, ppm	800	177
Potassium, %	2.50	1.28
Phosphorus, %	.27	.45

[a]Allen, 1984.

[b]Ravindran et al., 1982.

Cassava leaf meal contains about 8 to 9% ether extractable fraction, but only a third of this is lipids. Khor and Tan (1981) reported the lipid content of cassava leaves to be 3.0%. The lipids consist of 22.4% nonpolar lipids, 25.1% glycolipids and 48.2% phospholipids. All lipids, except steryl esters, were rich in polyunsaturated fatty acids.

Cassava leaves are good sources of minerals. They are particularly rich in calcium, magnesium, iron, manganese and zinc (Table 3). Cassava leaves are also rich in ascorbic acid (Caldwell, 1972; Watson, 1976) and vitamin A (Abbes, 1956; FAO, 1972), and contain significant amounts of riboflavin (Caldwell and Enoch, 1972). But considerable losses of vitamins, particularly of ascorbic acid, occurs during processing (Caldwell and Gim-Sai, 1973; Watson, 1976) and storage (Caldwell and Gim-Sai, 1973).

AMINO ACID COMPOSITION

Rogers and Milner (1963) were probably the first to report the amino acid content of cassava leaves. They analyzed the leaves of 20 Jamaican and Brazilian cultivars obtained from ten-month old healthy cassava plants and found that cassava leaf protein was deficient in methionine, but rich in lysine. Later studies (Eggum, 1970; Otul, 1973; Yeoh and Chew, 1976) on cassava leaf protein showed similar amino acid patterns, although considerable variability was observed for individual amino acids.

Table 4. Amino acid composition of alfalfa leaf meal and cassava leaf meal (g/16 g N)

Anubi Acid	Alfalfa Leaf Meal[a]	Cassava Leaf Meal		
		1[b]	2[c]	3[d]
Aspartic acid	-	10.14	9.63	9.77
Threonine	4.40	4.92	4.73	4.39
Serine	-	5.16	4.60	4.55
Glutamic acid	-	10.22	10.12	12.32
Proline	-	4.64	5.40	-
Glycine	5.00	5.39	5.32	4.86
Alanine	-	5.98	6.19	5.73
Valine	5.95	5.73	5.58	5.56
Cysteine	1.15	1.37	1.04	1.40
Methionine	1.65	1.65	1.71	1.86
Isoleucine	4.90	5.01	4.84	4.50
Leucine	7.50	8.89	8.85	8.19
Tyrosine	-	4.18	3.93	4.04
Phenylalnine	5.20	5.82	5.53	5.42
Lysine	4.35	7.20	6.33	5.87
Histidine	2.10	2.23	2.56	2.30
Arginine	4.90	5.28	6.12	5.34

[a]Allen, 1984
[b]Roger and Milner, 1963
[c]Roger and Milner, 1963
[d]Eggum, 1970

The variation in amino acid content of the leaves may be attributed to the stage of leaf maturity, sampling procedures, analytical methods and ecological conditions (Otul, 1973). Otul (1974) reported that variation among cultivars grown under identical conditions was insignificant. Similar observations were made by Yeoh and Chew (1976) suggesting little, if any, genotypic variation with respect to amino acid content. On the contrary, the data of Rogers and Milner (1963) show that there is a large variability in the amounts of individual amino acids between cultivars. However, these authors did not mention whether these cultivars were grown under similar conditions and sampled in an identical fashion.

The amino acid compositions of alfalfa leaf meal and cassava leaf meal are presented in Table 4. It can be seen that the essential amino acid profile of cassava leaf meal is either similar or superior to that of alfalfa meal.

PROTEIN QUALITY

The digestibility of cassava leaf protein has been investigated by Luyken *et al*. (1961) who found the digestibility to be 81% in young leaves and 67% in older leaves. However, the net protein utilization was low, 32% in young leaves and 39% in older leaves. Net protein utilization was increased to 61% by the addition of the most limiting amino acid, methionine.

Table 5. Percent amino acid availability in boiled cassava leaves[a]

Amino acid	Availability, %
Aspartic acid	72.3
Threonine	62.2
Serine	84.0
Glutamic acid	64.4
Glycine	58.7
Alanine	64.3
Valine	55.2
Cysteine	75.3
Methionine	59.2
Isoleucine	55.2
Leucine	61.0
Tyrosine	61.5
Phenylalanine	62.5
Lysine	72.8
Histidine	71.7
Arginine	65.8
Tryptophan	66.1

[a]Eggum, 1970.

Eggum (1970), using rat bioassays, studied the nutritional availability of individual amino acids in cassava leaves. The availability of amino aids varied widely ranging from 55% for valine and isoleucine to 84% for serine (Table 5). Only 59% of the methionine was biologically available, resulting in a low biological value of 49 to 57%. Supplementation with methionine improved the biological value to 80%.

Oke (1978) attributed the low protein digestibility values to the higher fiber content of cassava leaf meal. The presence of condensed tannins in cassava leaves has been recently reported (Reed et al., 1982) and this may be another factor responsible for the low protein utilization in cassava leaves. Tannins are known to lower the protein digestibility and amino acid availability by forming indigestible tannin-protein complexes with dietary proteins and/or by inhibiting digestive enzymes (Kumar and Singh, 1984).

PRODUCTIVITY OF CASSAVA LEAVES

The potential yield of cassava leaves varies considerably, depending on cultivars, age of plant, plant density, soil fertility, harvesting frequency and climate (Ahmad, 1973; Dahniya et al., 1981; Gomez and Valdivieso, 1984a; Montaldo and Montilla, 1976). Ahmad (1973), investigating the leaf dry matter productivity of two 12-month cultivars,

reported yields of up to 7490 kg/ha. In his study, five leaf harvests were made at intervals of six weeks starting from three months after planting. Leaf harvesting, however, lowered the root crop to almost one-half of the normal yield. Normanha (1962) harvested 9000 kg dry matter/ha in two harvests over a two-year period and obtained within 30% of the normal yield of roots.

Too frequent leaf harvests will not only result in lowered root yield (Dahniya et al., 1981), but also will make the leaves susceptible to cassava mosaic disease in endemic areas (Lutaldio and Ezumah, 1981). Methods need be developed to harvest cassava leaves while maintaining reasonable levels of root production. Dahniya et al. (1981) recommended a harvesting frequency of two to three months, starting from 4 months, for the best all round yields in 12-month cultivars. However, considerable variation exists among cultivars in their tolerance to leaf harvesting (Ahmad, 1973; Dahniya et al., 1981) and should be taken into consideration.

The leaf dry matter yields will be lower, if cassava leaves are obtained as a by-product at root harvest. Gomez and Valvidieso (1984a), evaluating two 12-month cultivars, reported the leaf dry matter yields at root maturity to be only 1170-1840 kg/ha.

When the cultivation of cassava is exclusively aimed towards leaf production, the plant density could be

increased and the harvesting frequency could be more intense. Under such conditions, annual leaf dry matter yields of over 34,000 kg/ha can be obtained. This represents a possible production of more than 6000 kg of protein per hectare per year (Montilla, 1976). Whether the aim should be root production, leaf production or an all round production of both would depend inter alia upon the relative prices of cassava roots, cassava leaf meal and traditional feedstuffs.

CYANOGENIC GLUCOSIDES

The toxic properties of cassava roots and leaves are generally associated with the free HCN that is liberated when their cyanogenic glucosides, namely linamarin and lotaustralin, are hydrolyzed. The former accounts for 96% of the total glucosides, and the latter, 4% (Conn, 1973). The release of free HCN is brought about by the action of either the endogenous enzyme linamarase in damaged plant tissues or β-glucosidases within the digestive tract of animals. The linamarase and glucosides do not come into contact in healthy cassava leaves, but contact occurs when the tissues are mechanically damaged or when the physiological integrity is lost as in the case of wilted leaves.

The cyanide content of cassava leaves has been determined by several workers (Table 6). The normal range of cyanide content is from 20 to 80 mg HCN per 100 g fresh leaf

weight, but occasional samples as low as 8 mg/100 g (Wood, 1965) or over 186 mg/100 g (Gondwe, 1974) have been reported. On a dry basis (assuming 25% dry matter in fresh leaves), the normal range of HCN content would correspond to 800 to 3200 mg/kg. These levels are substantially higher than the normal range of HCN reported for fresh cassava roots (Coursey, 1973). Yeoh and Oh (1979) found the leaf HCN levels to be six times higher than those of roots.

The wide variations observed in leaf cyanide levels may be attributed to genetic, physiological, edaphic and climatic differences, but have been exaggerated by problems associated with methodology of cyanide assay (Cooke and Coursey, 1981).

That there is a considerable genetic component in the variation of leaf cyanide levels is now well established (Chew, 1972; Gondwe, 1974; Yeoh and Oh, 1979). Chew (1972) reported a range of 17.4 to 62.2 mg HCN/100 g fresh weight in 18 cultivars grown under identical conditions. In a similar study involving 31 cultivars, Yeoh and Oh (1979) obtained a range of 12.5 to 85.4 mg HCN/100 g fresh weight. Leaf has been postulated as the site of glucoside synthesis (Nartey, 1968). The rate of glucoside synthesis is somewhat equal in all cultivars, but differences exist in the rate of degradation resulting in genetic variability among cultivars (De Bruijn, 1973). The cultivars with low leaf cyanide

Table 6. Reported levels of cyanide in fresh cassava leaves

Cyanide content (mg/100 g fresh weight)	References
46.6 - 63.0	Bassir and Fafunso, 1976
40.0 - 100.0	Charavanapavan, 1944
17.4 - 62.2	Chew, 1972
40.0 - 101.6	Gondwe, 1974
19.1 - 87.7	Joachim and Panditteseker e, 1944
20.5 - 149.7	Lutaladio et al., 1984
32.0 - 78.0	Sinha and Nair, 1967
8.3 - 16.2	Wood, 1965
12.5 - 85.4	Yeoh and Oh, 1979

levels evidently have a higher rate of degradation of glucoside than those of high leaf HCN-cultivars.

Stage of leaf maturity is another important factor causing variations in cyanide content. As in other cyanogenic plants, the glucoside concentration in cassava leaves decreases with age (Gondwe, 1974; Joachim and Panditteseker, 1944; Lutaladio et al., 1984; Obregon, 1968; Williams, 1979). In young, expanding leaves, the cyanide level in the petioles is higher than that in the leaf blades, whereas the reverse is true in the older leaves (De Bruijn, 1973).

Cyanide levels in the leaves are also influenced by the nutritional status of the plant. De Bruijn (1973) reported that leaf cyanide levels were increased by fertilizer nitrogen, whereas potassium and farmyard manure had the opposite effect. The effects of phosphate, calcium and magnesium were insignificant. Nitrogen and potassium are postulated to exert their influence by changing the amino acid content of leaves, particularly valine and isoleucine, which may be the precursors of linamarin (Conn, 1973). Sinha (1969) suggested that a change in the method of fertilizer nitrogen application, from soil to foliar, may check enhanced cyanogenesis caused by fertilizer nitrogen.

Leaves produced during prolonged drought were reported to contain high amounts of cyanide (De Bruijn, 1973). Short periods of water deficit generally had little effect as the

plants adapted by abscissing some leaves. Shading of young plants caused an increase in the leaf cyanide levels (De Bruijn, 1973a). Some evidence exists for a diurnal rhythm in cyanogenesis in cassava (Nartey, 1981). Goats and sheep browse cassava leaves at certain times of the day without any signs of toxicity, while ingestion during certain other times leads to poisoning and death.

Leaf cyanide levels have been used by several workers (CIAT, 1973; Sadik et al., 1974) in cassava screening programs to select cultivars with low root cyanide contents for breeding experiments. This procedure is convenient because leaves are more easily accessible than roots. Available data, however, suggest that no significant relationship exists between the cyanide contents in leaves and roots (Sinha, 1969; Yeoh and Oh, 1979).

CYANIDE TOXICITY

The animal body detoxifies cyanide via several pathways, but primarily by reaction with thiosulfate to form thiocyanate (Montgomery, 1969). When cyanide is converted to thiocyanate, a 200-fold reduction in toxicity occurs. This reaction is probably the detoxification mechanism by which the body copes with small amounts of cyanide consumed in food. Liver is the chief site of detoxification, where the enzyme rhodanese through combination with sulfur sources reacts with cyanide to form thiocyanate (Oke, 1973a).

Thiocyanate, is a potent goitrogen and has been implicated in the etiology of goiter in animals (Langer, 1966; Sihombing et al., 1971) and humans (Ekpechi et al., 1966).

Hydrocyanic acid is one of the most potent respiratory poisons known to man. In animals, while acute cases of cyanide toxicity usually result in sudden death, less severe cases may lead to gastrointestinal disorders and growth depression (Hill, 1973). Acute poisoning as a result of consuming cassava roots by man or domestic animals is not common, but by no means unknown. Despite its high content of HCN, documented cases of poisining due to the intoxication of cassava leaves are extremely rare. Perhaps the high content of HCN acts as a deterrent against excess consumption of cassava leaves by grazing animals (Swain, 1977).

In contrast to acute poisoning, relatively little is known about the chronic effects which results from the continuous ingestion of small amounts of cyanide. Perhaps this is because most reports of chronic toxicity are field or clinical cases where experimental controls were not used. Moreover, in such case, it is difficult to ascribe the effects specifically to cyanide, because a complex of factors are usually involved.

It is not usually recognized that low level chronic intoxication from cyanide does exist and can be quite incapacitating. Evidence accumulated during the last two decades is consistent with the hypothesis that long term

consumption of cassava containing low levels of HCN is a probable cause of tropical ataxic neuropathy and goiter in humans (Ekpechi et al., 1966; Osuntokun, 1973; Thilly et al., 1972; Oke, 1980). Other specific diseases implicated include Leber's optical atrophy, retrobulbar neuritis, cretinism, tobacco amblyopia and pernicious anemia (Way, 1981). Whether a parallel situation occurs with domestic animals is not known. However, it has been shown that continuous low level dosage of rats with potassium cyanide will produce lesions in the central nervous system (Smith et al., 1963). Studies have also demonstrated that thiocyanate formed during the detoxification of ingested cyanide interferes with the utilization of iodine for thyroxin production in pigs (Sihombing et al., 1971) and rats (Langer, 1966).

