Different levels of macadamia oil cake meal, and wood ash vs. feed lime as dietary sources of calcium on bone characteristics of slow-growing chickens

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Abstract

A study was conducted to evaluate the potential of macadamia oil cake meal (MOCM) and wood ash as feed ingredients for poultry under subsistence farming conditions. In this article, the effect of these ingredients on bone characteristics is reported. Two hundred and eighty eight day-old New Hampshire chickens were used in the study. The research was conducted as a 3 x 2 factorial design, and 48 chicks were randomly allocated per treatment. Three basic diets were formulated: one without MOCM and the other two containing 10% and 50% MOCM. The MOCM contained 132 g/kg of crude protein, 228 g/kg crude fat and 365 g/kg crude fibre on an 'as-fed' basis. Each of these three treatments was split into two: one receiving feed lime ($CaCO_3$) as the main source of calcium; and the other wood ash, which contained 257 g Ca/kg. All diets contained a Ca level of *ca*. 10 g/kg. The chickens received the experimental diets from 2 to 15 weeks of age. After week 15, eight chickens per treatment were killed and their right legs removed at the femorotibial articulation and frozen for later evaluation. Between Ca sources there were no significant differences in tibia weight, diameter, volume, density and breaking strength. The ash content and Ca, P and Mg concentrations in bone ash between Ca sources were similar. However, in the two Ca diets containing 50% MOCM the Ca and P concentrations of the tibiae were significantly lower than in the diets containing lower levels of MOCM. It was concluded that wood ash was as effective as feed lime in supplying Ca to chickens. However, some practical problems in the feeding of wood ash became apparent, such as that wood ash is a fine powder and does not mix well with other ingredients, except when the oil content of the diet is high, as with the treatments containing MOCM.

Keywords: Bone ash, bone strength, feed lime, subsistence farming, tibia
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Introduction

It would be advantageous to resource-poor poultry farmers under subsistence farming conditions if cheap and readily available feedstuffs could be identified to be included in the diets of their chickens.

Macadamia oil cake meal (MOCM), the residue after removal of the oil from the nuts, is a so-called opportunity feedstuff that would be accessible to subsistence farmers in the vicinity of macadamia processing plants. Limited research has apparently been done on the value of MOCM as a feedstuff for livestock, though Skenjana *et al*. (2006) demonstrated that MOCM has a relatively high *in vitro* digestibility and *in situ* degradability in sheep. Since macadamia nuts are readily consumed by human beings, it could be assumed that the cake meal should also be an ingredient that could be included successfully in poultry diets. The MOCM used in this experiment had high levels of crude fibre (385 g/kg DM) and crude fat (243 g/kg DM), while being a moderate source of crude protein (140 g/kg DM) (Phosa, 2009).

Investigations on the utilisation of wood ash in agriculture and forestry indicated that wood ash can serve as a liming agent and its application to the soil is a convenient way to recycle exported nutrient elements (Cronan & Grigal, 1995; Demeyer *et al*., 2001). Mineral supplements for livestock are not always readily available to resource-poor farmers under subsistence farming conditions. However, in many regions of the world, wood ash is accessible to subsistence farmers. There has been limited research conducted on the potential of wood ash as a mineral source for animals, though wood ash showed promise as a mineral supplement to sheep in the tropics (Imbeah, 1999).

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Wood ash is the inorganic residue that remains after the combustion of wood. Ash content and chemical composition vary depending on tree species, soil type and climate, as well as the temperature at which the wood was combusted (Campbell, 1990). In general, wood from trees originating in temperate climates yields 1 - 10 g ash/kg dry matter (DM), while tropical and subtropical woods yield up to 50 g/kg DM. Hardwoods, in general, contain more ash than softwoods (Campbell, 1990).

