Growth performance of pigs fed on diets containing *Acacia karroo*, *Acacia nilotica* and *Colophospermum mopane* leaf meals

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Abstract

Growth performance of pigs fed diets containing 10 % *Acacia karroo*, *Acacia nilotica* and *Colophospermum mopane* leaf meals was investigated using 20 individually penned male Large White pigs weighing 32.4 ± 5.86 kg (mean ± sd) over 56 days.

There was no significant effect of including leaf meals on average daily liveweight gain and feed conversion ratio (*P* > 0.05). Pigs supplemented with *Acacia karroo* had significantly higher intake than the other diets (*P* < 0.05). The control diet had a higher digestibility of crude protein and dry matter compared to the treatment diets (*P* < 0.05) except for the dry matter digestibility of the *Acacia nilotica* supplemented diet, which was not different to that of the control diet (*P* > 0.05). After the 56-day feeding period, there was an increase in the secretion of trichloroacetic acid soluble proteins from the parotid glands in all diets that contained leaf meals. No change was observed in the mandibular glands (*P* < 0.05). The activity of hepatic microsomal uridine diphosphate glucuronyl transferase increased significantly for pigs fed on the supplemented diets (*P* < 0.05). Indices of mitosis in the small intestine, the kidney and the liver were not affected by inclusion of leaf meals (*P* > 0.05).
In conclusion, inclusion of leguminous leaf meals in pig fattening diets reduced digestibility of the feed, but did not affect growth rate. Inclusion of leguminous leaf meals in the diets of pigs is, therefore, a feasible technology that farmers may adopt as part of their feeding strategy for pigs.

**Key words:** alternative feeds, flavonoids, salivary proteins, tannins, tropical legumes

**Introduction**

Pig production in the tropics is constrained by seasonal feed deficits, high costs of feed, erratic supply of feed ingredients and competition between humans and pigs for ingredients used in livestock feed (Makkar 1993; D'Mello 1995; Halimani et al 2005). However, there is a wide spectrum of alternative feeds including agricultural by-products and leaf meals from various tree species, which may be available in large quantities (Makkar 1993; Ravindran 1993). The potential of using leaf meals in this context is high due their relatively high crude protein contents and the ability of most candidate species to thrive in diverse and, in instances, adverse climatic and soil conditions (D'Mello 1995).

There are many candidate species available to various communities. The most commonly used tree species are Leucaena leucocephala (Laswai et al 1997) and Manihot esculenta (Ravindran 1993). Efforts have been made to identify new plant species that can be tested and exploited in feeding pigs and other monogastrics (D'Mello 1995; Halimani et al 2005; Leterme et al 2005).

Studies in which leaf meals have been included in the diet of pigs have shown that inclusion levels of up to 10% lead to weight gains and feed conversion efficiencies that are superior to conventional control diets (D'Mello 1995). However, most studies have shown that inclusion of leaf meals may increase growth rate but depress feed conversion efficiency (Ravindran 1993; Halimani et al 2005). Most of the studies have assessed the response of the pigs over relatively short periods (Halimani et al 2005; Leterme et al 2005). The objectives of the study were to investigate the growth performance, feed intake, digestibility, production of salivary proline rich proteins and indices of mitosis in the small intestine, liver and the kidneys in pigs fed on diets containing 10% Acacia karroo, Acacia nilotica and Colophospermum mopane over a 56-day fattening period taking pigs to market weight. The hypothesis to be tested was that inclusion of 10 percent leaf meal in pig diets had no effect on growth performance.

**Materials and methods**

**Study sites**

Three leguminous leaf meals; Acacia karroo, Acacia nilotica and Colophospermum mopane were
harvested at Matopos Research Station in Zimbabwe. Chemical analyses of the leguminous leaf meals were carried out at the School of Veterinary Science, Langford, University of Bristol, UK. Chemical analyses of the feed and faecal samples were carried out in the Department of Animal Science, University of Zimbabwe. The feeding trial was conducted at the Pig Industry Board, Arcturus, Zimbabwe.

