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Short communication

Effect of traditional processing methods on chemical composition and in vitro true dry matter digestibility of the Mophane worm (*Imbrasia belina*)

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Abstract

Effects of traditional processing methods on the nutritional value of Mophane worm (*Imbrasia belina*) were evaluated. Samples were degutted or not degutted. The worms were further subjected to either cooking for 1 h (remaining in boiling water for 45 min), hot ash roasted for 5–7 min or not treated (control). Treatment had an effect on the level of most minerals, on the in vitro true dry matter digestibility (IVTDMD) ($P<0.001$), crude protein (CP) and magnesium (Mg) ($P<0.05$). Fiber components, acid detergent insoluble nitrogen (ADIN) and manganese (Mn) and available CP (CP_{av}) were not affected ($P>0.05$). Degutting had an effect on CP, fiber components, minerals, ($P<0.001$), ADIN, IVTDMD and Mn ($P<0.01$). Leaves from Mophane vegetation in the undegutted Mophane worm samples diluted levels of CP, ADIN, Zn, Mn and IVTDMD but increased the levels of ash, fibre components, Ca, and P. Mg and CP_{av} were not ($P>0.05$) affected by degutting. There were no interactions ($P>0.05$) between treatment and degutting for ash CP, $CP_{av,}$ ADF, ADL, ADIN, P, Zn, Mn or Mg. An interaction was observed for NDF ($P<0.01$), Ca ($P<0.001$) and IVTDMD ($P<0.05$). A significant and negative correlation was observed between ADF and IVTDMD ($r=-0.86$, $P<0.001$). CP and IVTDMD were positively correlated ($r=0.88$, $P<0.001$). Therefore, it will not be necessary to degut Mophane worms destined for livestock feeding. High fibre levels in the undegutted Mophane worms would make it necessary to degut worms which are destined for human consumption. Given the scarcity of protein in Botswana it is important to evaluate locally available feed resources as potential livestock feeds.

Keywords: *Imbrasia belina*; Protein; Non degutted; Degutting; Livestock

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1. Introduction

Imbrasia belina (Mophane worm) is the larva of emperor moths (Lepidoptera) (Ditlhogo, 1996; Styles, 1996; Marais, 1996). This species feeds on Mophane trees (*Colophospermum mophane*), hence the worm is confined to areas of mophane woodland (Ditlhogo, 1996; Moruakgomo, 1996). In the semi-arid environments of Botswana, Namibia, South Africa and Zimbabwe, the Mophane worm is relished as a source of protein (Marais, 1996; Moruakgomo, 1996; Styles, 1996). Mophane worms harvested in Botswana are exported to South Africa where they are packaged and retailed or processed into livestock feed (Allotey and Mpuchane, 2003; Mphuchane et al., 2001). Moruakgomo (1996) reported that an average of 1113 tons/year of Mophane worm was exported to South Africa from 1991 to 1994. The amount of Mophane worm utilized locally is not known and harvesters have few markets in Botswana in which to sell their Mophane worms. This is compounded by the fact that there are no products processed from Mophane worms. Although the Mophane vegetation is known to provide nutrition to livestock (Macala et al., 1992; Mosimanyana and Kiflewahid, 1987), the value of the worm as livestock feed has not been tested.