It is well documented that wood ash is rich in calcium (Ca). Campbell (1990) reported that wood ash contains between 80 and 330 g Ca/kg, while Naylor & Schmidt (1986) reported concentrations of 132 to 321 g/kg. In a survey of the Ca content of ash from nine tree species in South Africa, Van Ryssen & Ndlovu (2003) recorded that the Ca content of the wood (without the bark) ranged from 91 to 318 g Ca/kg ash, while the Ca content of ash from the bark of these trees ranged from 283 to 337 g Ca/kg. The Ca is predominantly in the form of $CaCO_3$ (Demeyer *et al*., 2001; Ndlovu, 2007).

In formulating poultry diets, the inclusion of Ca sources is usually necessary to meet the Ca needs of chickens. It was hypothesized that the Ca in wood ash is readily available to poultry and can replace the Ca in feed lime ($CaCO_3$), which is most commonly used as Ca source in poultry feeds. In this article, the effects on bone characteristics are reported when different dietary levels of MOCM and two sources of Ca (feed lime and wood ash) were compared.

Material & Methods

Two hundred and eighty eight day-old New Hampshire chickens were obtained from "Fowls for Africa" at the ARC-Livestock Business Division (Pretoria, South Africa). The New Hampshire breed was chosen because it is a relatively slow-growing chicken, typical of what is found in subsistence farming situations in southern Africa. The research was conducted as a 3 x 2 factorial design and 48 unsexed chicks were randomly allocated to treatments, with four replications of 12 chicks per replicate. Three basic diets were formulated: one without MOCM, and the other two containing 10% and 50% MOCM. Each of these treatments was split into two, with one receiving feed lime ($CaCO_3$) and the other wood ash as the sources of Ca. A least cost feed formulation programme (Format International, UK) was used to formulate the six treatment diets (Tables 1 and 2) according to the NRC (1994) specification for poultry starter and grower diets. All the treatments received diets with a Ca level of *ca*. 10 g/kg.

Macadamia oil cake was obtained from the Royal Macadamia Processing Plant (Thohoyandou, South Africa) and prepared as described by Phosa (2009). Dry wood from a variety of tree species was collected on the experimental farm of the University of Pretoria, Hatfield, Pretoria, South Africa (coordinates 25°15'28.9"E, 25°45'03.6" S). The wood was burned in a furnace used for incinerating carcasses, and the ash was collected and sifted through a 1 mm sieve to remove any coarse material. A sample of the ash was taken for laboratory analysis. The Ca concentration in the wood ash was determined in advance to be used in the formulation of the experimental diets.

The study, approved by the Ethics Committee of the Faculty of Natural and Agricultural Sciences of the University of Pretoria (ethical clearance number: EC 030722-021), was conducted in an environmentally controlled broiler house at the experimental farm of the University of Pretoria. Inside the broiler house the replicate groups of chickens were placed randomly in pens (3 x 1.5 m) enclosed with chicken mesh. Shavings were used as litter material. The chickens received the experimental diets from 2 to 15 weeks of age. Feed and water were provided *ad libitum*. Light was provided for 23 h per day and infrared bulbs were used to supply heat. Room temperature was decreased from 32 °C during the early stages to 22 °C from day 21 onwards. The chickens were vaccinated according to the standard vaccination programme for the experimental site.

At the end of the growth trial at 15 weeks, two cockerels per replicate, giving eight per treatment, were killed humanely by cervical dislocation. The right leg from each chicken was removed at the femorotibial articulation, labelled, wrapped and stored in a freezer at −20 °C until the bone strength study commenced. For this, the legs were thawed, and boiling water was used to loosen the flesh whereupon adhering tissues were removed in order to extract the tibiae. The tibiae were further cleaned, the cartilage caps were removed, and the bone was wrapped in parafilm and placed in a refrigerator. The tibiae were weighed and their volume obtained through the displacement of water in a measuring cylinder. Before the breaking strength test was done, the bones were dried overnight in an oven at a temperature of between 60 and 80 °C. The tibiae were weighed in the air and in water to obtain bone density. Measurements of the major and minor exterior diameters were taken at the thinnest part of the tibia diaphysis with a veneer calliper (150 x 0.02 mm, Tricle Brand, Shangai, China).