Until recently, toxicity of cassava products was assumed to be associated with free HCN, 50 to 60 mg of which constitutes the lethal dose for an adult human (Bolhuis, 1954). The cyanogenic glucoside per se was thought to be of little consequence to animals, if endogenous linamarase had been inactivated (Montgomery, 1969). The hydrolysis of cyanogenic glucosides in the digestive tract of rats is now established (Spatz, 1968) and oral doses of linamarin, in the absence of linamarase, have been shown to produce physiological and biochemical effects in rats (Philbrick et al., 1977). Barrett et al. (1977) reported that linamarin

administered to rats resulted in toxicity symptoms similar to those observed with potassium cyanide.

The relative toxicities of free HCN and cyanogenic glucoside (bound cyanide), however, remain unclear. Studies of Sitompul (1977) with rats suggest that linamarin may be less toxic than free cyanide.

OTHER ANTI-NUTRITIONAL FACTORS

While the toxicity of cassava appears to arise from the presence of cyanogenic glucosides, it is noteworthy that several other potentially anti-nutritional factors have also been recorded in cassava leaves. An extremely wide range of oxalic acid levels, 99 to 3000 mg per 100 g fresh leaf weight, have been reported in the literature (Lancaster and Brooks, 1983). Ingestion of oxalate is known to interfere with calcium availability and utilization, and long term intake of small amounts of oxalate may lead to renal damage owing to the formation of urinary calculi (Blood and Henderson, 1974).

The presence of condensed tannins in cassava leaves may represent grounds for some concern. Condensed tannins can form unavailable complexes with protein, thus lowering the biological value of cassava leaf protein and increasing the amino acid requirements of animals fed cassava leaves (Reed et al., 1982).

The release of hydrogen sulfide from cassava leaves upon heating has been reported and suggested as a potential hazard in the consumption of cassava leaves (Ugochukwu and Osisiogu, 1977). The source of sulfur is obscure, especially since cassava leaves are low in sulfur-containing amino acids.

Several workers have referred to the presence of a toxic protein, toxalbumin, in cassava (Clark, 1936; Johnson and Raymond, 1965; Turnock, 1937), but the part of the plant from which it was isolated was not specified. Compounds of this class are typical of the family euphorbiaceae, to which cassava belong. Clark (1936) specifically mentioned post-mortem indications suggestive of toxalbumin in cases of cassava poisoning. The toxalbumin content of the leaves merits further study. The possible occurrence of yet unidentified potentially toxic substance(s) in cassava leaves should also not be ignored.

CASSAVA LEAVES AS HUMAN FOOD

While cassava leaves could play a significant role in improving the nutritional status of tropical population (Terra, 1964), consumption of leaves is not as widespread as that of roots. Except for central and west African regions where considerable quantities of cassava leaves are consumed daily, in many areas the leaves are not used at all or only

when other preferred leafy vegetables are unavailable (Jones, 1959).

The potential value of cassava leaves as human food, including traditional consumption patterns and processing techniques, had been recently reviewed (Lancaster and Brooks, 1983). The most common way of preparing cassava leaves for consumption is by pounding or chopping followed by several hours of boiling. The final product may be a sauce (thick paste) or a stew (thin soup). In Sri Lanka, the immature leaves are chopped, washed in water, mixed with ingredients such as coconut scrapings, onions, chillies and spices and fried in oil. The resulting product, called mallum, has a pleasant bitter taste.

In some areas, the cassava leaves are sun-dried before pounding and cooking (Tallantire and Goode, 1975). Dried cassava leaves are also stored for later use, sometimes ground into a flour (Velcich, 1963).

DETOXIFICATION OF CASSAVA LEAVES

Detoxification of cassava leaves may be partially accomplished by heating or boiling to inactivate linamanase or to drive off free HCN. However, this procedure would be insufficient to remove the bound cyanide in the form of linamarin. In light of the recent reports that linamarin per se can exert toxic effects (Barret et al., 1977; Philbrick et al., 1977), total detoxification may only be

achieved by complete autolysis of linamarin followed by removal of free HCN (Lancaster and Brooks, 1983). Because of the generally high linamarase levels in the leaves, the autolysis of linamarin will be more effective if the substrate and enzyme are brought into contact by processing techniques such as chopping or crushing of leaves. De Bruijn (1973) reported the linamarase activity of young expanding leaves to be almost 100 times that of peeled roots.

Although some earlier reports stated that simple boiling or cooking is sufficient to remove cyanide completely (Raymond et al., 1941), a review of literature has shown that residual quantities of cyanide always persist (Lancaster and Brooks, 1973). Thus, the potential danger of chronic cyanide toxicity associated with prolonged consumption of cassava leaves certainly exists and must be kept in perspective.

Bassir and Fafunso (1976) studied the effects of precooking on the cyanide content of cassava leaves. Washing the crushed leaves after boiling in water for 15 minutes was found to be the most effective means of lowering HCN levels. Almost 85% of the original level of 47 to 63 mg HCN per 100 g fresh leaves was eliminated. Mere bruising of the leaves in water resulted in a loss of about 77%, whereas boiling the leaves for 15 minutes removed only 57% of the HCN. Soaking the leaves in water lowered the HCN content only by

15%. This study along with others (De Bruijn, 1973; Williams, 1979) suggest that the leaves must be chopped or crushed prior to cooking to lower the HCN content to safer levels.

Limited published information exists on the HCN levels in dehydrated cassava leaves prepared for use in animal feeding. Obregon (1968) stated that sun drying eliminates most, if not all, of the cyanide in cassava leaves. Siriwardene and Ranaweera (1974) resorted to bruising and wilting of leaves to lower the cyanide levels. When oven-dried, the final products had a cyanide content of 49 mg/kg dry matter. Rajaguru et al. (1979) found that wilting in the shade for three days followed by oven-drying reduced the cyanide level in the leaves to 33 mg/kg dry matter.

FEEDING VALUE FOR POULTRY

At low levels of inclusion, the feeding value of cassava leaf meal for poultry is similar to that of dehydrated alfalfa meal. Ravindran et al (1983b) compared the performance of quails fed iso-nitrogenous diets containing 0, 2.5, 5.0, 7.5 and 10.0% levels of either cassava leaf meal or dehydrated alfalfa meal. Gains were not significantly influenced by the level of leaf meal inclusion, but feed intake and feed/gain were linearly increased as the leaf meals were incorporated above 5% level. The performances of birds fed cassava leaf meal and dehydrated alfalfa meal were

similar. On the contrary, Ross and Enriquez (1969) reported that the gains and feed efficiency of white leghorn cockerels fed diets containing above 5% cassava leaf meal were poorer than those fed corresponding levels of alfalfa meal. Cassava leaf meal used in their study contained 554 mg HCN/kg and may have contributed to the poor performance.

Ross and Enriquez (1969), in a series of trials, investigated the possible use of cassava leaf meal in chick rations. Cassava leaf meal, prepared by oven-drying the leaf materials overnight at 50°C, was substituted for corn and soybean meal. Progressive depression in performance was observed with increasing levels of cassava leaf meal. Supplementation of diets containing 20% cassava leaf meal with methionine and oil resulted in performance comparable to the control. It was suggested that methionine is the first limiting factor and energy the second to the high level usage of CLM. Addition of sodium thiosulfate, a known sulfur donor, to the 20% cassava leaf meal-ration significantly improved the chick performance, indicating that the beneficial effect of supplemental methionine was partly through the provision of sulfur for the detoxification of cyanide which was calculated to be 111 mg/kg in the cassava leaf meal-ration.

Siriwardene and Ranaweera (1974) evaluated cassava leaf meal as a substitute for coconut oil meal in broiler rations and concluded that up to 10% level can be used with

satisfactory results. Cassava leaf meal used in their study was prepared using a combination of bruising, wilting and drying, and contained 44 mg HCN/kg.

Montilla et al. (1976) reported depressions in gains and feed efficiency when cassava leaf meal was included at 10, 20 or 30% levels in broiler rations. Cassava leaf meal was used to replace parts of the cottonseed meal, sesame oil meal and corn in the basal ration. The depressing effects due to the inclusion of cassava leaf meal were partly overcome by pelleting.

Wyllie and Chamanga (1979) found cassava leaf meal to be a superior substitute for cottonseed meal in broiler rations. Replacement of cottonseed meal with 5 and 10% cassava leaf meal resulted in significant improvements in gains. However, when cassava leaf meal was substituted for sesame oil meal and sunflower oil meal the performance of broilers was poorer.

While the use of cassava leaf meal as a major source of protein for growing chickens is impressive, evidence suggests that cassava leaf meal may be better utilized by older birds. Cassava leaf meal can be included in layer rations up to 30% level without any adverse effect on egg production (Rajaguru, A. S. B., personal communication). The ability of layers to better utilize cassava leaf meal may be related to their higher tolerance to cyanide. Jalaludin and Yin (1972) found that layers were tolerant to cyanide levels as

high as 135 ppm. The suggested tolerance of HCN by growing chickens is only 10 to 15 mg/kg (Rajaguru, 1975).

Hutagalung et al. (1974) determined the metabolizable energy value of cassava leaf meal for poultry to be 1.59 kcal/g, whereas a higher value of 1.92 Kcal/g was reported by Siriwardene and Ranaweera (1974). The inconsistency in energy values is probably associated with differences in composition of the samples. Ravindran et al. (1983a) showed that the protein in cassava leaf meal was 63% digestible by poultry, but the dry matter digestiblity was only 35%.

The literature on the use of cassava leaf meal as a poultry feed is thus not only limited, but also inconclusive. The variability in the cyanide content of cassava leaf meal appears to be the major factor causing the conflicting results. Proper detoxification of cassava leaf meal and provision of sulfur sources, particularly of methionine, are essential to alleviate the problem of cyanide. High fiber content, along with the resultant low nutrient density and low nutrient digestibility, and bulkiness are other factors limiting the use of cassava leaf meal-based rations and, in this context, pelleting may prove beneficial.

FEEDING VALUE FOR SWINE

Limited published information exist regarding the use of cassava leaf meal in swine feeding. Mahendranathan

(1971) fed swine with fresh cassava leaves ad libitum, in addition to restricted amounts of a basal diet, and observed depressed performance. Though the animals consumed an average of 1.8 kg fresh cassava leaves daily, no clinical signs of cyanide poisoning were observed. This result may reflect the generally high tolerance of swine to cyanide.

Lee and Hutagalung (1972) found that inclusion of 10 and 20% cassava leaves reduced palatability and lowered gain and feed efficiency in growing-finishing swine. The depressing effects were evidently due to the high cyanide level in the fresh leaves, since supplemental methionine and thiosulfate proved effective in improving the performance. Addition of molasses and palm oil also tended to improve the gain and feed efficiency, indicating that energy may be the second limiting factor.

In preliminary studies involving 40 animals, Rajaguru et al (1979) substituted cassava leaf meal for coconut oil meal at levels up to 30% in diets for growing-finishing swine and observed no adverse effects on performance. During the grower phase, inclusion of cassava leaf meal significantly improved the performance which was attributed to its high lysine content.

There is an unexplored potential for the use of cassava leaf meal as the major source of protein in sow rations. Available evidence suggests that the energy in fibrous

feedstuffs are well utilized by sows (Allee, 1977; Boyd et al., 1976; Danielson and Noonan, 1975; Pollman et al., 1979).

CASSAVA LEAF PROTEIN CONCENTRATES

Although the potential for protein production from cassava leaves is enormous, other factors such as high fiber and cyanide content limit its use as a major source of protein for non-ruminants. These limitations could be largely overcome if the protein is separated from the fiber and a protein concentrate prepared by a juice extraction step and steam coagulation (Pirie, 1971).

Leaf protein concentrate (LPC) has been prepared from cassava leaves by a number of researchers (Byers, 1961; Fafunso and Oke, 1976; Fafunso et al., 1976; Nandakumaran et al., 1978; Singh, 1964; Tupynamba and Vieira, 1979). Byers (1961), who studied leaves from 60 tropical species in Ghana, found that protein was poorly extracted from the cassava leaf. Similarly, Singh (1964) in India and Kling et al., (1976) in Brazil reported poor extractions in cassava leaves. Because of the poor extraction, Telek and Martin (1983) are of the opinion that cassava has no potential for LPC production. In contrast, studies from Nigeria show that cassava leaf protein has a reasonably good extractability. An extractability of 70% was obtained by Oke (1973b). Fafunso and Oke (1976) extracted leaf protein from 15 culti-

vars of cassava. All cultivars had a similar extractability, with an average of 58.7%. Tupynamba and Vieira (1979) reported a variability in extraction ranging from 20.2 to 64.7%. The inconsistency in extractibility of cassava leaf protein may be related to differences in extraction techniques. Clearly further research is needed to develop refined technology to improve the protein extraction from cassava leaves.

Tupynamba and Vieira (1979) reported that cassava LPC, on an average, contained 46.1% crude protein, 3.5% crude fiber, 2.0% ash, 19.8% ether extracts and 28.6% nitrogen-free extracts. Over 75% of the protein was true protein and the papain digestibility of cassava LPC ranged from 52.8 to 60.9% (Fafunso and Oke, 1976). The amino acid content of cassava LPC is superior to that of oil seeds and comparable to that of animal protein supplements, with the exception of sulfur-containing amino acids (Oke, 1984; Tupynamba and Vieira, 1979).