Available data indicate that Mophane worms contain high levels of crude protein (CP) (47.5% by van Voorthuizen (1976), 55.3% by Sekhwela (1989) and 55.8% by Ohiokpehai et al. (1996) and O.R. Madibela, unpublished data); as well as high concentrations of calcium and phosphorous (O.R. Madibela, unpublished data; Sekhwela, 1989), and crude fat (Ohiokpehai et al., 1996; O.R. Madibela, unpublished data; Sekhwela, 1989). The profile of essential amino acids of Mophane worm was determined by Ohiokpehai et al. (1996), and is comparable to soyabean and fishmeal. Mophane worm contains greater concentrations of threonine, valine, phenylalanine, tryptophan than soyabean or fishmeal. Lysine and methionine levels, which are important in poultry nutrition, are comparable with the amounts in fishmeal. Zinzombe and George (1994) analysed Mophane worms for fatty acids and found that unsaturated fatty acids, oleic, linoleic and linolenic comprised 47.7% of total fatty acids. The primary source of conjugated linoleic acids (CLA) for human is food products from ruminants (Bauman et al., 2000). Foods rich in CLA have been suggested to have positive health benefits in humans, especially as anticarcinogens (detailed review: McGuire and McGuire, 2000). Therefore, manipulation of ruminant diets through supplementary feeding of diets rich in unsaturated fatty acids would improve the concentration of CLA and hence the nutritional benefits of milk and meat (Kott et al., 2003). Feeding Mophane worms to ruminants may serve this purpose. However, most of the samples that generated the available data were sourced from harvestors and it is not clear how the worms were processed. It is possible that different ways of processing Mophane worms (i.e. roasting, cooking or degutting) by harvestors may have an effect on the nutritional attributes. This study was carried out in order to find the effect of traditional processing on the chemical composition and in vitro true dry matter digestibility (IVTDMD) of Mophane worms.

2. Material and methods

2.1. Sampling site

Botswana is a semi-arid country where mean rainfall ranges from 650 mm in the extreme north east to 250 mm/year in the extreme south west. According to Burgess (2003) a

secondary maximum mean of 550 mm occurs in the higher areas in the south east and a minimum of 300 mm occurs on the lower areas of the Limpopo valley.

Mophane worms were sampled from Tati Siding, south of Francistown City, in the north east of the country. The land is a flat to undulating plain associated with the late erosion cycle. The soil type is classified as moderately deep to very deep, moderately well to slightly excessively drained, strong brown to dark red with sandy loams to clay loams (De Wit and Nachtergaele, 1990). Vegetation is hard veld tree savanna comprising C*olophospermum mopane* and *Acacia nigrescens* woodland (Bekker and De Wit, 1991).

2.2. Sampling

Samples of fresh Mophane worms were taken from Tati Siding. A handful of Mophane worms were obtained from five randomly selected trees using gloves to avoid sharp spines on the surface of the Mophane worm. The samples were pooled together to about 1 kg of sample, on a wet basis. This is because it has previously been found (Madibela and colleagues, unpublished data) that difference in site has no effect on most parameters tested.

2.3. Processing

A composite sample of Mophane worms was mixed and divided in two equal portions, one was degutted and the other was left with gut contents (not degutted). Three subsamples of the degutted and not degutted were subjected to; hot ash roasting for five to seven minutes, cooking for 1 h (held in boiling water for 45 min) and not cooked or roasted (control), to achieve a completely randomized design with two factors. The samples were analysed in duplicate. Degutting was achieved by forcibly squeezing the gut contents from the body by pushing the head towards the anal region (Siame et al., 1996). The worms were then cleaned with tap water and were sun dried for two days before being placed in clean papers bags.

2.4. Chemical analysis

Samples were oven dried at 105 °C for 24 h and was ground to pass through a 2 mm screen. The neutral detergent fiber (NDF) and acid detergent fiber (ADF) were analysed with the ANKOM fiber analyser using reagents described by Van Soest et al. (1991). Acid detergent lignin (ADL) was determined by digesting the ADF residue in 72% sulphuric acid and then burning the insoluble material in a muffle furnance. Sodium sulphite and amylase were used during the NDF determination. CP was determined using the Kjeldahl method (AOAC, 976.06, 1996). Acid detergent insoluble nitrogen (ADIN) was determined by estimating nitrogen using the Kjeldahl methods on the ADF residue. Available CP (CP_{av}) was determined as the difference between total CP and ADIN (Getachew et al., 2004). Calcium (Ca) was determined by atomic absorption spectrophotometry (AOAC, 968.08, 1996), while phosphorous (P) was determined with a UV-Vis spectrophotometer (AOAC, 965.17, 1996). IVTDMD was determined by a Daisy^{II} Incubator (ANKOM, Technology Corp) using multi-layered polyethylene cloth bags, (F57 filter bags; ANKOM, Technology Corp). The rumen fluid samples for IVTDMD were collected from three ruminally cannulated steers and combined. The steers were fed Bana grass ad lib. At the

end of the incubation, the bags were rinsed four times with distilled water, dried, weighed and placed in an ANKOM fiber analyser and boiled in neutral detergent solution for 60 min. IVTDMD was calculated as the difference between DM incubated and the residue after NDF analyses.