The strength of the tibia was determined with an Instron Universal Testing machine (Model No. 1011, Instron Corporation, Canton, MA, USA) using the standard bending test (ASAE, 1993). Each tibia was placed horizontally on a three-point bend fixture. The length of the tibia was measured in order to determine the centre where the plunger should be in contact with the bone for breaking. The plunger mounted on the crosshead of the testing machine was adjusted until it touched the bone at the centre. A set crosshead speed of 30 mm/min was used. After determination of breaking strength, the broken bones were crushed further and defatted in a Shoxhlet apparatus using petroleum ether. The bones were then dried, ground and stored for chemical analysis.

Table 1 Ingredient composition of the starter diet (g/kg, as fed)

Ingredients	Treatment					
	0%MOCM		10%MOCM		50%MOCM	
	Lime	Wood	Lime	Wood	Lime	Wood
Yellow maize	499	480	528	530	250	240
Wheat bran	132	140	110	110	66	70
Soya oilcake	188	183	152	155	94	92
Sunflower oilcake	80	81	97	91	40	41
Fish meal	73	73	80	81	36	36
Macadamia oilcake	0	0	110	110	483	483
Limestone	19	0	23	0	19	0
Wood ash	0	33	0	26	0	27
Salt	10	10	10	10	10	10
Vitamin & mineral premix*	3	3	3	3	3	3

MOCM: macadamia oil cake meal; Lime: limestone; Wood: wood ash;* no phytase included.

Table 2 Ingredient composition of the grower diet (g/kg, as fed)

Ingredients	Treatment					
	0%MOCM		10%MOCM		50%MOCM	
	Lime	Wood	Lime	Wood	Lime	Wood
Yellow maize	551	584	659	657	276	292
Wheat bran	321	168	0	0	110	84
Soya oilcake	111	120	143	140	56	60
Fish meal	86	86	76	78	43	43
Macadamia oilcake	0	0	97	97	493	493
Limestone	22	0	16	0	11	0
Wood ash	0	33	0	19	0	17
Salt	10	10	10	10	10	10
Vitamin & mineral premix*	3	3	3	3	3	3

MOCM: macadamia oil cake meal; Lime: limestone; Wood: wood ash; * no phytase included.

The Ca, total P and Mg levels in the dry fat-free bone powder were measured. The bone was also incinerated using a muffle furnace at 600 °C for 8 h to determine the ash content.

The following chemical analyses were performed on the bone, wood ash, MOCM and mixed rations according to the methods described in AOAC (2000): calcium and Mg, using atomic absorption spectrophotometry (Perkin Elmer 2380 Model), total P (method 968.08) and dry matter (DM) (method 934.01). Crude protein (CP) (method 968.06), ether extract (method 920.39) and crude fibre (CF) (method 962.09) of the MOCM and mixed rations were also measured. The crystalline forms of the metals in the wood ash were measured using the standard powder x-ray diffraction (XRD) technique on a Siemens D-501 automated diffractometer (Chung, 1974).

Data were analysed statistically as a randomized block design with the GLM model (SAS, 2004) for the average effects over time. Repeated measures analysis variance of the GLM model was used for repeated week or period measures. Means and standard deviations were calculated and significance of differences (P <0.05) between means was determined by the Fischer's test (Samuels, 1989). The linear model used, is described by the following equation:

$$Y = \mu + T_i + B_j + e$$

Where Y = variable studied during the period
μ = overall mean of the population
T = effect of the ith treatment
B = effect of the jth block
e = error associated with each Y.

Results and Discussion

The chemical composition of the MOCM used in this study (Table 3) indicated that the MOCM contained 132 g CP/kg. This is lower than the mean of 209 ± 53.3 g CP/kg DM (n = 8), reported by Skenjana (2011). Despite the fact that the oil was extracted, the MOCM still contained a relatively high concentration of crude fat (228 g/kg), and the total P concentration of 2.3 g/kg was higher than that of Ca, at 1.6 g/kg.