The production of LPC also overcomes the problem of cyanide in cassava leaves. Only a low level of cyanide remains in the protein concentrate (Balasundaram et al., 1976; Fafunso et al., 1976; Oke, 1973b). Almost 75% of the cyanide in fresh leaves is lost during pulping and pressing. The remaining cyanide in the wet-leaf fraction is further reduced by drying, particularly by freeze drying or

oven drying rather than sun drying (Fafunso et al., 1976).

Despite its low cyanide content and good amino acid profile, nutritional evaluation of cassava LPC has shown poor animal performance (Cheeke et al., 1980). Tupynamba and Vieira (1979) reported that rats fed on diets containing cassava LPC lost weight, resulting in a negative PER value. The rats responded to supplemental methionine, but the PER value remained low, 0.28, as against 2.97 for those fed on the caesin diet. The apparent digestibility of cassava LPC has been shown to be only 50% (Vieira, 1983). The low digestibility along with the possible presence of anti-nutritional factor(s) may explain the poor nutritive value of cassava LPC. Quinoids formed by polyphenol oxidase during extraction could react with proteins to lower the nutritional value. In a thin layer chromatographic study of cassava leaves, Thakur et al. (1974) found 55 phenolic constituents; some of these compounds can form quinoids and react with the amino group of lysine, thus making it unavailable (Telek and Martin, 1983).

Studies with poultry, however, show that cassava LPC could be used satisfactorily as a source of protein for chick. In fact, cassava LPC has proven to be a more effective protein source than fish meal (Adegbola and Oke, 1973). The fibrous residue that remains after leaf protein extraction can still be used as a feed for ruminants (Pirie,

1971), thus making the production of LPC a more economically justifiable venture.

Chapter III

EVALUATION OF CASSAVA LEAF MEAL AS A REPLACEMENT FOR COCONUT OIL MEAL IN BROILER DIETS

INTRODUCTION

The scope for increased poultry production in the tropical regions is greatly handicapped by the rising cost and chronic shortages of conventional concentrate feeds. It is now recognized that the long-term growth of poultry farming would depend on the better utilization of lesser-known new feed sources. Cassava (Manihot esculenta Crantz) leaf meal warrants investigation in this context.

Cassava is cultivated extensively in the tropics, where its starchy roots provide the staple food for over 500 million people (Lancaster et al., 1982). It also produces a lush, high protein foliage which is at present returned to the soil as a green manure. The protein content of cassava leaves is extremely high for a non-legume and may be as high as 39.9% in young leaves (Tupynamba and Vieira, 1979). Eggum (1970) reported that cassava leaf protein is deficient in methinine, but rich in lysine. Cassava leaf meal (CLM) is also a good source of calcium and trace minerals (Ravindran et al., 1982). Despite its availability in large quantities and potential as a protein supplement, there is

little published data regarding the feeding value of CLM for poultry. This is perhaps due to the high levels of HCN in fresh cassava leaves (Gondwe, 1974), but can be lowered to safer levels by proper processing (Chapter V).

Ross and Enriquez (1969) observed progressive depression in gain and feed efficiency of broilers with increasing levels of CLM in a corn-soybean meal diet. Supplementation of diets containing 20% CLM with methionine and soybean oil resulted in performance comparable to that of the control. Ravindran et al. (1983b) reported that, at low levels of inclusion, feeding value of CLM for Japanese quail is similar to that of dehydrated alfalfa meal. Wyllie and Chamanga (1979) found CLM to be a superior substitute for cottonseed meal in broiler diets. The present study was undertaken to evaluate CLM as a replacement for coconut oil meal (COM) in broiler diets. COM is the major protein supplement used for poultry feeding in many tropical countries. Other objectives were to establish the highest possible replacement level within the physical and nutritional limitation of CLM and to determine whether various dietary additives would improve the utilization of diets high in CLM.

MATERIALS AND METHODS

Preparation of CLM

CLM was prepared from fresh cassava leaves and petioles harvested at two-month intervals from plants maintained for leaf production at the experimental plots of the Department of Animal Science, University of Peradeniya, Sri Lanka. No attempt was made to separate the old leaves from young leaves. The material was initially wilted in the shade for 2 to 3 days to lower the HCN content, dried overnight in an Unitherm oven at 65°C and then ground into a semi-powdery form.

Analytical Procedures

Because of the paucity of information on the nutrient composition of CLM, meals were also prepared from different parts of the leaves and from leaves of different maturity and stored for subsequent chemical analyses. Proximate analyses were performed on representative samples according to standard AOAC (1970) methods. Nitrogen-free extract content was calculated by difference. The samples were ground to pass through a 40 mesh screen and analysed for acid detergent fiber (ADF), permanganate lignin and cellulose using the procedures of Goering and Van Soest (1970). Neutral detergent fiber (NDF) was determined using the modified procedures of Robertson and Van Soest (1977). Hemi-

cellulose was calculated as the difference between ADF and NDF. HCN content was determined using the AOAC (1970) procedure as modified by Rajaguru (1972/73). The bulk density (g/cc) of COM and CLM were determined by placing a known weight of meals in a measuring cylinder. The cylinder was tapped 20 times for the samples to settle and the volume was recorded. All determinations were made in quadruplicates.

General Procedures

A total of nine feeding trials were conducted using unsexed, Cornish x White Plymoth Rock broiler chicks. The chicks were obtained from the Sri Lankan state hatchery, placed in electrically heated battery brooders and fed a 20% protein commercial mash for six days. On day 7, the chicks were weighed, and the heaviest and lightest were removed. The remaining were wing-banded and randomly assigned in groups of eight to pens to give groups of similar weight range.

All trials, except trial 4, lasted eight weeks. Trial 4 was terminated at three weeks. The chicks were kept in electrically heated battery brooders during the first four weeks of the trial and then transferred to colony cages. During these periods, they were fed a starter (21% protein) and a finisher (18% protein) diet, respectively. The birds had continuous access to feed and water. Diets were fed in mash form.

Table 7. Composition of broiler starter diets fed during 7 to 35 d. of age (Trials 1 and 2)

Ingredient	International reference number	Basal	Cassava leaf meal, %		
			10	20	30
		%			
Corn	4-26-023	35	35	35	35
Rice bran	4-03-928	14	14	14	14
Coconut oil meal	5-01-573	30	20	10	-
Cassava leaf meal	1-10-768	-	10	20	30
Fish meal (52% CP)	-	10	10	10	10
Sesame oil meal	5-04-220	5	5	5	5
Skim milk powder	5-01-175	5	5	5	5
Bone meal	6-00-400	1.25	1.25	1.25	1.25
Vitamin-mineral premix[a]		0.50	0.50	0.50	0.50
Salt	6-04-152	0.25	0.25	0.25	0.25
Chemical Composition, %					
Crude protein, %[b]		20.7	21.3	21.6	21.4
Crude fiber, %[b]		6.66	6.87	7.24	7.34
Metabolizable energy, kcal/kg[c]		2935	2930	2924	2920
Methionine and cystine, %[c]		0.76	0.77	0.78	0.79
Lysine, %[c]		1.01	1.07	1.13	1.19
Arginine, %[c]		1.84	1.65	1.46	1.27
Calcium, %[c]		1.36	1.49	1.62	1.75
Phosphorus, %[c]		1.22	1.24	1.26	1.28
HCN, mg/kg[c]		-	7	14	21

[a]Provided the following per kilogram of diet: vitamin A, 33000 IU; vitamin D_3, 2640 IU; vitamin E, 22 IU; vitamin K, 6.6 mg; riboflavin, 9.8 mg: calcium pantothenate, 11.0 mg; niacin, 40 mg; choline chloride, 55 mg; vitamin B_{12}, 22 mcg; manganese sulfate, 26.4 mg; calcium iodate, 6.6 mg; cobalt sulfate, 2.76 mg; zinc oxide, 2.76 mg.
[b]Determined values.
[c]Calculated values.

Table 8. Composition of broiler finisher diets fed during 36 to 63 d of age (Trials 1 and 2)

Ingredient	Basal	Cassava leaf meal, %		
		10	20	30
Corn	37	37	37	37
Rice bran	17	17	17	17
Coconut oil meal	30	20	10	-
Cassava leaf meal	-	10	20	30
Fish meal (52% CP)	7	7	7	7
Sesame oil meal	4	4	4	4
Skim milk powder	3.25	3.25	3.25	3.25
Bone meal	1	1	1	1
Vitamin-mineral premix[a]	0.50	0.50	0.50	0.50
Salt	0.25	0.25	0.25	0.25
Chemical Composition, %				
Crude protein, %[b]	18.3	18.5	18.3	18.6
Crude fiber, %[b]	6.51	6.86	7.02	7.16
Metabolizable energy Kcal/kg[c]	2961	2956	2951	2946
Methionine and cystine, %[c]	0.68	0.69	0.69	0.70
Lysine, %[c]	0.84	0.90	0.96	1.02
Arginine, %[c]	1.70	1.51	1.32	1.13
Calcium, %[c]	1.06	1.19	1.32	1.45
Phosphorus, %[c]	1.09	1.11	1.13	1.15

[a]See Table 7 for premix composition.

[b]Determined values.

[c]Calculated values.

Diets were fed to four replicates (pens) of eight chicks each in trials 1 and 2, and three replicates of eight chicks in trials 3 to 9. Individual body weights, group feed consumption and mortality data were monitored throughout the trials and feed/gain was calculated.

Treatments and Design

Trials 1 and 2 were conducted to assess the COM replacement value of CLM. Three experimental diets were formulated from the starter (Table 7) and finisher (Table 8) basal diets by replacing 10, 20 and 30% of the COM with CLM on a weight to weight basis. The diets were formulated to be isocaloric and isonitrogenous. At the termination of trial 1, birds were fasted for 12 hours and four birds from each pen were randomly selected, weighed and killed by exsanguination. Carcass recovery, and weights of liver, spleen, heart and pancreas were recorded. Carcass pigmentation rank was evaluated independently by three assessors using the Roche color fan.

Since increasing the level of CLM from 10 to 20 depressed broiler performance, trial 3 was carried out to determine the exact level of replacement at which growth depression begins. The basal diets were similar to those used in trials 1 and 2, and the experimental diets contained 10, 12.5, 15, 17.5 and 20% CLM. CLM used in trials 1 to 3 contained an average of 22.1% crude protein and 78 ppm HCN.

Trials 4 to 8 were concerned with overcoming the growth depression observed at high level inclusion of CLM. Twenty per cent CLM level was chosen, because it induced a moderate rather than severe depression that might be favourably influenced by dietary additives.

The effects of supplementing the basal and 20% CLM diets with methionine were studied in trial 4. A 2 x 3 factorial experiment was undertaken in which three levels of methionine (0, .125 and .25%) were added either to the basal or 20% CLM diets.

Trials 5 and 6 further examined the effects of methionine or thiosulfate supplemtation to the basal or 20% CLM diets. These supplements were added at a level of .25%. Thiosulfate, a sulfur donor, is known to provide the sulfur for the detoxification of cyanide.

CLM is a low energy poultry feed (Ravindran et al., 1983a). Thus the use of high levels of CLM would necessitate supplementation with high energy sources. Trials 7 and 8 were designed to study the effects of supplementing the basal and 20% CLM diets with 3% soyabean oil or a combination of 3% soyabean oil and .25% methionine. CLM used in trials 4 to 8 contained an average of 21.5% crude protein and 91 ppm HCN.

Trial 9 was designed to study the contribution, if any, of HCN to the growth depressing effects of CLM at 20% level of inclusion. The broilers were fed the basal diet and

experimental diets containing 25, 50, 100, 150 and 200 ppm potassium cyanide.

Data were analyzed by least-squares analysis of variance according to the procedures of Statistical Analysis System (SAS, 1979). Linear and quadratic effects of levels of CLM and cyanide were also tested.

RESULTS AND DISCUSSION

Nutrient composition

Nutrient composition of CLM as influenced by the age of leaves is presented in Table 9. Crude protein, nitrogen-free extract and HCN contents decreased with maturity, whereas fiber, ash and ether extract contents increased. Very young leaves contained 37.4% crude protein and this decreased to 19.5% in old, mature leaves. Use of only young leaves would therefore result in a high protein feedstuff, but this procedure would produce a meal with relatively high levels of HCN. The decrease in HCN content with leaf maturity is in accordance with the results of several workers (Gondwe, 1974; Lutaladio *et al.*, 1984; Williams, 1979).

Petioles had lower crude protein and higher fiber contents than the leaf blades, but HCN contents were similar (Table 10). Thus, the amount of leaf blades present will largely determine the nutritive value of CLM. A leaf meal of high nutritive value could be prepared by using leaf

Table 9. Chemical composition and hydrocyanic acid content of dehydrated cassava leaves as influenced by the age of leaves (dry matter basis)

Component	Very young leaves[a]	Young leaves[b]	Old leaves[c]
Crude protein, %	37.37 ± 1.28[d]	28.73 ± 0.76[d]	19.50 ± 0.42[d]
Crude fiber, %	8.33 ± 0.83	17.03 ± 0.41	28.13 ± 0.49
Ether extract, %	3.83 ± 0.51	6.07 ± 0.18	6.97 ± 0.41
Ash, %	4.00 ± 1.16	5.37 ± 0.59	7.87 ± 0.12
Nitrogen-free extract, %	46.47 ± 0.71	42.80 ± 1.10	37.67 ± 0.29
Neutral detergent fiber, %	18.43 ± 0.59	32.43 ± 1.96	47.40 ± 0.56
Acid detergent fiber, %	9.30 ± 0.56	17.97 ± 0.48	31.27 ± 0.54
Hemicellulose, %	9.13 ± 0.18	14.40 ± 2.22	15.13 ± 0.35
Cellulose, %	8.37 ± 0.54	13.27 ± 0.66	22.67 ± 0.62
Lignin, %	0.93 ± 0.09	4.37 ± 0.18	8.37 ± 0.27
Hydrocyanic acid, mg/kg[e]	210.67 ± 12.35	185.33 + 14.5	94.00 ± 4.02

[a]Expanding leaves.