2.5. Statistical analysis

Data was subjected to analysis by the general linear model (GLM) procedures of SAS (1990) to determine the effects of roasting and degutting on chemical composition and IVTDMD. Correlations between CP and ADIN, CP and ADF, CP and IVTDMD, ADF and ADIN, NDF and IVTDMD, ADF and IVTDMD were determined. The difference in chemical composition and IVTDMD between degutting and roasting were tested for significance by least significance differences (LSD) (Sokal and Rohlf (1969)). Results are reported as mean $\pm$ standard error.

3. Results

Cooking methods had an effect on ash, Ca, P, Zn, IVTDMD ($P<0.001$), CP and Mg ($P<0.05$). Fibre components (NDF, ADF and ADL), ADIN, Mn and CP_{av} were not affected ($P>0.05$) (Table 2).

Degutting had an effect on almost all parameters tested, i.e. ash, CP, fiber components, Ca, P, Zn, ($P<0.001$), ADIN, IVTDMD and Mn ($P<0.01$). Mg and CP_{av} were not ($P>0.05$) affected by degutting (Table 1).

Ash was significantly lower ($P<0.001$) in degutted/control than undegutted/control and also lower ($P<0.01$) than degutted/roasted samples. Degutted/control, degutted/roasted and degutted/cooked samples were significantly lower ($P<0.001$) in NDF than those from undegutted/control, undegutted/roasted and undegutted/cooked. ADF was significantly lower ($P<0.01$) in degutted/control than undegutted/control, undegutted/roasted and undegutted/cooked. Degutted/control samples had significantly higher ($P<0.001$) IVTDMD than undegutted/control and undegutted/cooked samples. CP was higher ($P<0.01$) in degutted/control samples than undegutted/control and undegutted/roasted samples (Table 2).

A significant and negative correlation was observed between NDF and IVTDMD ($r=-0.77$, $P<0.01$), between ADF and IVTDMD ($r=-0.86$, $P<0.001$) and between ADF and ADIN ($r=-78$, $P<0.01$). Only ADF seem to be a better predictor of IVTDMD than NDF, according to the relationships $Y=-1.0737X+1017.6$, where Y is IVTDMD in g/kg DM and X is ADF in g/kg DM and $Y=-0.7333X+996.2$, where Y is IVTDMD in g/kg DM and X is NDF in g/kg DM, respectively. The correlation between CP and ADF was highly significant ($r=-0.92$, $P<0.001$). CP was also significantly correlated to ADIN ($r=0.92$, $P<0.001$). CP and IVTDMD were positively correlated ($r=0.88$, $P<0.001$), and CP levels explained 78% of the variation in IVTDMD, according to the relationship $Y=0.4552X+658.3$, where Y is IVTDMD in g/kg DM and X is CP in g/kg DM.

Table 1
The effect of degutting on chemical composition and in vitro true dry matter digestibility of Mophane worms[a]

Parameters	Degutted	Non degutted	SEM[b]	Probability
Ash	50.5[b]	71.6[a]	1.6	<0.001
CP	480.7[a]	355.5[b]	8.9	<0.001
$\%CP_{av}$	59.0	62.0	1.7	NS
NDF	163.8[b]	238.9[a]	1.9	<0.001
ADF	132.6[b]	182.3[a]	5.4	<0.001
ADL	15.5[b]	43.5[a]	2.6	<0.001
ADIN	196.4[a]	136.6[b]	7.2	<0.01
Ca	3.6[b]	4.6[a]	0.1	<0.001
P	1.4[b]	1.5[a]	0.02	<0.01
Zn	11.9[b]	10.6[a]	0.2	<0.001
Mn	1.9[a]	1.3[b]	0.1	<0.01
Mg	1.2	1.0	0.1	NS
IVTDMD	875.2[a]	823.0[b]	4.3	<0.01

[a]Units are expressed as g/kg DM except for Zn and Mn, which are in ppm.
[b]Standard error of the mean. Figures with different superscript across rows are different at $P<0.05$.