Table 3 Chemical composition of macadamia oil cake meal

Nutrients	g/kg (as fed)
Dry matter	940
Ash	28
Crude protein	132
Crude fibre	365
Crude fat	228
Calcium	1.6
Total phosphorus	2.3

The wood ash in the present study contained 257 g Ca/kg, which is in agreement with results reported by Demeyer *et al*. (2001) and Van Ryssen & Ndlovu (2003). The crystalline forms of the elements in the wood ash used in the study are presented in Table 4. According to the XRD analysis, the wood ash contained 58.8% calcite ($CaCO_3$), a crystalline form similar to that in limestone. This is followed by periclase

Table 4 Approximate proportions of crystalline forms (XRD analysis) of minerals in the wood ash

Crystalline form[1]	Symbol	Percentage
Calcite	$CaCO_3$	58.8
Periclase	MgO	19.4
Hydroxylapatite	$Ca_5(PO_4)_3(OH)$	7.4
Fairchildlite	$K_2Ca(CO_3)_2$	6.5
Lime	CaO	2.2
Ankerite	$Ca(Fe{+}_2,Mg)(CO_3)_2$	1.8
Quartz	SiO_2	1.4
Halite	KO_4NaO_6Cl	1.3
Sylvite	KCl	1.2

[1] Terminology as used in geology.

(MgO) at a concentration of 19.4%. Other minerals include hydroxylapatite ($Ca_5(PO_4)_3(OH)$) and sylvite (KCl) at concentrations of approximately 7.4 and 1.2%, respectively. However, it should be pointed out that Olanders & Steenari (1995) demonstrated that the crystalline phases in ash depend on the temperature of the ashing process.

The comparable concentrations of Ca in wood ash and limestone were 257 and 380 g/kg, respectively. This necessitated a higher inclusion rate of wood ash in the diet than with feed lime to replace feed lime with wood ash, resulting in a greater dilution of other nutrients in the ash diets. Using relatively slow growing chickens, this higher dilution apparently did not affect the performance of the chickens (Phosa, 2009).

The chemical composition of the experimental diets is presented in Tables 5 and 6. The diets contained *ca*. 10 g Ca/kg. The fat concentration in the diets ranged between 40 and 60 g/kg for the 0% and 10% MOCM diets and *ca*. 80 g fat/kg for the diets containing 50% MOCM, except for the 50% MOCM plus wood ash treatment in the starter diet, which contained 98 g fat/kg feed.

Table 5 Chemical composition (g/kg) of the different treatments of the starter diets (as fed, full fat basis)

	Treatment					
	0% MOCM		**10% MOCM**		**50% MOCM**	
	Lime	**Wood**	**Lime**	**Wood**	**Lime**	**Wood**
Crude protein	211	231	225	226	189	182
Fat	45	42	52	54	79	98
Crude fibre	62	67	92	88	220	224
Calcium	12	10	11	10	10	10
Total phosphorus	5.2	7.0	6.0	6.1	4.3	4.0

MOCM: macadamia oil cake meal; Lime: limestone, Wood: wood ash.

Table 6 Chemical compositions of different treatments of the grower diets (as fed, full fat basis)

	Treatments					
g/kg	**0% MOCM**		**10% MOCM**		**50% MOCM**	
	Lime	**Wood**	**Lime**	**Wood**	**Lime**	**Wood**
Crude protein	190	190	182	180	180	160
Fat	33	30	41	40	80	81
Crude fibre	41	40	52	50	164	190
Crude ash	74	73	64	62	72	80
Calcium	11	10	9	8	13	13
Total phosphorus	6	7	5	5	4	5
Magnesium	2	3	2	2	2	4
ME (MJ/kg)	12.3	12.2	12.5	12.6	11.7	11.4

MOCM: macadamia oil cake meal; Lime: limestone; Wood: wood ash.
ME: metabolisable energy (calculated).

Tibia weight, volume, density breaking strength and shaft diameters are presented in Table 7. No significant differences ($P > 0.05$) were recorded between these measurements. Bone breaking strength as a factor of Ca availability and deposition from the source provided to the chickens was the same for both limestone and wood ash, although it showed a tendency to be lower at the 50% MOCM than at the other treatments. The results suggest that wood ash can be used as an alternative for limestone. This finding is in agreement with Waldenstedt (2006), who stated that Ca has high availability in most sources.