[b]Fully developed, but immature leaves.

[c]Mature leaves.

[d]Mean ± S.E. (n=3).

[e]Fresh material from very young, young and old leaves contained 444 ± 24.6, 289.6 ± 12.6 and 158.7 ± 9.34 mg HCN per kg, respectively. The corresponding values for percent dry matter were 10.9 ± 0.10, 16.27 ± 0.24 and 21.47 ± 0.33, respectively.

Table 10. Chemical composition and hydrocyanic acid content of meals prepared from different parts of casssava leaves (dry matter basis)

Components	Petioles	Leaf blades alone	Whole leaves
Crude protein, %	11.70 ± 0.75[a]	28.80 ± 0.76[a]	23.03 ± 0.79[a]
Crude fiber, %	33.27 ± 0.74	17.10 ± 0.25	24.37 ± 0.81
Ether extract, %	1.73 ± 0.03	10.57 ± 0.33	7.50 ± 0.15
Ash, %	9.67 ± 0.30	7.23 ± 0.12	8.03 ± 0.19
Nitrogen-free extract, %	43.63 ± 1.64	36.30 ± 0.10	37.07 ± 0.54
Neutral detergent fiber, %	51.37 ± 0.39	42.70 ± 0.63	46.13 ± 0.66
Acid detergent fiber, %	34.40 ± 1.18	32.33 ± 0.22	34.40 ± 0.91
Hemicellulose, %	16.97 ± 0.79	10.37 ± 0.83	11.63 ± 0.75
Cellulose, %	18.47 ± 0.46	27.43 ± 0.69	26.23 ± 0.75
Lignin, %	15.67 ± 1.09	4.60 ± 0.50	7.60 ± 0.41
Hydrocyanic acid, mg/kg[b]	103.67 ± 2.41	108.33 + 5.55	103.67 ± 2.61

[a]Mean ± S.E. (n=3).

[b]Fresh material from petioles, leaf blades and whole leaves contained 154.67 ± 6.75, 211.00 ± 6.44 and 192.67 ± 3.48 mg HCN per kg, respectively. The corresponding values for percent dry matter content were 17.20 ± 0.35, 24.53 ± 1.11 and 20.77 ± 0.72, respectively.

blades alone. This will, however, be uneconomical, since petioles constitute about 25 to 30% of the foliage dry matter yields.

Level of COM replacement

Substitution of CLM for COM had significant linear and quadratic effects on gains ($P < .001$), feed intake ($P < .001$) and feed efficiency ($P < .01$) of broilers (Table 11). Broiler performance was improved at 10% CLM level and this may be attributed to a better balance of amino acids, since the amino acid profile of CLM is superior to that of COM (Ravindran et al., 1982). The growth response may also be partly attributed to overcoming the lysine deficiency caused by an excess of arginine in the COM-based basal diet. Excess arginine has been reported to increase lysine requirement at deficient levels of dietary lysine (Scott et al., 1982).

High levels (20 and 30%) of CLM inclusion resulted in depressions in gain, feed intake and feed efficiency. Reduced feed intake was probably due to the increased bulkiness (g/cc) of the diets. In the present study, CLM was determined to be 3.25 times bulkier than COM. High levels of CLM increased bulkiness to the extent that feed intake was physically reduced. The semi-powdery nature (dustiness) of CLM could have been another contributing factor.

Table 11. Performance of broilers fed diets containing various levels of cassava leaf meal (Trials 1 and 2)[a]

Diet	Gain per bird(g)[b,d]	Feed intake per bird(g)[b,e]	Feed per gain[c,e]	Mortality
Basal	983	3205	3.26	1/64
10% CLM	1027	3229	3.14	2/64
20% CLM	922	3017	3.27	1/64
30% CLM	725	2472	3.41	3/64
SEM[f]	14	114	0.04	-

[a] Average of eight replicates of eight birds each.

[b] Treatment effect ($P < .001$); linear effect ($P < .001$).

[c] Treatment effect ($P < .01$); linear effect ($P < .01$).

[d,e] Quadratic effect ($P < .001$, $< .01$).

[f] Standard error of means.

The depression in weight gain at 20% CLM inclusion parallels the depression in feed intake, suggesting that feed intake may be the primary factor causing the poor growth. At 30% level, however, reduced feed intake could not totally explain the reduced growth. Presence of anti-nutritional factors, such as HCN and tannin, in CLM may have been partially reponsible for the observed effects on growth. CLM used in these trials was determined to contain 78 ppm HCN. The slight increases in dietary fiber level with increasing levels of CLM may also have contributed to the poor growth. It has been shown by some workers that birds fed fibrous and bulky (low density) diets spent three to nine times longer eating the feed than those fed diets of high density (Jensen et al., 1962; Reddy et al., 1962). Similarly in the present study, birds eating high CLM diets appeared restless and spent more time eating. The increased time spent on feeding would mean a lowered available energy for metabolism and a reduced growth rate.

Mortality seemed to be normal, ranging between 2 and 5%, and was not influenced by dietary treatments. The gains of birds in trials 1 and 2 were appreciably lower than those in the other trials. This is probably due to the severe hot weather experienced during the trial period.

Dietary treatments had significant ($P < .05$) quadratio effect on the carcass recovery percentage of broilers, with a slight decrease at 30% CLM level (Table 12). Liver and

spleen weights (as a % of body weight) linearly ($P < .01$) increased as the level of dietary CLM increased. Pancreatic weight (as a % of body weight) was heavier ($P < .05$) in the 30% CLM diet compared to the other treatments. No gross abnormalities were observed in the external appearance of the organs. Hepatic and splenic hypertrophy provides further evidence to the possible involvement of anti-nutritional factors in causing growth depression at high dietary levels of CLM. The pancreatic enlargement in birds fed 30% CLM diets may be suggestive of the pancreatic compensatory response to trypsin inhibitor. The presence of trypsin inhibitor in cassava leaves has never been reported. Explanation for this hypertrophy may probably lie in the tannins contained in CLM. Griffiths and Mosley (1980) showed that the presence of tannin stimulates increased pancreatic secretion in rats. Increased pancreatic secretion, on the other hand, has been shown to increase pancreatic weight in the rat (Green and Lyman, 1972).

Table 12. Carcass characteristics of broilers fed diets containing varying levels of cassava leaf meal (Trial 1)[a]

Diet	Percent of body weight					Carcass pigmentation rank[e]
	Carcass[b]	Liver[c]	Spleen[c]	Heart	Pancreas[d]	
Basal	69.6	2.33	0.18	0.49	0.28	1.72
10% CLM	70.3	2.37	0.20	0.53	0.28	2.53
20% CLM	69.9	2.40	0.22	0.50	0.28	2.66
30% CLM	68.3	2.78	0.26	0.50	0.33	2.25
SEM[f]	0.5	0.09	0.01	0.02	0.01	0.19

[a]Average of 16 birds

[b]Treatment effect ($P < .05$); quadratic effect ($P < .05$).

[c]Treatment effect ($P < .01$); linear effect ($P < .01$).

[d]Treatment effect ($P < .05$).

[e]Treatment effect ($P < .01$); linear effect ($P < .05$); quadratic effect ($P < .05$).

[f]Standard error of means.

Table 13. Performance of broilers fed diets containing 0, 10, 12.5, 15, 17.5 and 20% cassava leaf meal (Trial 3)[a]

Diet	Gain per bird(g)[b]	Feed intake per bird(g)[c]	Feed per gain[d]
Basal	1297	3129	2.41
10% CLM	1375	3058	2.22
12.5% CLM	1338	3053	2.28
15% CLM	1294	3014	2.33
17.5% CLM	1213	2922	2.41
20% CLM	1189	2833	2.38
SEM[e]	26	40	0.04

[a]Average of three replicates of eight birds each.

[b]Treatment effect ($P < .001$); linear effect ($P < .001$); quadratic effect ($P < .001$).

[c]Treatment effect ($P < .01$).

[d]Treatment effect ($P < .05$); quadratic effect ($P < .05$).

[e]Standard error of means.

Carcass pigmentation values responded ($P < .05$) linearly and quadratically with increasing levels of CLM in the diet. CLM has been reported to be a rich source of xanthophylls (Agudu, 1972), and the use of CLM in broiler diets, therefore, offers an added advantage by giving a more desirable color to the broiler skin.

Results of trial 3 confirm the growth stimulatory effect of CLM at 10% level of inclusion (Table 13). Growth improvement was maintained up to 12.5% CLM level. Birds fed a level of 15% CLM consumed 3.7% less feed, but performed as well as those fed the basal diet. Incorporating levels of higher than 15% markedly depressed feed intake and growth. It appears that broiler chicks will readily accept and tolerate a level of 15% CLM without adversely affecting growth.

Effects of dietary supplementation

In trial 4, level of methionine did not significantly ($P > .10$) influence the gains and feed efficiency of broilers fed either the basal and 20% CLM diet (Table 14). However, a numerical trend toward improved growth was observed when methionine was added to diets containing 20% CLM. Methionine supplementation tended ($P < .10$) to increase the feed intake of CLM-based diets.

Results of trials 5 and 6 are summarized in Table 15. Supplementation of methionine and sodium thiosulfate sig-

nificantly ($P < .001$) improved the gains of birds fed diets containing 20% CLM without, however, having any beneficial effect when added to the basal diet. Irrespective of the supplementation, the performance of birds fed CLM diets in both cases was inferior to that of birds fed the basal diet. The lack of growth response to methionine in the basal diet was unexpected, since the calculated methionine contents (0.76 and 0.68% during the starter and finisher phases, respectively) of the basal and 20% CLM diets were similar. This lack of response may be attributable to the deficiency of other amino acids that may limit the utilization of methionine. COM is known to be deficient in lysine (Creswell and Brooks, 1971; Ravindran et al., 1982). The calculated lysine content of COM-based basal diet was 0.12% less than that of the 20% CLM diet (Tables 7 and 8). Furthermore, Mee and Brooks (1973) reported the availability and true digestibility of lysine in COM to be extremely poor. Poor availability of lysine is due to its binding with aldehydes produced during peroxidation of polyunsaturated fats at the high processing temperatures of coconut meal (Butterworth and Fox, 1963; Samson, 1971). Thus a deficiency of lysine may have been responsible for the lack of response to the methionine supplementation.

Sodium thiosulfate is a known antidote against cyanide poisoning, supplying the labile sulfur for detoxification (Way, 1981). The response to thiosulfate addition in CLM-

Table 14. Effects of methionine supplementation to basal and 20% cassava leaf meal diets on broiler performance (Trial 4)[a]

Diet	Gain per bird(g)[b]	Feed intake per bird(g)[b,c]	Feed per gain
Basal	274	630	2.30
Basal + 0.125% methionine	271	614	2.27
Basal + 0.25% methionine	277	651	2.35
20% CLM	257	581	2.26
20% CLM + 0.125% methionine	264	602	2.28
20% CLM + 0.25% methionine	269	627	2.33
SEM[d]	6	16	0.04

[a]Average of three replicates of eight birds each.

[b]Cassava leaf meal effect ($P < .05$)

[c]Methionine effect ($P < .10$).; methionine linear effect ($P < .05$).

[d]Standard error of means.

Table 15. Effects of methionine and sodium thiosulfate supplementation to basal and 20% cassava leaf meal diets on broiler performance (Trials 5 and 6)[a]

Diet	Gain per bird(g)[b]	Feed intake per bird(g)[c]	Feed per gain
Basal	1221	3129	2.56
Basal + 0.25% methionine	1229	3174	2.58
Basal + 0.25% thiosulfate	1208	3138	2.60
20% CLM	1096	2869	2.61
20% CLM + 0.25% methionine	1169	2950	2.52
20% CLM + 0.25% thiosulfate	1136	2925	2.57
SEM[d]	20	87	0.05

[a]Average of six replicates of eight birds each.

[b]CLM effect ($P < .001$); supplement effect ($P < .001$); unsupplemented vs methionine ($P < .01$); CLM x supplement interaction ($P < .06$).

[c]CLM effect ($P < .05$).

[d]Standard error of means.

based diets may indicate that the improvements observed with methionine may be partly attributed to its role as a source of labile sulfur. The beneficial effect of methionine on CLM-based diets is consistent with the findings of Ross and Enriquez (1969). The results of the present study lend more credibility to their hypothesis that methionine serves both to overcome a deficiency of sulfur amino acids and as a source of sulfur for cyanide detoxification.

All response criteria were improved ($P < .01$) when 3% soybean oil was added to the basal or to the 20% CLM diet (Table 16), suggesting that energy was limiting in both diets. Further addition of methionine had no effect on the growth response of the birds fed the basal diet. Though the addition of methionine to 20% CLM diet improved ($P < .05$) body weight gain, it failed to support growth numerically equivalent to that obtained with the basal diet. The significant CLM x supplement interaction ($P < .05$) indicate that dietary supplementation of soybean oil and methionine improved feed intake to a much greater degree in broilers fed 20% CLM diet compared to those fed the basal diet. Irrespective of dietary supplementation, feed intake of CLM-based diets remained lower than that of controls. The overall results suggest that the bulkiness, low energy content, methionine deficiency and presence of anti-nutritional factors are the major factors limiting the high level use of CLM in broiler diets.

Table 16. Performance of broilers fed the basal and 20% cassava leaf meal diets supplemented with soybean oil or soybean oil plus methionine (Trials 7 and 8)[a]

Diet	Gain per bird(g)[b,c]	Feed intake per bird(g)[b,d]	Feed per gain[b,c]
Basal	1229	3088	2.51
Basal + 3% SBO	1345	3151	2.34
Basal + 3% SBO + 0.25% meth.	1341	3076	2.29
20% CLM	1071	2822	2.64
20% CLM + 3% SBO	1145	2934	2.56
20% CLM + 3% SBO + 0.25% meth.	1205	2976	2.47
SEM[e]	15	22	0.02

[a]Average of six replicates of eight birds each.