**Chemical analysis**

The chemical compositions of the leaf meals were determined according to the Association of Official Analytical Chemists (AOAC 1990). Levels of neutral detergent fibre (NDF), acid detergent fibre (ADF) and the acid detergent lignin (ADL) were determined according to Goering and Van Soest (1970). Crude fibre was analysed using the method of the AOAC (1990). Ash was obtained by igniting the samples in a muffle furnace. Gross energy (GE) was obtained using a bomb calorimeter. The total phenolics were determined gravimetrically using the Ytterbium acetate precipitation method described by Giner-Chavez et al (1997).

**Diets**

Diets were formulated using the Format International computer package (Specialised Animal Feed Company, Ltd, Zimbabwe), using data from the proximate analyses of the forages. The diets were isonitrogenous (about 206g CP/kg) and iso-energetic (13.14 MJ DE/kg), as shown in Table 1. Chromium (III) oxide was added to the diet at the rate of 2 g/kg as an external digestibility marker.

<table>
<thead>
<tr>
<th>Component</th>
<th>Control</th>
<th>10 % Colophospermum mopane</th>
<th>10 % Acacia karroo</th>
<th>10 % Acacia nilotica</th>
</tr>
</thead>
<tbody>
<tr>
<td>White maize</td>
<td>400.0</td>
<td>400.0</td>
<td>400.0</td>
<td>400.0</td>
</tr>
<tr>
<td>Acid oil</td>
<td>97.0</td>
<td>84.0</td>
<td>82.0</td>
<td>73.0</td>
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<tr>
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<td>100.0</td>
<td>110.0</td>
<td>120.0</td>
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<tr>
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<td>Limestone flour</td>
<td>30.0</td>
<td>0.0</td>
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<tr>
<td>Monocalcium phosphate</td>
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<td>12.0</td>
<td>6.0</td>
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<tr>
<td>Fine salt</td>
<td>16.7</td>
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<td>15.8</td>
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<tr>
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<td>0.1</td>
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</tr>
<tr>
<td>banox†</td>
<td>0.2</td>
<td>0.2</td>
<td>0.2</td>
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</tr>
<tr>
<td>SS3‡</td>
<td>3.0</td>
<td>3.0</td>
<td>3.0</td>
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<tr>
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<td>150.0</td>
<td>120.0</td>
<td>112.0</td>
<td>110.0</td>
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<tr>
<td>Forage</td>
<td>0.0</td>
<td>100.0</td>
<td>100.0</td>
<td>100.0</td>
</tr>
</tbody>
</table>

**Chemical Analysis**

CP

200.1  209.6  206.8  209.6
Animals

Twenty male Large White pigs weighing between 26 and 47.5 kg (mean ± sd: 32.4 ± 5.56) were randomly assigned to each of the four diets. The average weights of the treatment groups were balanced. The pigs were individually penned in pens measuring 4 m x 3 m. The experiment was carried out over 56 days, which mimics the on-farm fattening period of between 8 and 12 weeks. The animals were slaughtered at the end of the trial. The animals were individually fed on an ad libitum basis with the feed provided in a trough. The animals had free access to water, which was available through low-pressure nipple drinkers. All animals in the study were treated in compliance with internationally recognized codes and standards for animal welfare.

Feed intake was calculated as the difference between the weight of the feed offered and the weight of the refusals. The pigs were weighed at the end of every week and the daily liveweight gain was calculated. Weighing was done at 0800 hours, before feed was offered. Feed conversion ratio (FCR) was computed as weekly weight gain divided by amount of fed consumed per week. Chromium (III) oxide was used as an external digestibility marker and chromium analysis was conducted according to the procedures described in AOAC (1990).

Post-slaughter analyses

All the pigs were slaughtered by stunning and ex-sanguination, in conformity with Zimbabwean legislation on handling and slaughter of animals. Immediately after slaughter, the mandibular salivary glands, parotid salivary glands, two liver samples, a kidney sample and a small intestine sample were collected. The intestinal sample was collected at 25% of the length of the small intestine. Of the two liver samples, one was snap frozen in liquid nitrogen and then stored at -70°C.