The chemical composition of the tibiae of birds on the MCOM treatments is presented in Table 8. Bone ash as percentage of dry, fat-free bone ranged from 60.8% to 64.4%, in line with values reported by Zhang & Coon (1997) and Field (2000). In the present study the ash percentage in the dry, fat-free tibiae from the chickens on the 50% MOCM plus wood ash was lower (P <0.05) than those of the other treatments. This corresponds with lower concentrations of Ca and P in the tibiae in this treatment compared to the other treatments, except that the Ca and P concentrations in the 50% MOCM plus feed lime was also lower (P <0.05) than those in the diets containing 0% and 10% MOCM. This is furthermore supported by the lower (P <0.05) percentage of P in the bone ash of both the treatments containing 50% MOCM, and the lower (P <0.05) Ca% in the bone ash in the 50% MOCM plus feed lime (Table 8). It is evident from these results that at an inclusion rate of 50% MOCM concentrations of Ca and P in the bone were depressed.

Table 7 Bone characteristics of the right tibia bone of chickens at 15 weeks of age fed different calcium sources and levels of macadamia oil cake meal (MOCM)

Tibia[1] characteristics	0% MOCM		10% MOCM		50% MOCM		SE
	Lime	Wood	Lime	Wood	Lime	Wood	
Weight (g)	5.7	5.8	5.1	5.5	5.8	5.3	0.30
Volume (L)	4.3	4.7	4.2	4.4	4.6	4.3	0.31
Density (g/L)	1.4	1.2	1.2	1.3	1.3	1.2	0.041
Breaking strength (N)[1]	−235	−234	−236	−241	−221	−169	27.8
Diameter major (mm)	0.62	0.65	0.65	0.61	0.63	0.58	0.024
Diameter minor (mm)	0.27	0.28	0.27	0.28	0.34	0.25	0.034

Lime: feed lime ($CaCO_3$); Wood: wood ash. [1] N: Newtons.
Differences between means within rows were statistically insignificant (P >0.05).

Table 8 Effect of dietary level of macadamia oil cake meal (MOCM) and calcium source on the means (g/kg) (SEM) of the elemental composition of tibia bone (dry, fat-free basis) and as percentage of bone ash

Tibia characteristics	0% MOCM		10% MOCM		50% MOCM		SE
	Lime	Wood	Lime	Wood	Lime	Wood	
Proportion of dry, fat-free bone (g/100 g)							
Bone ash	63.3^{a}	62.9^{a}	64.4^{a}	63.6^{a}	63.6^{a}	60.8^{b}	0.59
Calcium	22.5^{ab}	22.0^{b}	22.8^{a}	21.9^{b}	21.3^{c}	21.2^{c}	0.18
Phosphorus[1]	11.1^{a}	11.3^{a}	11.6^{a}	11.3^{a}	10.2^{b}	10.2^{b}	0.15
Magnesium	0.40	0.41	0.40	0.42	0.40	0.40	0.01
% of bone ash							
Calcium	35.4^{a}	34.9^{ac}	35.4^{a}	34.5^{c}	33.5^{b}	34.9^{ac}	0.27
Phosphorus[1]	17.6^{a}	18.0^{a}	17.9^{a}	17.7^{a}	16.0^{b}	16.8^{b}	0.23
Magnesium	0.64	0.66	0.61	0.66	0.63	0.66	0.01

[abc] Means within rows with different superscripts are significant at P <0.05. SE: standard error.
[1] Interaction between phosphorus concentration and level of MOCM significant at P <0.05.
Lime: feed lime ($CaCO_3$); Wood: wood ash.

When considering the combined effects of Ca source (Table 9) on the percentage ash and Ca in the dry fat-free bone, the percentages were lower in the wood ash treatments than in the feed lime treatments. However, no differences between Ca sources were measured when the Ca, P and Mg were expressed as percentages of bone ash (Table 9). Thus, Ca levels in the tibiae were lower when the chickens were fed wood ash, but the mineral composition of the ash itself stayed constant between treatments.