[b]CLM effect ($P < .001$); supplement effect ($P < .001$); unsupplemented vs SBO ($P < .01$); unsupplemented vs SBO plus methionine ($P < .01$).

[c]SBO vs SBO plus methionine ($P < .05$).

[d]CLM x supplement interaction ($P < .05$).

[e]Standard error of means.

Effects of cyanide level

Gains of broilers tended to decrease linearly ($P < .08$) with increasing levels of cyanide (Table 17). It appears that a dietary level of 25 ppm cyanide may produce a measurable, as well as economically significant depression in performance of broilers. Levels of 25 ppm cyanide decreased gain by 3%.

Significant linear ($P < .01$) and quadratic ($P < .05$) effects on feed intake were observed, as the level of cyanide was increased. Feed intake was lowered beyond 100 ppm cyanide level. Feed efficiency and mortality were not influenced by dietary cyanide level. No signs of toxicity were observed even in birds fed diets containing as high as 200 ppm added cyanide.

Diets containing 20 and 30% CLM were calculated to contain 14 and 21 ppm HCN, respectively. Results of Trial 9 therefore imply that cyanide may be a factor contributing to the growth depression observed at high dietary levels of CLM.

CONCLUSIONS

The present results confirm the previous suggestions that CLM could be used as a poultry feed ingredient (Ravindran et al., 1983b; Ross and Enriquez, 1969). Because HCN appears not to be a problem at 33.3% replacement of COM by CLM, poultry producers in the tropics could benefit Table

17. Performance of broilers fed diets containing different levels of potassium cyanide (Trial 9)[a]

	Gain per bird(g)[b]	Feed intake per bird(g)[c]	Feed per gain	Mortality
Basal	1236	3055	2.47	0/24
Basal + 25 ppm CN^{-1}	1200	3052	2.54	1/24
Basal + 50 ppm CN^{-1}	1217	3078	2.53	0/24
Basal + 100 ppm CN^{-1}	1196	3058	2.56	0/24
Basal + 150 ppm CN^{-1}	1186	2979	2.51	1/24
Basal + 200 ppm CN^{-1}	1179	2952	2.50	0/24
SEM[d]	27	19	0.02	-

[a]Average of three replicates of eight birds each.

[b]Linear effect ($P < .08$).

[c]Treatment effect ($P < .01$); linear effect ($P < .01$); quadratic effect ($P < .05$).

[d]Standard error of means.

economically by incorporating more CLM in feed formulations. CLM has potential in poultry diets both as a source of protein and xanthophylls.

It appears that the unfavourable effects of high dietary levels of CLM are due to bulkiness, reduced energy intake, methionine deficiency and cyanide. The role of methionine in supplying labile sulfur for cyanide detoxification further aggravates its inherent deficiency in CLM. Bulkiness probably is the major limiting factor. Future studies should use pelleting to remove this variable, so that any depression in growth can be attributed to factors other than bulkiness and dustiness.

Cyanide is not the only anti-nutritional factor known to be present in cassava leaves. Other deleterious agents identified include condensed tannins and oxalate (Lancaster and Brooks, 1983). Presence of condensed tannins in cassava leaves was first reported by Reed et al. (1982). Condensed tannins form strong molecular complexes with protein (Oh et al., 1980), thereby lowering amino acid availability. Condensed tannins have also been implicated in forming unavailable complexes with methionine (Ford and Hewitt, 1974). Reduced digestibilities of amino acids with increasing tannin contents have been reported in chicks (Stephenson et al., 1971; Nelson et al., 1975). Several studies (Elkin et al., 1978; Armstrong et al., 1974; Armanious et al., 1973) have shown that the detrimental effects of high tannin sor-

ghum on chicks can be partly ameliorated by supplementing practical diets with methionine. The condensed tannins in CLM could, therefore, increase the methionine requirement when fed to poultry and may actually compound the effects of cyanogenic glucosides. The nutritional significance of cassava leaf tannins need to be evaluated in future studies.

The results suggest that the growth depression caused at high levels of CLM could be overcome, to a great extent, by fortifying such diets with soybean oil and methionine, but this would add more cost to formulations and may nullify the economic advantage of including CLM.

Chapter IV

FEEDING VALUE AND DIGESTIBILITY OF CASSAVA LEAF MEAL FOR SWINE

INTRODUCTION

High cost of conventional proteins in livestock feeding has evoked considerable interest to evaluate green leaves as alternate sources of protein. It is known that leaves offer the highest yield of protein of all crops (Telek and Martin, 1983). Cassava (Manihot esculenta Crantz) leaves, a by-product of cassava root production, represent one such potential source of protein available in the tropical region that warrants evaluation.

Mature cassava leaves contain around 22% crude protein on a dry matter basis (Ravindran et al., 1982), whereas values of up to 39.9% has been reported for young leaves (Tupynamba and Vieira, 1979). Cassava leaf protein is deficient in methionine, posibly marginal in tryptophan but rich in lysine (Rogers and Milner, 1969; Eggum, 1970). Ravindran et al. (1983c) reported that the amino acid profile of cassava leaf meal (CLM) compares favorably with that of the alfalfa meal, and of coconut oil meal (COM) which is the major protein supplement in livestock diets in many tropical countries, including Sri Lanka. CLM, however, was richer in lysine. CLM is also a good source of minerals, especially

of calcium and trace minerals (Ravindran et al., 1982). These attributes qualify CLM for use as a swine feed in the tropics, but this potential remains unexploited probably because of its HCN content. Though fresh cassava leaves may contain up to 1630 mg HCN per kg dry matter, this should not be a major concern since cyanide levels can be lowered to safer levels by processing (Chapter V).

Published information on the use of CLM in swine feeding is extremely limited. Early studies of feeding fresh cassava leaves showed that palatability was depressed and growth performance was lowered with increasing amounts of leaves in swine diets (Lee and Hutagalung, 1972; Mahendranathan, 1971). The adverse effects were evidently due to the high cyanide levels in the fresh leaves, since supplemental methionine and thiosulfate proved effective in improving the performance (Lee and Hutagalung, 1972). Cassava leaves, however, were well accepted as rabbit feed (Harris et al., 1980). Studies with poultry suggest that CLM, if properly processed to lower the cyanide level, could be successfully utilized as a source of vegetable protein in the tropics (Ross and Enriquez, 1969; Ravindran et al., 1983b). The objectives of the present study were to (1) evaluate CLM as a replacement for COM in swine diets and (2) determine the nutrient digestibility and, protein and energy utilization values of CLM for swine.

MATERIALS AND METHODS

Preparation of CLM

CLM was prepared from fresh cassava leaves and petioles harvested at two-month intervals from plants maintained for leaf production at the experimental plots of the Department of Animal Science, University of Peradeniya, Sri Lanka. No attempt was made to separate the old leaves from young leaves. The material was initially wilted in the shade for 2 to 3 days to lower the HCN content, dried overnight in an Unitherm oven at 65°C and then ground into a semi-powdery form.

Feeding trials

Two trials were conducted to evaluate CLM as a replacement for COM in pig diets. COM and CLM used in these trials contained an average of 20.6 and 21.5% crude protein, respectively (Table 18). In each trial, 36 crossbred pigs with an average initial weight of 14.5 kg were assigned to pens by sex and weight, with two barrows and one gilt per pen. Treatments were allocated to pens at random within each of the three replicates. The basal diet contained 40% COM and the experimental diets were formulated by replacing 0,33.3,66.7 and 100% COM with CLM on a weight to weight basis (0,13.3,26.7 and 40% CLM in the total diet). The composition of the diets is presented in Table 19.

A 4 x 4 Latin square change-over design, with an extra period, was employed (Lucas, 1957). Each period was of 14 days duration. Treatment pattern during the extra period was identical to that in the last period of the Latin square design and permits estimation of residual effects, that are carried over for than one period.

The pigs were housed in concrete floored pens (5.4 m^2/pen) under an open shed. Pigs were limit-fed twice daily at 1000 and 1700 h. The amount of feed was determined by the replicate group consuming the least during a 30-minute feeding period at each weighing. Feeding levels were held constant until next weighing. Body weights were determined and feed intake was adjusted at fortnightly intervals. In most periods, feeding level was determined by pigs fed 40% CLM. Diets were mixed with water (1 liter/kg) immediately before feeding to enhance consumption. Water was provided ad libitum.

Data were analysed according to Lucas (1957) using the procedures of Statistical Analysis System (SAS, 1979). Linear and quadratic effects of levels of CLM were also tested. Average daily gain and feed to gain ratio were computed on a pen basis.

Table 18. Chemical composition of coconut oil meal and cassava leaf meal (dry matter basis)

Item	Coconut oil meal	Cassava leaf meal
Gross energy, kcal/g	4.72	4.81
Crude protein (N x 6.25), %	20.6	21.5
Ether extract, %	8.6	7.8
Ash, %	5.6	7.2
Neutral detergent fiber, %	50.6	41.4
Cell contents, %	49.4	58.6
Acid detergent fiber, %	21.1	30.3
Hemicellulose, %	29.5	11.1
Cellulose, %	20.8	23.8
Permanganate lignin, %	4.8	6.9
HCN, mg/kg	-	76

Table 19. Composition of diets used in the feeding and balance trials.

Ingredient	International feed number	Basal	Cassava leaf meal, %		
			13.3	26.7	40.0
		%			
Corn	4-26-023	25	25	25	25
Rice bran	4-03-928	25	25	25	25
Coconut oil meal	5-01-573	40	26.7	13.3	-
Cassava leaf meal	1-10-768	-	13.3	26.7	40
Fish meal (44% CP)	-	6	6	6	6
Skim milk powder	5-01-175	2.25	2.25	2.25	2.25
Bone meal	6-00-400	1	1	1	1
Vitamin-mineral premix[a]	-	0.50	0.50	0.50	0.50
Salt	6-04-152	0.25	0.25	0.25	0.25
Chemical Composition, (% DM basis)					
Crude protein, %[b]		17.68	17.68	18.16	18.41
Acid detergent fiber, %[b]		9.21	10.83	11.61	12.55
Metabolizable energy, kcal/kg[c]		3.61	-	-	-
Methionine + cystine, %[c]		0.60	0.61	0.63	0.64
Lysine, %[c]		0.78	0.85	0.93	1.00
Arginine, %[c]		1.74	1.49	1.24	0.99
Calcium, %[c]		0.89	1.06	1.23	1.40
Phosphorus, %[c]		1.16	1.14	1.12	1.10
HCN, mg/kg[c]		-	10	20	30

[a] Supplied (per kilogram of premix): 6,600,000 IU vitamin A, 528,000 IU vitamin D_3, 880 IU vitamin E, 1.98 g ribiflavin, 4.4 g calcium pantothenate, 8.8 g niacinamide, 22.0 g choline chloride, 8.8 g vitamin B_{12}, 1.32 g vitamin K, 5.28 g manganese sulfate, 1.98 g; copper carbonate, 0.99 g calcium iodate, 0.55 g cobalt sulfate and 0.22 g zinc oxide.

[b] Determined values.

[c] Calculated values.

Balance trials

Two balance trials, each with two collection periods, were conducted using 16 crossbred barrows with an average initial weight of 37.2 kg (8 barrows per trial). In each trial, two randomized blocks were formed based on weight and diets were assigned at random within each block. The four dietary treatments were similar to those used in the feeding trials (Table 19).

Pigs were housed in individual metabolism cages in an open shed and allowed 7 days to adapt to the cages. During this period, the basal diet was offered ad libitum to all pigs. During the following 7 days, pigs were adjusted to an equal feeding regimen wherein the experimental diets were offered 90% of the ad libitum consumption of the pig eating the least feed within each outcome group. Diets, mixed with equal quantity of water, were fed twice daily at 0900 and 1700 h. One hour after feeding, any refused feed was removed, weighed, sampled and oven-dried. The amount of weigh-back was then adjusted for moisture level. There were, however, only few instances of feed refusal. Dry matter intakes for trials 1 and 2 were 1,780 and 2.236 g/d, respectively. Water was provided by nipple waterers. Each pig completed two 5-day total collection periods separated by a 7-day interval.

Fecal material was collected twice daily, weighed and dried in an Unitherm oven at 60°C for 36 h. After drying,

the samples were allowed to attain equilibrium with atmospheric moisture, reweighed and stored in sealed two-ply paper bags. At the end of the collection period, the dried feces were pooled and coarsely ground in a hammer mill before representative samples were taken. Urine was collected in plastic buckets containing 60 ml 25% v/v hydrochloric acid. Urine pH was checked daily and additional acid was added to maintain the pH below 5. Urine collections were pooled daily and volume was determined on day 5. Aliquots were taken for analyses and frozen. Samples of feed, feces and urine were analysed as described by Ravindran et al. (1984).

Data were analyzed by a randomized complete block design using analysis of variance (SAS, 1979). Nutrient digestibility, and energy and protein utilization values for CLM were calculated by linear regression analysis. Calculations for each component was based on the percentage of that component in the diets that was from CLM. The method assumed that there were no associative effects.

RESULTS AND DISCUSSION

Feeding trials

The carry-over effects of treatments between periods, as measured by the extra-period after the latin square, were non-significant ($P > .10$). Dietary level of CLM had significant ($P < .001$) linear and quadratic effects on average

daily gain and feed efficiency of pigs (Table 20). Pig performance was improved at 33.3% replacement of COM by CLM (13.3% CLM in the total diet) and this may be attributed to a better balance of amino acids, since the amino acid profile of CLM is superior to that of COM (Ravindran et al., 1982). CLM is a rich source of lysine (Eggum, 1970; Ravindran et al., 1982), whereas the lysine in COM is both low (Creswell and Brooks, 1971) and poorly available to pigs (Mee and Brooks, 1973). The growth response at 13.3% CLM level may also be partly attributed to overcoming the lysine deficiency caused by an excess of arginine in the COM-based basal diet (Table 19).