<table>
<thead>
<tr>
<th></th>
<th>Fat (g/kg)</th>
<th>CF (g/kg)</th>
<th>Ash (g/kg)</th>
<th>Ca (g/kg)</th>
<th>P (g/kg)</th>
<th>NaCl (g/kg)</th>
<th>MJ DE/kg</th>
<th>Cys (g/kg)</th>
<th>Met (g/kg)</th>
<th>M+C (g/kg)</th>
<th>Lysine (g/kg)</th>
<th>Try (g/kg)</th>
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<tbody>
<tr>
<td>1</td>
<td>166.3</td>
<td>34.1</td>
<td>92.4</td>
<td>9.3</td>
<td>7.0</td>
<td>19.6</td>
<td>13.0</td>
<td>2.9</td>
<td>4.6</td>
<td>7.5</td>
<td>12.0</td>
<td>2.4</td>
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<td>2</td>
<td>88.4</td>
<td>46.6</td>
<td>85.9</td>
<td>8.9</td>
<td>8.2</td>
<td>19.5</td>
<td>13.2</td>
<td>2.9</td>
<td>4.9</td>
<td>7.5</td>
<td>12.3</td>
<td>2.5</td>
</tr>
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<td>3</td>
<td>95.9</td>
<td>41.8</td>
<td>75.6</td>
<td>9.9</td>
<td>7.0</td>
<td>19.2</td>
<td>13.2</td>
<td>2.9</td>
<td>4.5</td>
<td>7.4</td>
<td>12.2</td>
<td>2.5</td>
</tr>
<tr>
<td>4</td>
<td>108.8</td>
<td>40.8</td>
<td>73.4</td>
<td>10.0</td>
<td>7.1</td>
<td>19.5</td>
<td>13.2</td>
<td>2.9</td>
<td>4.6</td>
<td>7.6</td>
<td>12.4</td>
<td>2.6</td>
</tr>
</tbody>
</table>

† Banox; ‡ SS3 — feed additives
C. The other was placed in 10 % neutral buffered formalin. The salivary glands were removed and stored in plastic bags immediately after slaughter. Trichloroacetic acid soluble proteins (presumed to be proline rich proteins) were extracted using the methods described by Mehansho et al (1992). All the carcasses were kept in a cold room at 4°C for 24 hours, after which cold dressed mass was determined.

The Folin-Lowry (Lowry et al 1951) method was used to estimate the quantities of proteins in the 10 % trichloroacetic acid extract. Five millilitres of the alkaline solution (50 ml of 20 g/l Na₂CO₃ in 0.1 mol/l NaOH and 1 ml of 5 g/l CuSO₄.5H₂O in 10 g/l Na, K tartrate) was added to 1 ml of the test solution. The mixture was stirred thoroughly and allowed to stand at room temperature for 20 minutes then 0.5 ml of the diluted Folin-Ciocalteau reagent (water: reagent at 1:1) was added rapidly to the mixture and mixed immediately. The absorbance was read against a blank at 750 nm. Bovine serum albumin was used as standard.

The liver, kidney and small intestine samples were fixated in 10 % neutral buffered formalin (NBF) (100 ml formalin supplied as 40 % formaldehyde, 6.5 g anhydrous disodium hydrogen orthophosphate and 4 g sodium dihydrogen orthophosphate monohydrate made up with distilled water) and processed using the method described earlier (Halimani et al 2005) and adapted from Banks (1981) and Bancroft and Stevens (1977). Indices of mitosis (cells in metaphase per 100 cells) were estimated.

The assay for hepatic microsomal uridine diphosphate glucoronyl transferase (UDPGT) was adapted from Tavoloni et al (1983). Details for the procedure used have been reported earlier (Halimani et al 2005).

**Statistical analysis**

Daily liveweight gain, daily feed intake, crude protein digestibility, dry matter digestibility, 10 % TCA soluble proteins, indices of mitosis, UDPGT activity and feed intake were analysed using a general linear model as follows:

\[ Y_{ij} = m + a_i + E_{ij} \]

Where;

- \( Y_{ij} \) = response (DLWG, DFI, CP and DM digestibility, TCA soluble proteins, index of mitosis, UDPGT activity);
- \( m \) = overall mean;
- \( a_i \) = effect of diet;
- \( E_{ij} \) = residual error.