Table 2
Chemical composition and in vitro true dry matter digestibility of Mophane worm of degutted and non degutted of cooked, roasted and raw samples[a]

Effects/Parameters		Ash	CP	$\%CP_{av}$	NDF	ADF	ADL	ADIN	Ca	P	Zn	Mn	Mg	IVTDMD
Degutted	Control	45.1	524.3	60.4	165.8	112.7	11.8	207.5	3.3	1.1	13.1	2.0	1.1	904.2
	Roasted	64.5	468.5	61.0	167.2	132.8	22.8	182.9	3.5	1.7	12.0	1.8	1.6	868.5
	Cooked	41.9	449.4	55.7	158.5	152.3	11.9	198.9	3.9	1.2	10.6	1.8	1.1	852.8
Non degutted	Control	70.9	382.9	57.0	237.6	177.1	46.6	164.6	3.5	1.3	11.8	1.3	1.0	822.3
	Roasted	83.1	363.8	62.8	226.6	179.4	47.2	135.1	6.8	1.9	9.7	1.4	1.3	853.8
	Cooked	60.9	319.8	66.2	252.6	190.4	36.7	110.1	3.6	1.3	8.7	1.2	0.7	789.9
SEM[a]		2.7	15.3	3.0	3.3	9.4	4.5	12.5	0.2	0.04	0.3	0.1	0.1	7.5
Main effects	Treatment	***	*	NS	NS	NS	NS	NS	***	***	***	NS	*	**
	Degutting[c]	***	***	NS	***	***	***	**	***	**	***	**	NS	**
Trt x Deg[b]		NS	NS	NS	**	NS	NS	NS	***	NS	NS	NS	NS	*

SEM = Standard error of the mean. NS = $P>0.05$; * = $P<0.05$; ** = $P<0.01$; *** = $P<0.001$.
[a]Units are expressed as g/kg DM except for Zn and Mn, which are in ppm.
[b]Trt × Deg = Treatment × Degutting interaction.
[c]See Table 1.

4. Discussion

It is important to use proteins to the best effect because they are a valuable (Ørskov et al., 1980) and an expensive resource (Abdullah and Awawdeh, 2004). Botswana does not produce oil seed cakes. Although blood and carcass meals are produced locally, their inclusion in livestock diets is prohibited. This gives animal scientists a challenge in their search for alternative protein sources. Results of the present study indicate that Mophane worms have CP at concentrations ranging from 320 to 524 g/kg DM. These figures are more than the CP level reported by van Voorthuizen (1976) but less than those reported by

Sekhwela (1989), Ohiokpehai et al. (1996) and O.R. Madibela, unpublished data. The difference between the present study and those studies is that it is not known how the latter samples were processed before they were analysed. Our samples were either degutted or left intact, and either cooked, roasted or raw when analysed. Cooking lowered the CP content of Mophane worm relative to the uncooked, possibly by some protein dissolving in the cooking water which was the discarded. For those samples that were degutted, there was a high concentration of CP (Table 1). Apparently, the undigested leaves of Mophane vegetation, still in the gut of Mophane worm, may have reduced the CP concentration. However, the CP level of 355.5 g/kg DM for the undegutted samples is still considered high compared to some plant proteins and is comparable to oil cakes. According to McDonald et al. (2002), residues of groundnut, cottonseed, linseed and soyabean has protein levels ranging from 200 to 500 g/kg. This means that if Mophane worm were to be considered as a potential ruminant protein source, it would not be necessary to degut the worm. This would speed up the harvesting of Mophane worm by harvestors since time would not be spent on degutting. However, it would be necessary to determine if this leafy material inside the worm does have anti-nutritional compounds or whether the worm enzymes were able to degrade these compounds. Analysis of Mophane leaves in our laboratory has shown that it has condensed tannin level of 25 g/kg (O.R. Madibela and colleagues, unpublished data).