Waldenstedt (2006) pointed out that different factors can affect Ca and P absorption in chickens, including chelating agents and mineral interactions, gastrointestinal tract pH, and interactions with dietary protein, fat and carbohydrates. Furthermore, feed sources of plant origin often contain anti-nutritional substances including phytate, which contains P, which is unavailable for absorption, and oxalic acid, which depresses Ca absorption (Woyengo & Nyachoti, 2011). In this investigation, the diets were not tested for anti-nutritional substances, though MOCM might have contained some, which could have affected the Ca and/or P absorption at the high level of MOCM inclusion in the two diets. Furthermore, Atteh & Leeson (1984) reported that at elevated dietary fat levels, a decrease in Ca absorption can occur with unavailable Ca soaps being excreted through the faeces. In the present study the fat content ranged between 40 and 60 g/kg DM in the 0% and 10% MOCM diets, but was 80 and 98 g/kg for the 50% MOCM plus limestone and 50% MOCM plus wood ash, respectively. Therefore, the relatively high fat content in the 50% MOCM treatments diets could have caused a decrease in Ca absorption leading to lower levels of Ca and P in bone ash of these two treatments, irrespective of the Ca source.

Table 9 Combined effects of the calcium sources on the chemical composition of the dry fat-free bone and the bone ash

Chemical composition	Feed lime	Wood ash	SE
Dry, fat-free bone			
Bone ash (%)	63.8^{a}	62.4^{b}	0.34
Calcium (%)	22.2^{a}	21.7^{b}	0.11
Phosphorus (%)	11.0	10.9	0.08
Magnesium (%)	0.40	0.41	0.005
Bone ash			
Calcium (%)	34.8	34.8	0.29
Phosphorus (%)	17.2	17.5	0.23
Magnesium (%)	0.63	0.66	0.01

^{ab} Means within rows with different superscripts are significant at P <0.05.
SE: standard error.

In southern Africa, cereal grains such as maize and sorghum form a large proportion of the diet of people living under subsistence conditions. These cereals contain low concentrations of Ca (Bredon *et al*., 1987). Scavenging poultry would have access to leftovers from human meals, and if that constitutes a large proportion of the diets of the chickens, they may suffer from a Ca deficiency. Likewise, cassava roots that are fed to livestock in subsistence farming situations contain very low levels of Ca (Oke, 1978; Smith, 1988). This demonstrates that there is a potential need for supplementing Ca to chicken diets under subsistence farming conditions and evidence from the present study suggests that wood ash would be an easily accessible source of Ca.

However, although the utilization of the Ca in wood ash did not differ from that of Ca in feed lime, there seems to be some limitations in using wood ash as a Ca source in subsistence farming situations. These include the fact that effective methods of presenting the ash to the birds could be a problem, since it is unlikely that fowls would consume fine ash as such. In the 0% MOCM plus wood ash treatment in the present study, it was observed that the ash which was in a fine powder form, tended to separate from the other feed ingredients. This did not occur in the other treatment diets containing MOCM, probably because of the relatively high fat content that was in an oily form, which probably prevented such a separation. Despite this separation it did not affect the measurements as evidenced by bone strength, suggesting that the chickens consumed sufficient wood ash to meet their requirements. However, this does indicate that effective methods of retaining the wood ash in other diet ingredients would be required to prevent the separation of the fine ash particles.

Conclusion

Data from the present study suggested that at a 50% inclusion of MOCM in the broiler diet, the mineralization of the bone would be affected. On the other hand, wood ash was found to be an effective substitute for feed lime in supplying Ca in poultry diets. However, some practical problems in the feeding of wood ash became apparent, such as, that wood ash is a fine powder and does not mix well with other ingredients except when the oil content of the diet is high, as was the case with the treatments containing MOCM.

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References

AOAC, 2000. Official methods of analysis. (17^{th} ed.). Association of Official Analytical Chemists, Inc., Maryland, USA.

ASAE, 1993. Bone, shear and three point bending, S349. ASAE standards 40, 462463.