Induction weight was used a covariate in the analysis of daily live weight gain. Analyses were carried using the PROC GLM procedure (SAS 2000).
Results

Effect of leaf meal inclusion on feed intake, daily liveweight gain and feed conversion ratio

Pigs fed on 10% *Acacia karroo* had the highest feed intake (*P*<0.05). Daily feed intake for pigs fed on *Acacia nilotica, Colophospermum mopane* was similar to pigs fed on the control diet (*P>*0.05). Although the growth rate of *Acacia karroo* fed animals (g/day) tended to be higher than the rest, this was not statistically significant (*P>*0.05). There were no significant differences in the FCR and cold dressed mass for pigs fed on diets containing the leaf meals and those on the control diet (*P>*0.05) (Table 2).

Table 2. Effect of low dietary inclusion levels of leguminous leaf meals on pig performance (least square means)

<table>
<thead>
<tr>
<th></th>
<th>Control</th>
<th>Acacia karroo</th>
<th>Colophospermum mopane</th>
<th>Acacia nilotica</th>
<th>SEM</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td>DFI</td>
<td>2312.9b</td>
<td>2622.5a</td>
<td>2373.6b</td>
<td>2471.8b</td>
<td>98.64</td>
<td>*</td>
</tr>
<tr>
<td>ADLWG</td>
<td>1039.3</td>
<td>1080.4</td>
<td>1003.6</td>
<td>987.5</td>
<td>42.69</td>
<td>NS</td>
</tr>
<tr>
<td>FCR</td>
<td>2.2</td>
<td>2.4</td>
<td>2.4</td>
<td>2.5</td>
<td>0.10</td>
<td>NS</td>
</tr>
<tr>
<td>CDM</td>
<td>72.1</td>
<td>73.4</td>
<td>73.0</td>
<td>73.9</td>
<td>0.95</td>
<td>NS</td>
</tr>
<tr>
<td>CPD</td>
<td>93.4a</td>
<td>84.2c</td>
<td>83.5c</td>
<td>88.6b</td>
<td>0.39</td>
<td>**</td>
</tr>
<tr>
<td>DMD</td>
<td>89.7a</td>
<td>85.9b</td>
<td>82.7b</td>
<td>89.8a</td>
<td>1.44</td>
<td>*</td>
</tr>
</tbody>
</table>

*Systemic effects*

<p>| | | | | |</p>
<table>
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<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>MPRP</td>
<td>0.8</td>
<td>1.0</td>
<td>0.8</td>
<td>1.4</td>
</tr>
<tr>
<td>PPRP</td>
<td>0.8c</td>
<td>2.5b</td>
<td>2.6b</td>
<td>3.5a</td>
</tr>
<tr>
<td>UDPGTA</td>
<td>1251.2d</td>
<td>1456.6c</td>
<td>1651.2b</td>
<td>2232.0a</td>
</tr>
</tbody>
</table>

ADLWG - average daily live weight gain (g/day); DFI - daily feed intake (g/day); FCR - feed conversion ratio (g feed/g gained); CDM - cold dressed mass (%); CPD - crude protein digestibility; DMD - dry matter digestibility; PPRP - parotid proline-rich proteins (mg/g gland tissue); MPRP - mandibular protein rich proteins (mg/g gland tissue); UDPGTA - uridine diphosphate glucuronyl transferase activity (A/ mg protein).

*a,b,c* LSMs in the same row with different superscripts are significantly different (*P*<0.05); NS - not significant; *P*<0.05; **P*<0.01.

Crude protein digestibility was significantly higher in the diet containing *Acacia nilotica* than diets containing either *Acacia karroo* or *Colophospermum mopane* (*P*<0.05). The control diet, however, had a higher crude protein digestibility (*P*<0.05) than for diets containing leaf meals. Dry matter digestibility for the diet containing *Acacia nilotica* was similar (*P>*0.05) to the
Growth performance of pigs fed on diets containing Acacia karroo, Acacia nilotica and Colophospermum mopane leaf meals

control diet, and was higher than for diets containing either Acacia karroo or Colophospermum mopane (Table 2).