There was no interaction between cooking methods and degutting for all the parameters except for NDF, Ca and IVTDMD, indicating that different treatments had the same effect on both degutting and non-degutting in all these parameters except NDF, Ca and IVTDMD (Table 2). Hot ash roasting did not heat damage Mophane proteins but probably burnt off the spines from the Mophane skin. This is reflected by the lower level of ADIN in the roasted samples than in the control (non-treated samples). Siame et al. (1996) recorded a protein level of 770 g/kg on the skins which included the spines and head capsules. Siame et al. (1996) also found that these fractions of dried Mophane worm contain 27% chitin. Microfibrils, which are formed from adjacent chitin chains, are embedded in a protein matrix, and thus, proteins are major constituents of insect cuticle (Chapman, 1982). In ADF analysis, ADF residue recovers cutin, hence, the skin, spines and head capsules fractions of Mophane worms in the control samples would contribute to the high ADIN.

The elevated levels of minerals in the roasted samples are most likely due to contamination by the hot ash that was used to roast the samples. The ash was obtained from Mophane wood. Botswana soils are deficient in P resulting in lower levels of this mineral in forages. According to APRU (1980), P levels in grass barely reach 1.0 g/kg. The P level in Mophane worms in all samples was higher than 1.0 g/kg and this means Mophane worm may be used to supply this mineral. Calcium, which is associated with P in bone metabolism, was higher than 3 g/kg in all treatments. A ratio of Ca:P of 2:1 is recommended for optimal metabolism of these minerals (Abdulrazak et al., 2000) and the average ratio of 2.9:1 found in Mophane worm will not cause a major mineral imbalance of these minerals.

Degutting affected the fibre components producing degutted samples with low levels of NDF, ADF and ADL. It is suggested that Mophane leaves inside the Mophane worm gut of the undegutted samples elevated these parameters. In vitro digestibility was more than 800 g/kg in all samples, which is consistent with the high available protein. In the present study, protein accounted for three quarters of the variance in in vitro digestibility. CP in

natural grass averaged 70 g/kg during the wet season and dropped to 40 g/kg during the dry season (APRU, 1975). Moleele (1998) observed that browsing of green material by cattle decreased in July, during the dry season, while foraging of litter on the ground increased dramatically and this resulted in a decrease in the supply of nutrients required by the animals. During such times, dried Mophane worms would be useful as a supplementary source of protein for the animals.

Although the methods of processing Mophane worms reported in the present study are traditional and may be considered crude (since temperature and time were not controlled), these results show that in a controlled environment, Mophane worm quality would vary only slightly depending on the processing technology. If Mophane worm is destined for animal feeding it would not be necessary to degut it, but since undegutted Mophane worm has high fibre levels it would need to be degutted for human consumption, as has been the tradition.

Where the environment and weather permits, Mophane worms represent a potential source of protein and energy in diets of livestock. However, the absence of regulations and monitoring of the harvesting process has led to a possibility for abusive pupal harvest (Arntzen and Fidzani, 1998), thus putting doubts on the sustainability of this natural resource. This is compounded by the fact that availability of Mophane worms depends on rainfall and in turn the productivity of the Mophane vegetation. But, in Botswana, rainfall is erratic and its distribution in space and time is unpredictable. The availability of Mophane worms also depends on a tight synchrony of the availability of vegetative material on Mophane trees and the hatching of the moth eggs. Thus, it would be worth exploring the possibility of breeding Mophane worms in captivity and releasing the butterfly to re-seed the Mophane vegetation when the conditions are favourable (Stack, 2003).

Further studies on rumen degradability and on animal performance are needed to determine supplementary feeding levels that would cater for physiological needs of different animals.