Atteh, J.O. & Leeson, S., 1984. Effects of dietary saturated or unsaturated fatty acids and calcium levels on performance and mineral metabolism of broiler chicks. Poult. Sci. 61, 2252-2260.

Bredon, R.M., Stewart, P.G. & Dugmore, T.J., 1987. A manual on the nutritive value and chemical composition of commonly used South African farm feeds. Natal Region, Dept. of Agriculture and Water Affairs, South Africa.

Campbell, A.G., 1990. Recycling and disposing of wood ash. Tappi J. (September), 141-146.

Chung, F.H., 1974. Quantitative interpretation of X-ray diffraction patterns of mixtures. I. Matrix flushing method for quantitative multicomponent analysis. J. Appl. Cryst. 7, 519-525.

Cronan, C.S. & Grigal, D.F., 1995. Use of calcium/aluminium ratios as indicator of stress in forest ecosystems. J. Environ. Qual. 24, 209-226.

Demeyer, A., Voundi Kkana, J.C. & Verloo, M.G., 2001. Characteristics of wood ash and influence of soil properties and nutrient uptake: an overview. Biores. Technol. 77, 287-295.

Field, R.A., 2000. Ash and calcium as measures of bone in meat and bone mixtures. Meat Sci. 55, 255-264.

Imbeah, M., 1999. Wood ash as mineral supplement for growing lambs under village conditions in the tropics. Small Rumin. Res. 32, 191-194.

Naylor, L.M. & Schmidt, E.J., 1986. Agricultural use of wood ash as a fertilizer and liming material. Tappi J. (October), 114-119.

Ndlovu, H., 2007. Ash from homestead fireplaces and wood as possible sources of minerals for livestock. M. Inst. Agrar. (Anim. Prod.) thesis, University of Pretoria, South Africa.

NRC, 1994. Nutrient Requirements of Poultry. National Academy Press, Washington, D.C., USA.

Oke, O.L., 1978. Problems in the use of cassava as animal feed. Anim. Feed Sci. Technol. 3, 345-380.

Olanders, B. & Steenari, B-M., 1995. Characteristics of ashes from wood and straw. Biomass & Bioenergy 8, 105-115.

Phosa, M.A., 2009. The nutritive value of macadamia oil cake meal and wood ash as alternative feed ingredients for chickens in rural areas. M.Inst. Agrar. thesis, University of Pretoria, South Africa.

Samuels, M.L., 1989. Statistics for the Life Sciences. Collier MacMillian Publishers, London, UK.

SAS, 2004. Statistical Analysis System user's guide. Release Version 7.00. SAS Institute, Cary, North Carolina, USA.

Skenjana, A., 2011. The potential nutritive value of waste products from the sub-tropical fruit processing industry as livestock feed. MSc Agric. Thesis, University of Pretoria, South Africa.

Skenjana, A., Van Ryssen, J.B.J. & Van Niekerk, W.A., 2006. *In vitro* digestibility and *in situ* degradability of avocado meal and macadamia waste products in sheep. S. Afr. J. Anim. Sci. 36 (5, Suppl.1), 78-81.

Smith, O.B., 1988, Workshop on processing and utilization of cassava by small-scale farmers in Africa, http://www.fao.org/Wairdocs/ILRI/x5458E/x5458eOd.htm

Van Ryssen, J.B.J. & Ndlovu, H., 2003. Wood and homestead ash as potential source of minerals to livestock. Proc. 8th World Conference on Animal Production. October 26-31, Porto Alegre, Brazil. ID 9713, pp. 12-13.

Waldenstedt, L., 2006. Nutritional factors of importance for optimal leg health in broilers: A review. Anim. Feed Sci. Technol. 126, 291-307.

Woyengo, T.A. & Nyachoti, C.M., 2011. Review: Supplementation of phytase and carbohydrases to diets for poultry. Can. J. Anim. Sci. 91, 177-192.

Zhang, B. & Coon, C.N., 1997. The relationship of various bone measurements in hens. Poult. Sci. 76, 1698-1701.