Based on the superior amino acid profile, pig performance might be expected to improve when dietary CLM level is increased. Gains and feed efficiency of pigs fed 26.7% CLM, however, were similar to those fed the basal diet. An adverse effect on performance was observed when CLM totally replaced COM. Pigs fed diets with 40% CLM gained 15% slower and required 18% more feed per unit gain than did controls. Presence of anti-nutritional factors, such as HCN and tannin, may explain the loss of benefit of the superior amino acid profile of CLM at these levels. The increases in dietary fiber level with increasing levels of CLM may also have contributed to this loss of benefit.

The calculated HCN contents of diets containing 26.7 and 40% CLM were 20 and 30 ppm, respectively. Based on the

Table 20. Performance of growing pigs fed diets containing varying levels of cassava leaf meal (Trials 1 and 2)[a]

Diet	Average daily gain(Kg)[b,c,d]	Feed per gain[b,c,d]
Basal	0.38	3.3
13.3% CLM	0.44	2.8
26.7% CLM	0.39	3.2
40% CLM	0.32	3.9
SEM[e]	0.03	0.12

[a]Each mean represents six replicates of three pigs each, used in an extra-period latin-square change over design. Average initial weight was 14.5 kg.

[b]Treatment effect ($P < .001$)

[c]Linear effect ($P < .001$)

[d]Quadratic effect ($P < .001$)

[e]Standard error of means

results of the present study, it appears that pigs could very well tolerate a level of 20 ppm HCN without any adverse effect on growth performance. Presence of condensed tannins in CLM has recently been reported (Reed et al., 1982). Condensed tannins have been implicated in forming unavailable complexes with proteins (Oh et al., 1980), thereby lowering amino acid availability (Stephenson et al., 1971; Nelson et al., 1975). Nutritional importance of cassava leaf tannins needs to be investigated in future studies.

Balance trials

Daily gain and dry matter per gain for pigs during the balance trials followed a trend similar to those observed in the feeding trials (Table 21). Fecal dry matter output increased ($P < .001$) and percentage dry matter of fresh feces decreased ($P < .001$) with increasing dietary levels of CLM. Other researchers (Cooper and Tyler, 1959a,b; Kornegay, 1978, 1981; Kass et al., 1980) have made similar observations with inceasing levels of fibrous feedstuffs. The increased moisture content of the feces as a result of fiber addition is attributed to the water holding capacity of fiber (Monte, 1981), particularly of cellulose and hemicellulose fractions (Mendeloff, 1984).

Urine volume linearly decreased ($P < .001$) as the level of CLM was increased in the diet. This response is difficult to explain, but compliments the results of Henry

Table 21. Amount of feed, feces and urine, fecal dry matter, average daily gain and dry matter per gain of barrows in the balance trials

Item	Cassava leaf meal, %				SEM[a]
	0	13.3	26.7	40	
No. of observations	8	8	8	8	-
Avg. initial weight, kg	37.4	37.4	37.3	37.6	0.83
Avg. final weight, kg	39.6	39.7	39.3	39.2	0.84
Dry matter intake, kg/d	2.00	2.01	2.01	2.02	0.04
Avg. daily gain, kg/d[b,c,d]	0.44	0.46	0.40	0.32	0.02
Dry matter per gain[b,c,d]	4.52	4.37	5.03	6.31	0.20
Fecal dry matter output, kg/d[b,c]	0.44	0.49	0.59	0.70	0.02
Dry matter of fresh feces, %[b,c,d]	35.3	32.4	30.0	29.6	0.42
Urine output, liters/d[b,c]	2.38	2.33	1.96	1.89	0.05

[a]Standard error of means

[b]Treatment effects ($P < .001$)

[c]Linear effect ($P < .001$)

[d]Quadratic effect ($P < .01$)

Table 22. Apparent nutrient digestibility of diets containing different levels of cassava leaf meal

Criteria	Cassava leaf meal, %				SEM[a]	Trt effect[b]	CLM[c]
	0	13.1	26.7	40			
Dry matter	78.1[d]	75.4	70.5	65.5	.22	***	49.3
Ether extract	87.7	85.0	80.7	75.9	.42	***	40.5
Ash	3.6	46.1	45.1	44.9	1.61	***	40.5
Cell contents	83.4	81.4	77.1	72.5	.20	***	53.4
Cell wall	68.8	64.3	59.1	53.0	.60	***	41.3
Acid detergent fiber	52.1	55.1	49.1	40.6	.91	***	31.3
Hemicellulose	86.5	76.6	73.9	71.9	1.26	***	46.0
Cellulose	63.1	68.3	63.4	55.5	.60	***	47.9
Lignin	11.2	24.4	25.5	27.8	1.41	***	30.0

[a]Standard error of means

[b]Probability, *** P < .001)

[c]Calculated using linear regression based on the % of nNtrient in the diets that was from CLM.

[d]Mean of eight observations

(1976), Kornegay (1978) and Lindemann et al. (1985) who reported a negative effect of fiber level on urine volume. The data of Lindemann et al. (1985), however, indicate that the total water excreted via feces and urine for all dietary fiber levels would be similar if fecal moisture is added to urine volume.

Effects of dietary levels of CLM on apparent nutrient digestibility are presented in Table 22. Digestion coefficients for dry matter, ether extract, cell contents, cell wall were depressed ($P < .001$), while those for ash and lignin were improved ($P < .001$) as CLM was substituted for COM. Digestion coefficients for acid detergent fiber and cellulose were improved at 13.3% level of CLM inclusion, but declined with further substitution.

The digestibility of dry matter of the basal diet was much lower than the value of 89.3% reported by Kornegay (1973) for corn - soybean meal basal diet. The high fiber content of our basal diet could account for this difference.

The predicted digestibilities for various components in CLM calculated by linear regression using the percentage of nutrient that was from CLM were (%): dry matter, 49.3; ether extract, 54.5; ash, 40.5; cell contents, 53.4; cell wall, 41.3; acid detergent fiber, 31.3; hemicellulose, 46.0; cellulose, 47.9; and lignin, 30.0.

Inclusion of CLM to the diets produced a pronounced effect on the nitrogen metabolism of pigs (Table 23). Fecal

nitrogen excretion increased ($P < .001$), while urinary nitrogen excretion and digested nitrogen decreased ($P < .01$). Nitrogen retention, although not significantly different, appeared to be lower for pigs fed the diets containing 40% CLM. The increase in fecal nitrogen excretion may suggest that a part of the nitrogen in CLM is bound to some compound, possibly tannins, and excreted undigested. Hale and McCormick (1981) observed a similar nitrogen excretory pattern in pigs fed diets containing peanut skin, a feed ingredient high in tannin.

Digestibility of protein was reduced ($P < .001$) as CLM protein was substituted for COM protein. The protein digestibility was depressed 3.32% for each 1% increase in acid detergent fiber content, which is much greater than the values of 0.9 and 1.52 for diets containing soybean hulls (Kornegay, 1978) and dehydrated alfalfa meal (Kass et al., 1980), respectively. The numerically larger depression may suggest that, in addition to fiber, condensed tannins may also have been partially responsible for the poor digestibility of CLM protein. Naturally occurring tannins, due to the formation of indigestible tannin - protein complexes or tannin effects on enzyme activity (Reed et al., 1982), are known to depress protein digestibility in pigs (Almond et al., 1979; Cousins et al., 1981; Ford and Hewitt, 1979; Hale and McCormick, 1981). Using linear regression, the protein digestibility of CLM was estimated to be 56.2%. It is much

Table 23. Apparent digestible protein and apparent protein utilization values of cassava leaf meal for swine

Item	Level of CLM				SEM[a]	Trt effect[b]	CLM[c]
	0	13.3	26.7	40.0			
Daily dry matter intake, kg	2.00[d]	2.01	2.01	2.02			
N per kg of dry matter							
intake	31.1	31.6	32.2	32.5	.64	**	-
fecal	6.7	7.4	9.1	10.7	.41	***	-
urinary	9.3	8.4	7.7	7.5	.60	**	-
digested	24.4	24.2	23.1	21.8	.99	**	-
retained	15.1	15.8	15.4	14.3	1.20	NS	-
Digestible protein, %	78.5	76.6	71.7	67.1	.39	***	56.2
Apparent NPU, %[e]	48.6	50.0	47.8	44.0	1.12	**	39.1
Apparent BV, %[f]	61.9	65.3	66.7	65.6	1.47	*	72.6

[a]Standard error of means.

[b]Probability, NS = nonsignificant, * < .05, ** P < .01, *** P < .001.

[c]Calculated using linear regression based on the % of N in the diets that was from CLM.

[d]Mean of eight observations.

[e]Apparent net protein utilization = N retention expressed as a % of gross N consumed.

[f]Apparent biological value = N retention expressed as a % of apparent N digested.

Table 24. Digestible energy, metabolizable energy and metabolizable energy corrected for nitrogen retention of cassava leaf meal (dry matter basis)

Item	Level of CLM				SEM[a]	trt effect[b]	CLM[c]
	0	13.3	26.7	40.0			
Energy,							
digestible, %	82.3[d]	80.2	75.9	73.0	.20	***	60.6
metabolizable, %	81.0	79.1	74.8	72.0	.21	***	58.8
metabolizable, corrected for N retention, %[e]	78.9	77.0	72.4	69.8	.24	***	57.3
Energy per gram of diet, kcal							
digestible	3.77	3.70	3.53	3.46	.01	***	2.91
metabolizable	3.71	3.65	3.48	3.46	.01	***	2.83
metabolizable, corrected for N retention	3.62	3.56	3.37	3.31	.01	**	2.76

[a]Standard error of means.

[b]Probability, *** P < .001.

[c]Calculated using linear regression based on the % of energy in the diets that was from CLM.

[d]Mean of eight observations.

[e]The correction factor for N retention was calculated to be 7.41 kcal of energy per gram of retained N.

lower than the value of 73% reported by Loosli et al. (1954) for COM protein.

The apparent NPU value was maximum at 13.3% CLM level and was linearly ($P < .01$) depressed with further addition of CLM. Apparent BV of all CLM-based diets were higher than that of control. It is apparent that pigs efficiently utilize cassava leaf protein even when dietery COM was completely replaced by CLM. This is consistent with the better amino acid balance of CLM-based diets. Regression analyses on the balance data yielded estimates of 39.1 and 72.6% for apparent NPU and apparent BV, respectively.

The determined energy utilization values for the diets and the calculated energy utilization values for the CLM are shown in Table 24. As CLM was incorporated to the basal diet, coefficients for digestible energy (DE), metabolizable energy (ME) and ME corrected for nitrogen retention (ME_n) linearly decreased ($P < .001$), indicating that gross energy of CLM is less efficiently utilized than that of COM. The predicted DE, ME and ME_n values for CLM were 2.91, 2.83 and 2.76 kcal/g, respectively. The author is not aware of any published energy utilization values of CLM for swine. The 2.83 kcal/g ME value is considerably higher than the 1.99 kcal/g true ME value reported by Ravindran et al. (1983a) for poultry. The difference between species probably reflects the greater microbial cellulytic activity of the pig.

The DE value of CLM obtained in our study was much lower than the value of 3.6 kcal/g reported for COM by Creswell and Brooks (1971). This would imply that less energy was available for metabolism with each increment of dietary CLM. Thus the poor performance of swine at high dietary levels of CLM may also be partly related to a reduced DE intake.

The ME of CLM was 97.3% of the DE. Diggs et al. (1965), evaluating 18 feedstuffs, found the average efficiency of utilization of DE to be 94.7%. May and Bell (1971) reported that ME values are approximately 98% of the DE values, although individual feed ingredients vary considerably.

Conclusions

The results indicate that when judiciously used CLM is an acceptable source of protein for swine. Its high lysine content permits considerable savings in the expensive COM for balancing swine diets. The present study has demonstrated that CLM may replace up to 66.7% of the COM (26.7% of the total diet) in growing swine diets without depressing performance. Most efficient gains were observed at 33.3% replacement of COM. The findings also indicate that cassava leaf protein is utilized more efficiently, although the nutrients in CLM are not as digestible as those in COM.

The poorer performance of pigs when CLM totally replaced COM could be explained on the basis the low energy content of CLM. CLM is deficient in methionine (Eggum, 1970) and this deficiency is further aggravated by the need for additional methionine to detoxify cyanide (Maner and Gomez, 1973) and tannin (Campadadal et al., 1976; Elkin et al., 1978). Evaluation of methods to overcome these effects would improve the feasibility of using high levels of CLM in swine feeding.

Chapter V

INFLUENCE OF PROCESSING AND STORAGE ON THE HYDROCYANIDE CONTENT OF CASSAVA LEAF MEAL

INTRODUCTION

Cassava leaf meal (CLM) has good potential as an animal feed in the tropics on the basis of its protein, amino acid and mineral contents (Ravindran et al., 1982). Use of CLM in animal nutrition, however, has been limited due to the presence of the two cyanogenic glucosides, linamarin, and lotaustralin (Conn, 1973). Upon hydrolysis by the action of either the endogenous enzyme linamarase in damaged plant tissues or the β-glucosidases within the digestive tract of animals, these glucosides liberate free hydrogen cyanide (HCN) which is highly toxic (Nestel and MacIntyre, 1973).

Fresh cassava leaves contain high levels of cyanogenic glucosides (Gondwe, 1974). Yeoh and Oh (1979) found the leaf HCN levels to be six times higher than those of cassava roots. But this should not be a major deterrent, since simple drying of leaves has been reported to eliminate most, if not all, of the cyanide (Obregon, 1968).

Because of the increased need to utilize non-conventional feeds, justified by the scarcity of protein sources in Sri Lanka, the present study was initiated with the object of developing an effective processing technique

to lower the HCN content of CLM to safer levels suitable for animal feeding. Efficacy of three processing methods, namely drying, chopping and wilting, and their combinations were evaluated. The influence of storage time on the HCN and crude protein contents of CLM was also studied.