**Effect of leaf meal inclusion on production of TCA soluble proteins and induction of liver enzymes**

The mean amount of mandibular TCA soluble proteins was 1.0 mg/g gland tissue. There was no significant effect of feeding leguminous leaf meal supplemented diets on mandibular TCA soluble proteins ($P>0.05$). *Acacia nilotica* induced higher levels of parotid TCA soluble proteins than the other supplements ($P<0.05$), which, in turn, induced higher levels of TCA soluble proteins than the control diet ($P<0.05$). There was a four-fold increase in parotid proline rich proteins in the diet containing *Acacia nilotica* than the control diet. Diets containing leaf meals led to the production of more parotid proline rich proteins than the control diet ($P<0.05$). No differences were observed in the parotid PRP production between pigs that consumed diets containing *Acacia karroo* and *Colophospermum mopane* ($P>0.05$). The control diet had the least UDPGT activity while *Acacia nilotica* had the highest ($P<0.05$). The UDPGT activity was ranked as Control $<$ *Acacia karroo* $<$ *Colophospermum mopane* $<$ *Acacia nilotica* ($P<0.05$) (Table 2).

There was no significant effect of feeding leaf meals on the index of mitosis in the liver, kidney and the small intestine ($P>0.05$) (Table 3).

| Table 3. Effect of feeding low levels of leguminous leaf meals in the diet on the index of mitosis in the small intestine, liver and kidney of pigs |
|---|---|---|
| **Diet** | **Small intestine** | **Kidney** | **Liver** |
| Control | 1.9 | 3.2 | 2.7 |
| *Acacia karroo* | 1.7 | 2.5 | 2.3 |
| *Acacia nilotica* | 1.6 | 2.6 | 2.5 |
| *Colophospermum mopane* | 2.5 | 2.7 | 2.2 |
| s.e.m. | 0.61 | 0.71 | 0.30 |
| Significance | NS | NS | NS |

*NS - not significant (P>0.05)*

**Discussion**

The value of locally available leguminous leaf meals on pig performance has not been determined. These leaf meals have the potential to partially replace the conventional protein sources, such as
soyabean and sunflower, which are expensive. Higher levels of intake of these leaf meals been shown to depress digestibility of nutrients and to increase the activity of enzymes that are responsible for detoxification of toxic substances in the body (Halimani et al 2005).

The increased voluntary feed intake of animals fed *Acacia karroo* agrees with D'Mello (1995) who observed a similar pattern when feeding 10% *Leucaena leucocephala* leaves to pigs. Further increases of leaf meals have, however, been shown to depress intake. For example, Ravindran (1990) observed a decline in intake in pigs fed on incremental levels of *Manihot esculenta*. Twenty percent *Leucaena leucocephala* fed to growing pigs also markedly depressed intake (D'Mello 1995; Laswai et al 1997). In this study, the intake of pigs fed on *Acacia nilotica* and *Colophospermum mopane* were not different from pigs fed on the control diet. The reported differences in the intake of diets containing leaf meals could be due to differences in the basal diets used and in the nature of the leaf meal supplements. A rise in intake in pigs fed on *Acacia karroo* could be due to unavailability of essential amino acids in the leaf meal as a result of interacting with fibre or phenolic constituents leading to the pigs eating more in order to compensate (Forbes 1995; MacDonald et al 1995). Amino acid side chains containing -SH and -NH₂ groups such as methionine and lysine have a higher tendency to form covalent bonds with phenolics. In addition, Halimani et al (2005) observed that *Acacia karroo* contains five times more CP-bound phenolic compounds than *Acacia nilotica*. Fibre-bound phenolics were also higher in *Acacia karroo* than in *Colophospermum mopane*. Using tannic acid as a standard, the radial diffusion assay showed that *Acacia karroo* had twice as much phenolic compounds than in *Colophospermum mopane* (Halimani et al 2005). Several studies have reported a reduction in intake in pigs fed on phenolic-containing diets (Laswai et al 1997; Halimani et al 2005). In addition, other substances have been reported to reduce intake of diets containing leaf meals.

The daily live weight gain of pigs fed on *Acacia nilotica*, *Acacia karroo* and *Colophospermum mopane* was not different from control. This may be due to inadequate replication in this experiment. Literature indicates a general decline in the daily live weight gain of pigs fed low inclusion levels of leaf meals (Mtenga and Laswai 1994; Laswai et al 1997; Halimani et al 2005). Other reports have indicated an increase in daily live weight gain in pigs fed on low inclusion levels of leaf meals (D'Mello 1995; Halimani et al 2005). Pigs fed diets including leaf meals have been shown to have low nitrogen retention, which could explain the decline in weight gain (Phuc et al 2000; Cheverria et al 2002). The feed conversion ratio, though not significantly different from control, had a tendency to be higher for the diets containing the leaf meals compared to control. Laswai et al (1997) reported a depression in feed conversion efficiency in pigs fed 20 % *Leucaena leucocephala*. Reduced nitrogen retention (Cheverria et al 2002) is a result of a reduction in crude protein digestibility and an increase in loss of endogenous protein (Jansman et al 1995; Lindberg and Andersson 1998; Phuc et al 2000).