MATERIALS AND METHODS

Processing methods

Cassava leaves with petioles were obtained at two-month intervals from plants (variety MU 22) maintained for leaf production at the experimental unit of the Department of Animal Science, University of Peradeniya, Sri Lanka. Two kilogram samples each of freshly harvested cassava leaves were subjected to one of 16 treatment combinations, involving two methods of drying (sun- or oven-), two methods of leaf preparation (unchopped or chopped) and four methods of wilting (no wilting or wilting for 1, 2 or 3 days).

Sun-drying was carried out by spreading the leaf material on aluminum trays and keeping the trays on a cement floor. Oven-drying of leaf materials was done in an unitherm oven at 60°C for 6-30 hours. Chopping was done manually using knives. Wilting was carried out in a well ventillated room by spreading the leaves on the floor. Leaves were turned twice per day to avoid mold formation. The study was quadruplicated in time to obtain more reliable data.

Storage

Four 10 kg samples of CLM were stored in airtight three-ply paper bags in room temperature. The bags were opened monthly during a 8-month post-processing storage, mixed thoroughly and sampled for HCN and crude protein (CP) estimations.

Chemical analyses

Representative samples were analyzed in duplicate for their dry matter and CP contents according to standard AOAC (1970) procedures. HCN contents were determined by the alkaline titration method (AOAC, 1970), as modified by Rajaguru (1972/73). Samples of fresh cassava leaves of different maturity stages were also analyzed for HCN.

RESULTS AND DISCUSSION

HCN content of fresh cassava leaves

Cyanide levels decreased as the leaves matured (Table 25), consistent with the reports of De Bruijn (1973) that stage of maturity is one of the major factors governing cyanide content of cassava leaves. Separate analyses of petioles and leaf blades also revealed a similar declining trend. On a dry matter basis, very young, young and old leaves contained 4073, 1766 and 745 mg HCN/kg, respectively.

In expanding leaves the cyanide level in petioles was higher than in the blades, whereas in old leaves the reverse

Table 25. Hydrocyanic acid content of fresh cassava leaves as influenced by stage of maturity

Stage of maturity	HCN, mg/kg fresh weight[a]		
	Petioles	Leaf Blades	Whole leaves
Very young[b]	525.0	398.3	444.0
Young[c]	268.3	294.3	289.6
Old[d]	104.7	174.3	158.7

[a]Mean of three samples

[b]Expanding leaves

[c]Fully expanded, but immature leaves

[d]Mature, green leaves

was true. Similar observations have been reported by De Bruijn (1973).

Effects of Processing

Studies of Cooke and Madwagwu (1978) and Gomez and Valdiviesa (1984b) show that sun-drying was more effective than oven-drying in lowering the cyanide content of cassava roots. The relatively greater efficiency of sun-drying was attributed to its slower rate of drying. In the present study, no such differences were observed between sun- and oven-drying (Table 26). Both drying methods were equally effective in lowering the cyanide levels in cassava leaves.

Drying by artificial means is a costly process requiring substantial investment and operational costs, and may not be a feasible idea in developing countries. Use of sun-drying, on the other hand, is limited because of its dependence on climatic conditions. The rate of sun-drying was primarily determined by the wind and relative humidity. The time taken to reduce the moisture content to 10% or less was greater when the relative humidity was more than 80%. Lowering the moisture level to 13 to 15% was found to permit safe storage of the product, but was insufficient for grinding. When the prevailing conditions were dry and windy, complete drying was achieved in two days. In general, drying rates were enhanced by chopping and wilting prior to drying. Chopping and wilting the leaves for three days

Table 26. Hydrocyanic acid content (mg/kg dry matter) of cassava leaf meal as influenced by processing methods

Method of Wilting	Oven-drying		Sun-drying	
	Full[a]	Chopped[b]	Full[a]	Chopped[b]
No wilting	170[c]	106	173	109
1-day wilting[d]	146	85	141	88
2-day wilting	117	75	114	72
3-day wilting	86	56	93	53

[a]Freshly harvested cassava leaves contained an average of 1436 mg HCN/kg dry matter (range 1225 to 1627).

[b]Freshly chopped leaves (analyzed 3 to 6 hours after chopping) contained an average of 1045 mg HCN/kg dry matter (range 921 to 1228).

[c]Each value represent mean of four samples.

[d]Wilted in the shade.

Table 27. Reduction in HCN content (as a % of initial level in freshly harvested leaves) of cassava leaf meal as influenced by different processing methods

Method of	Oven- or sun-drying	
	Full leaves	Chopped leaves[a]
No wilting	87.8	92.5
1-day wilting	90.0	94.0
2-day wilting	91.9	94.8
3-day wilting	93.7	96.2

[a]Chopping alone resulted in 27.2% loss in HCN of freshly harvested leaves.

reduced the moisture content from 79 to 26% and the leaves thus processed required only a day of sun-drying under optimum conditions.

The results indicate that simple drying of the cassava leaves resulted in considerable reduction of HCN content, eliminating 88 - 92% of the initial cyanide level (Table 27). Cyanide loss during the drying process appears to be more complex than simply driving off the readily volatizable free cyanide. It probably also involves an initial drying phase during which the cells lose physiological integrity, causing the enzyme linamarase to come into contact with cyanogenic glucosides (bound form) and liberating free cyanide.

Chopping and wilting prior to drying resulted in further reductions in the cyanide content of the final product (Table 26 and 27). Cyanide levels were linearly decreased with increasing duration of wilting. The mechanism of action in both cases is due to endogenous linamarase activity on glucosides following either loss of cell integrity (wilting) or mechanical tissue damage (chopping).

A combination of chopping and 3-day wilting prior to drying proved most effective, lowering the cyanide content of CLM to around 55 mg/kg dry matter. Thus substantial amounts of cyanide remains in the processed produc, but cyanide contained is within the safety levels for poultry and pig feeding (Chapter III and IV).

Table 28. HCN and crude protein contents of cassava leaf meal as influenced by storage time (dry matter basis)

Storage time (months)	HCN content (mg/kg)	HCN loss as a % of initial level	Crude protein,%
0	91[a]	-	22.7[a]
1	78	14.3	-
2	68	25.3	22.6
3	59	35.2	-
4	49	46.2	21.7
5	43	52.7	-
6	40	56.0	20.9
7	38	58.2	-
8	38	58.2	20.3

[a]Mean of four samples.

Effects of storage time

The HCN and CP contents of CLM as influenced by storage time are present in Table 28. The cyanide levels diminished at a rapid rate during the first four months of storage and then the rate of decrease gradually slowed. Almost 46% of the initial HCN level of 91 mg/kg was lost during the first four months, while only a further 12% was lost during the next four months. Narahari and Kothandaraman (1983) observed a similar diminishing trend in cyanide levels with storage of rubber seeds.

A gradual diminishing trend in the CP content of CLM was also observed with storage time. The initial CP content of 22.7% declined to 21.7 and 20.3% after four and eight months of storage, respectively. The results suggest that four-month storage time appears to be a compromise to further lower the cyanide level with minimum loss in feeding value.

CONCLUSIONS

The significance of the present study had been to ascertain that it is possible to produce low cyanide CLM using simple processing techniques. It is evident that drying alone can effect considerable detoxification, and that when combined with chopping and wilting the cyanide content in the final product can be reduced to levels which safe for non-ruminants. The cyanide content could be further

reduced, with little sacrifice in nutritive value, by merely storing the processed meal at room temperature for a period of four months.

CHAPTER VI

EFFECTS OF LEAF HARVESTING ON ROOT AND LEAF YIELDS OF CASSAVA

INTRODUCTION

High cost of conventional feedstuffs and concern for maximum utilization of resources have stimulated interest in the use of nutrients in agricultural residues, such as cassava (Manihot esculenta Crantz) leaves, for animal feeding. Cassava leaves are a good source of protein, calcium and trace minerals (Ravindran et al., 1982). Recent studies show that properly processed cassava leaf meal could be successfully used as a partial replacement for coconut oil meal in poultry and swine diets (Chapters III and IV). At low levels of inclusion, the feeding value of cassava leaf meal was found to be equivalent to that of alfalfa meal (Ravindran et al., 1983b).

Harvesting the leaves during the growing season are known to depress the cassava root yields. Ahmad (1973) reported that frequent leaf harvesting lowered the root crop to almost one-half of the normal production. Cassava is cultivated primarily for its tuberous roots; therefore it is imperative that a suitable defoliation practice which would not greatly reduce root yields, should be established. The present investigation was conducted to study the effects of

frequency of leaf harvesting on the root and leaf yields of a short-age cassava variety, MU 22.

MATERIALS AND METHODS

The trial was conducted from October 1983 to May 1984 at Peradeniya (longitude 80° 29'E, latitude 7° 13' N, 488 m above sea level), Sri Lanka. Mean daily temperatures of the area vary from 23.1°C in January to 26.0°C in April, with an annual average of 24.4°C (Domros, 1974). The rainfall during the trial period was 1512 mm and was well distributed. The soil at the experimental site was reddish brown latasolic soil with a pH of 6.1.

The trial was laid out in a randomized block design with four replicates. Plot size was 1.2 m x 4.8 m. Cuttings of about 40 cm from mature healthy stems of variety MU 22 were planted at 60 cm spacings on ridges which were 60 cm apart (27,225 plants/ha) on October 2, 1983. Details of the treatments are presented below:

Treatment 1 - No leaf harvesting during the growing season.

Treatment 2 - One leaf harvest during the growing season. Leaves were cut three months after planting.

Treatment 3 - Two leaf harvests during the growing season. Leaves were cut at three and five months after planting.

At final harvest, seven months after planting, both root and leaf weights were recorded in all three treatments.

Each plot received a basal application of one ton of poultry litter/ha (equivalent to approximately 52 kg N, 36 kg P_2O_5 and 35 kg K_2O per hectare) at planting. No fertilizers were applied, because fertilizers are not normally used under traditional subsistence farming conditions. The plots were hand-weeded thrice during the first two months after planting.

At each leaf harvest, stems were cut with knives 90 cm above ground level, and the leaves were stripped off the harvested tops. Any green leaves remaining on the plant were also removed. Fresh leaf weights were recorded at each harvest and representative samples were taken for dry matter (DM) and crude protein (CP) determinations. The number and weights of roots in each plot were recorded and fresh root yields in tons/ha were computed. The numbers of total and marketable roots per plant were also counted. Roots with a minimum diameter of 7.5 cm were considered marketable.

The DM and CP contents of leaf samples were determined using standard AOAC (1970) procedures. Leaf CP yield was

calculated by multiplying the total leaf DM yield by the per cent CP in the appropriate treatment.

Data were analyzed using the Statistical Analysis System (SAS, 1979). Differences among treatment effects were tested using the Duncan's multiple range test.

RESULTS AND DISCUSSION

Effects of leaf harvesting on root and leaf production attributes of cassava are presented in Table 29. Total and marketable fresh root yields were significantly ($P < .05$) depressed by leaf harvesting. The depression was pronounced when leaves were harvested twice during the growing season. One complete defoliation during the growing season lowered total fresh root yield by 12.9%, whereas two defoliations resulted in a 56.1% depression of root yield. The corresponding decreases in marketable fresh root yield were 21.7 and 75.0%, respectively. Marketable roots accounted for 66.2% of the total fresh root production in the controls. This decreased ($P < .05$) to 59.5 and 37.7%, respectively when leaves were harvested once and twice during the growing season. The reduction in root yields with leaf harvesting may be attributed to a decrease in effective photosynthetic area and the consequent reduction in carbohydrate supply for root enlargement (Hunt et al., 1977).

The number of roots per plant and weight per individual root are considered as determinants of tuberous root yield

of cassava (Dahniya et al., 1981). The number of roots per plant was unaffected when leaves were harvested once during the growing season, but harvesting twice resulted in a reduction ($P < .05$) in the number of roots per plant. Dahniya et al. (1981) reported that leaf harvesting frequency had little effect on the number of roots produced, but markedly lowered the individual root size.

The number of marketable roots per plant decreased ($P < .05$) with increasing number of leaf harvests during the growing season. Almost half the plants which were defoliated twice did not have any marketable roots. Defoliation had no effect on the size of marketable roots.

Leaf harvesting during the growing season resulted in increases ($P < .05$) in fresh leaf yield. When leaves were harvested only at root maturity, a fresh leaf yield of 21.2 t/ha was obtained. Defoliating once and twice during the growing season increased leaf yields by 53.7 and 83.4%, respectively. Similar trends ($P < .05$) were observed for leaf DM and leaf CP yields. The DM content of leaves decreased ($P < .05$) and CP content increased ($P < .05$), when the leaves were harvested more frequently.

It is noteworthy that a leaf DM production of 4.63 t/ha (equivalent to 7.92 t/ha/year) with a CP content of 20.4% is possible as a by-product of cassava root production. This yield is much higher than the leaf DM yields of 1.17 and 1.85 t/ha/year obtained at root harvest for two cassava

Table 29. Effects of leaf harvesting on various root and leaf production attributes of cassava

Measurement	No. of leaf harvests during growing season			SEM
	0	1	2	
Total fresh root yield, t/ha	13.93[a]	12.07[b]	6.14[c]	0.32
No. of roots/plant	3.49[a]	3.52[a]	1.95[c]	0.10
Avg. weight/root, g	147[a]	126[b]	116[b]	3.04
Marketable fresh root yield, t/ha	9.24[a]	7.23[b]	2.26[c]	0.28
No. of marketable roots/plant	1.68[a]	1.39[b]	0.40[c]	0.07
Avg. weight/marketable root, g	206[a]	198[a]	218[a]	16.21
% marketable root yield	66.33[a]	59.90[a]	36.81[b]	2.38
Fresh leaf yield, t/ha	21.19[c]	32.57[b]	38.87[a]	0.63
Leaf DM, %	21.90[a]	20.71[b]	19.63[c]	0.23
Leaf CP, %	20.40[c]	23.33[b]	26.98[c]	0.18
Leaf DM yield, t/ha	4.64[c]	6.75[b]	7.63[a]	0.11
Leaf CP yield, t/ha	0.95[c]	1.57[b]	2.06[a]	0.26

[a,b,c]Means in the same row with different superscripts differ significantly ($P < .05$), based on Duncan's multiple range test.

varieties by Gomez and Valdivieso (1984a) in Columbia. The high rainfall conditions experienced may have partly account for the high leaf production in the present study. Varietal differences observed in terms of total leaf production (Lutaladio, 1984) may also partly explain this discrepancy.