There was a reduction in the dry matter and crude protein digestibility in pigs fed low inclusion levels of leaf meals compared to the conventional control diet. This is consistent with literature (Laswai et al 1997; Ly et al 1998; Cheverria et al 2002). A reduction in dry matter digestibility can result from an increase in the flow of digesta and total tract excretion of nutrients and energy as a result of the higher insoluble fibre content of leguminous leaf meals (Lindberg and Cortova 1995). Low digestibility of protein may be due to protein being bound by polyphenols and fibre
or physically entrapped by fibre in the leaf meals (Phuc et al 2000). It may also be due to an increase in hindgut fermentation leading to higher loss of bacterial nitrogen or due to enhanced secretion of endogenous protein coupled with reduced degradation and reabsorption of endogenously secreted protein (Jansman et al 1995; Lindberg and Andersson 1998).

There was a significant increase in the production of TCA-soluble proteins from the parotid salivary glands of pigs fed diets including leaf meals but not in the mandibular gland. There is tissue specific expression of the genes that code for salivary proline rich proteins (Bennick 2002). Pigs have been reported to produce proline rich proteins in their salivary glands as a normal part of the secretions from these glands (Mole et al 1990; Patamia et al 2005). In some animal species, feeding diets containing polyphenols increases the production of these proteins that bind the polyphenols as a sacrifice for both dietary protein and other endogenous proteins (Mehansho et al 1992). A direct measurement of proline-rich proteins in this study showed an increase of approximately three times the basal rate of secretion, which is lower than the increase recorded for rats (Mehansho et al 1992). Grala et al (1993) reported a reduction in the apparent total tract disappearance of several amino acids when feeding pigs diets containing tannins. The amino acids that showed the highest reduction were proline (-31.8 units), cystine (-17.2 units) and glycine (-16.4 units). Proline and glycine constitute 61% of salivary proline rich proteins in the pig (Mole et al 1990). Jansman et al (1995) reported that tannin associated proteins in the ileum of pigs fed faba bean hulls were markedly higher in content of proline, alanine, glycine but low in glutamic acid compared to dietary protein, indicating that these amino acids are derived from endogenous proteins. Halimani et al (2005) reported data on the direct determination of salivary proline rich proteins from pigs fed high tannin diets using polyacrylamide gel electrophoresis. Pigs were observed to increase their production of PRPs with apparent molecular weights of 24 600, 54 000, 66 000, and 74 000 dalton. These findings indicate that production of proline rich proteins, though significant, may not the main method of defence against tannins in the pig as the increase in secretion above basal rate is not as high as in other species.

The increase in the activity of hepatic microsomal UDPGT in pigs fed diets containing leaf meals is consistent with literature (Makkar 1993; Halimani et al 2005). Feeding high tannin diets increases the activity of enzymes involved in detoxification. This shows that some phenolics are absorbed and exert systemic effects. The highest increase was recorded for Acacia nilotica whose polyphenolics are catechin gallates (Mueller-Harvey et al 1986), which may be more easily absorbed, compared to proanthocyanidins.

There was no significant effect of feeding leaf meals on the index of mitosis in the liver, kidney and small intestine. This indicates that the leaf meals and their constituents do not have a major effect on the gross turnover of cells in these organs and is consistent with literature reports (Van Leeuwen et al 1995).
Conclusions

- It is concluded that feeding leaf meals at low (10%) inclusion levels in the diet of pigs leads to a depression in digestibility of nutrients, an increase in endogenous protein secretion and an increase in the activity of liver enzymes.

- Inclusion of leaf meals in the fattening diets of pigs did not reduce growth rate and is, therefore, a potentially feasible technology that farmers can include in their pig production strategies.

Acknowledgements

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