The data suggest that in short-age (early maturing) cassava varieties good yields of both leaves and roots can be obtained by defoliating once during the growing season. Two defoliations during the growing season increased the leaf DM and leaf CP yields by 67 and 111%, respectively, but depressed the root yield by more than half. On the contrary, Dahniya et al. (1981) evaluating two long-age (late maturing) cassava varieties reported that reasonable yields of both leaves and roots were maintained even after 3 to 4 leaf harvests during the growing season. It appears that cassava varieties respond differently to defoliation in terms of root yield.

When leaves were harvested twice during the growing season, a leaf DM yield of 7.7 t/ha (2.06 t CP/ha) was obtained. Projected to an annual basis, this would correspond to 13.2 t DM/ha (3.5 t CP/ha). The data suggest that if cassava is maintained exclusively for leaf production, it can be planted more closely at a density of 95,000 plants/ha with a potential leaf DM production of over 40 t/ha.

Whether the aim of cassava cultivation should be leaf production or a compromise towards root production will depend on the relative prices of CLM, cassava root meal and traditional feedstuffs. The results of the present study, however, demonstrate that acceptable root yields could be obtained by the adoption of suitable defoliation practices for a particular cassava variety.

Chapter VII

GENERAL CONCLUSIONS

A healthy animal industry is necessary to help achieve improved nutrition in the developing nations of the tropics. Attempts to expand the animal industries in these areas has long been hampered by the everrising cost and chronic shortages of traditional animal feeds. Seasonal and unreliable rainfall, marginal soil fertility and subsistence farming conditions leave such nations with an erratic supply of locally grown sources of animal feed. Against this background, the importance of a successful search for cheaper, non-traditional feed ingredients becomes obvious.

The overall objectives of the present study was to investigate the feasibility of developing CLM as an animal feed in the tropics. The results of the feeding and balance trials have demonstrated the scope for using CLM as a replacement for COM in non-ruminant diets and for reducing the cost of feed formulations in the tropics. Adequately processed CLM could be used up to a level of 15% (50% replacement of COM) in balanced poultry diets, whereas dietary levels of up to 26.7% CLM (66.7% replacement of COM) were well tolerated by the swine. Results of the balance study with swine indicate that cassava leaf protein is utilized more efficiently, although the nutrients in CLM are

not as digestible as those in COM. With both poultry and swine, the most efficient and economical gains were obtained when CLM replaced a third of COM, suggesting that use of low levels of CLM in feed formulations will permit greater savings in feed cost compared to moderately high levels of CLM.

Possible reasons for the unfavourable effects on broiler and swine growth of high levels of CLM are as follows: The bulkiness and dusty nature of CLM was probably the major contributing factor. Future studies should use pelleted CLM in an effort to remove this variable. Presence of anti-nutritional factors such as cyanide and tannin in CLM may also be of concern. The toxicity of these anti-nutritional factors, however, tends to be associated with poor nutritional levels when intake of protein and sulfur is insufficient. It is also conceivable that other factors such as high fiber, low methionine and low energy contents of CLM could have been partly responsible for its poor feeding value at high levels of inclusion.

The existence of cyanogenic glucosides has made some form of processing a prerequisite for the use of cassava leaves as an animal feed. The present study has shown that simple drying is sufficient to eliminate 90% of the initial cyanide level. A combination of chopping and 3-day wilting prior to drying proved the most efficient method of processing. Processed CLM could be stored for up to four months with little loss in feeding value.

A processing method effective in lowering cyanide levels in varieties of low toxicity may not necessarily be as efficient when used for a high toxic variety. Since the cyanide content of fresh cassava leaves is known to vary widely (Table 6), additional studies relating the effectiveness of processing methods to the initial cyanide levels in the fresh leaves are needed.

The data presented show that a cassava leaf yield of 4640 kg dry matter per hectare could be obtained as a by-product of cassava root production of a seven-month variety. The results also indicate that it is possible to harvest 6750 kg cassava leaf dry matter per hectare by defoliating once during a 7-month period and to produce within 86% of the normal yield of roots. Cassava is primarily grown for its starchy roots. The present study has demonstrated that reasonable yield of roots could be produced by adopting suitable defoliation practices for a particular cassava variety.

It is evident from the data presented thus far that potential of CLM as a non-ruminant feed in the tropics is too great to be ignored. Properly processed CLM can be successfully used in poultry and swine diets provided that care is taken to balance the energy and sulfur amino acid contents. The economics and technology of processing cassava leaves, however, has to be evaluated if the production and utilization of CLM are to reach commercial levels.

Dehydration of cassava leaves by artificial means is an expensive and impractical option for the developing countries of the tropics due to the high cost of equipment and energy required. On the other hand, sun-drying on concrete floors represent a simple way of drying cassava leaves. Sun-drying, however, is totally dependent on climatic conditions and hence restricted to the dry seasons which imposes a limitation on the year-round production of CLM. Research programs are urgently needed to develop appropriate technology for the production and processing of CLM.

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APPENDIX

TABLE 1. PERFORMANCE OF BROILERS FED VARYING LEVELS OF CASSAVA LEAF MEAL (TRIAL 1)[a]

	Gain per bird(g)[b]	Feed intake per bird(g)[c]	Feed per gain[d]
Basal	991	3253	2.38
10% CLM	1016	3191	3.14
20% CLM	924	3065	3.32
30% CLM	773	2557	3.31

[a]Average of four replicates of eight birds each.

[b]Treatment effect ($P < .001$); linear effect ($P < .001$); quadratic effect ($P < .001$).

[c]Treatment effect ($P < .01$); linear effect ($P < .01$); quadratic effect ($P < .08$).

[d]Treatment effect ($P < .01$); quadratic effect ($P < .09$).

TABLE 2. PERFORMANCE OF BROILERS FED DIETS CONTAINING VARYING LEVELS OF CASSAVA LEAF MEAL (TRIAL 2)[a]

	Gain per bird(g)[b]	Feed intake per bird(g)[c]	Feed per gain[d]
Basal	975	3157	3.24
10% CLM	1038	3279	3.16
20% CLM	921	2969	3.22
30% CLM	677	2386	3.52

[a]Average of four replicates of eight birds each.

[b]Treatment effect ($P < .001$); linear effect ($P < .001$); quadratic effect ($P < .001$).

[c]Treatment effect ($P < .01$); linear effect ($P < .001$); quadratic effect ($P < .01$).

[d]Treatment effect ($P < .05$); linear effect ($P < .06$); quadratic effect ($P < .05$).

TABLE 3. EFFECTS OF METHIONINE AND SODIUM THIOSULFATE TO BASAL AND 20% CLM DIETS ON BROILER PERFORMANCE (TRIAL 5)[a]

	Gain per bird(g)[b]	Feed intake per bird(g)[c]	Feed per gain[b,c]
Basal	1155	3019	2.61
Basal + .25% meth.	1167	3102	2.66
Basal + .25% thiosulf.	1136	3004	2.64
20% CLM	1052	2776	2.64
20% CLM + .25% meth.	1121	2888	2.58
20% CLM + .25% thiosulf.	1109	2871	2.59

[a]Average of three replicates of eight birds each.

[b]CLM effect ($P < .05$).

[c]CLM x supplement interaction ($P < .06$).

TABLE 4. EFFECTS OF METHIONINE AND THIOSULFATE SUPPLEMENTATION TO BASAL AND 20% CLM DIETS ON BROILER PERFORMANCE (TRIAL 6)[a]

	Gain per bird(g)[b]	Feed intake per bird(g)[c]	Feed per gain[b,c]
Basal	1287	3239	2.52
Basal + .25% meth.	1291	3246	2.51
Basal + .25% thiosulf.	1279	3272	2.56
20% CLM	1121	2942	2.62
20% CLM + .25% meth.	1218	3012	2.47
20% CLM + .25% thisulf.	1146	2978	2.56

[a]Average of three replicates of eight birds each.

[b]CLM effect ($P < .001$); basal vs methionine ($P < .05$).

[c]CLM effect ($P < .05$); supplement effect ($P < .07$).

TABLE 5. PERFORMANCE OF BROILERS FED 20% CLM AND BASAL DIETS SUPPLEMENTED WITH SOYBEAN OIL OR A COMBINATION OF SOYBEAN OIL PLUS METHIONINE (TRIAL 7)[a]

	Gain per bird(g)[b]	Feed intake per bird(g)[c]	Feed per gain[d]
Basal	1239	3129	2.53
Basal + 3% SBO	1342	3141	2.34
Basal + 3% SBO + .25% meth.	1343	3054	2.27
20% CLM	1067	2839	2.66
20% CLM + 3% SBO	1133	2985	2.63
20% CLM + 3% SBO + .25% meth.	1202	3044	2.53

[a]Average of three replicates of eight birds each.

[b]CLM effect ($P < .001$); supplement effect ($P < .001$); basal vs SBO ($P < .001$); basal plus meth. ($P < .001$).

[c]CLM effect ($P < .001$); supplement effect ($P < .06$); CLM x supplement interaction ($P < .06$): basal vs SBO ($P < .07$); basal vs SBO plus meth. ($P < .05$).

[d]CLM effect ($P < .001$); supplement effect ($P < .01$); basal vs SBO ($P < .01$); basal vs SBO plus meth. ($P < .001$); SBO vs SBO plus meth ($P < .08$).

TABLE 6. PERFORMANCE OF BROILERS FED BASAL AND 20% CLM DIETS SUPPLEMENTED WITH SOYBEAN OIL OR A COMBINATION OF SOYBEAN OIL PLUS METHIONINE (TRIAL 8)[a]

	Gain per bird(g)[b]	Feed intake per bird(g)[c]	Feed per gain[b,d]
Basal	1218	3047	2.50
Basal + 3% SBO	1347	3162	2.35
Basal + 3% SBO + .25% meth.	1339	3096	2.31
20% CLM	1075	2806	2.61
20% CLM + 3% SBO	1157	2883	2.49
20% CLM + 3% SBO + .25% meth.	1208	2908	2.41

[a]Average of three replicates of eight birds each.

[b]CLM effect ($P < .001$); supplement effect ($P < .001$); basal vs SBO ($P < .001$); basal vs SBO plus meth. ($P < .001$).

[c]CLM effect ($P < .001$); supplement effect ($P < .01$); basal vs SBO ($P < .01$); basal vs SBO plus meth. ($P < .01$).

[d]SBO vs SBO plus meth. ($P > .09$).

TABLE 7. THE EXTRA-PERIOD LATIN-SQUARE CHANGE-OVER EXPERIMENTAL DESIGN USED IN THE SWINE FEEDING TRIALS

Period	Block 1				Block 2				Block 3			
	1[a]	2	3	4	1	2	3	4	1	2	3	4
1	1[b]	2	3	4	1	2	3	4	1	2	3	4
2	2	1	4	3	3	4	1	2	4	3	2	1
3	3	4	1	2	4	3	2	1	2	1	4	3
4	4	3	2	1	2	1	4	3	3	4	1	2
5	4	3	2	1	2	1	4	3	3	4	1	2

[a]Represents the treatment sequence.

[b]The numbers in the heart of the table represent the treatments.

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DEVELOPMENT OF CASSAVA (*MANIHOT ESCULENTA* CRANTZ) LEAF MEAL AS AN ANIMAL FEED

by

Velmurugu Ravindran

Committee Chairman: Ervin T. Kornegay

Animal Science

(ABSTRACT)

Research was conducted in Sri Lanka to evaluate the feasibility of developing cassava (*Manihot esculenta* Crantz) leaf meal as an animal feed. In feeding trials with broilers, improvements in performance were observed at 10% inclusion of cassava leaf meal (CLM). High levels (20 and 30%) of CLM depressed gain, feed intake and feed per gain. Weights of spleen and liver (% of body weight) linearly increased with increasing levels of CLM. Carcass pigmentation values favored the CLM-based diets. Dietary additives (methionine, sodium thiosulfate or soybean oil plus methionine) improved the growth of broilers fed 20% CLM diet without, however, having any beneficial effect when added to the basal diet. Gain of broilers tended to decrease with increasing dietary levels of cyanide.

Feeding trials with growing pigs showed that CLM can be included up to 26.7% level without any effect on performance. Gain and feed per gain were improved at 13.3% CLM level, whereas marked depressions were noted at 40% CLM level. Results of the balance trials indicated that cassava leaf protein is utilized more efficiently by the pigs, although the nutrients in CLM are not as digestible as those in coconut oil meal. The overall results suggest that bulkiness, low energy content, methionine deficiency and presence of anti-nutritional factors, are the major factors limiting the high level use of CLM in non-ruminant diets.

Studies on the processing of CLM revealed that simple drying is sufficient to eliminate almost 90% of the initial cyanide level in the fresh cassava leaves. A combination of chopping and 3-day wilting prior to drying proved most effective in lowering the cyanide level of CLM.

Field trials conducted with a short-age cassava variety, MU 22, demonstrated that it is possible to increase cassava leaf dry matter yields by defoliating once during the growing season and to produce within 86% of the normal yield of roots. Two defoliations during the growing season depressed the root crop by more than half.