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References

Abdullah, A.Y., Awawdeh, F.T., 2004. The effect of protein source and formaldehyde treatment on growth and carcass composition of Awass lambs. Asian-Australasian Journal Animal Science 17, 1080–1087.

Abdulrazak, S.A., Fujihara, T., Onkiek, J.K., Ørskov, E.R., 2000. Nutrient evaluation of some Acacia leaves from Kenya. Animal Feed Science and Technology 85, 89–98.

Allotey, J., Mpuchane S. 2003. Utilization of useful insects as food source. African Journal of Food, Agriculture, Nutrition and Development 3(3) 〈http://www.ajfand.net/Issue-V-files/IssueV-Short%20Communication%20-%20Allotey.htm〉 (downloaded on 28/9/2004).

AOAC, 1996. Official Methods of Analysis. Association of Official Analytical Chemists, Washington, DC.

APRU, 1975. Animal Production Research Unit (APRU 1975). A Five-Year Programme of Intergrated Beef Cattle and Range Research in Botswana, 1970–1975. Part 1 Technical Results. Government Printers, Gaborone, Botswana.

APRU, 1980. Animal Production Research Unit (APRU 1980). Beef Production and Range Management in Botswana. Government Printers, Gaborone, Botswana.

Arntzen, J., Fidzani, H., 1998. Incentives for sustainable natural resource management and economic diversification in Botswana. A Final Report of the Environmental Economics, Sub-component in the BNCS Action Plan 〈http://www.envirobotswana.gov.bw/pdffiles/vol2.pdf〉 (accessed 17/12/2005).

Bauman, D.E., Baumgard, L.H., Corl, B.A., Giinari, J.M., 2000. Biosynthesis of conjugated linoleic acid in ruminants. Proceedings of American Society of Animal Science 1999. 〈http://www.asas.org/jas/symposia/proceedings/0937.pdf〉 (accessed 24/06/2004).

Bekker, R.P., De Wit, P.V., 1991. Vegetation map of republic of Botswana. Soil Mapping and Advisory Services Project. AG:DP/BOT/85/011. Food and Agricultural Organisation and Government of Botswana.

Burgess, J., 2003. Country pasture/forage profile; Botswana. 〈www.fao.org/ag/FAOINFO/AGRICULT/AGP/AGPC/doc/counprof/Botswana/botswana.htm#3〉 (accessed; 05/01/2006).

Chapman, R.F., 1982. The Insects: Structure and Function, third ed. Harvard University Press, Cambridge.

De Wit, P.V., Nachtergaele, F.O., 1990. Explanatory Note on the Soil Map of Republic of Botswana. Soil Mapping and Advisory Services Botswana. AG: BOT/85/001 Field Document 30. Food and Agricultural Organisation and Government of Botswana.

Ditlhogo, M., 1996. The ecology of *Imbrasia belina* (westwood0 in north-Eastern Botswana In: Gashe B.A. Mpuchane, S.F., (Eds.), Phane, Proceedings of the Multidisciplinary Symposium on Phane, Held in Gaborone, Botswana 18 June 1996. Department of Biological Sciences and Kalahari Conservation Society.

Getachew, G., Robinson, P.H., DePeters, E.J., Taylor, S.J., 2004. Relationships between chemical composition, dry matter degradation and in vitro gas production of several ruminant. Animal Feed Science and Technology 111, 57–71.

Kott, R.W., Hatfield, P.G., Bergman, J.W., Flynn, C.R., Van Wagoner, H., Boles, J.A., 2003. Feedlot performance, carcass composition, and muscle and fat CLA concentrations of lambs fed diets supplemented with safflower seeds. Small Ruminant Research 49, 11–17.

Macala, J., Sebolai, B., Majinda, R.R., 1992. *Colophospermum mopane* browse plant and sorghum stover as feed resources for ruminants during the dry season in Botswana. In: Stares, J.E.S., Said, A.N., Kategile, J.A., (Eds.), The Complementary of Feed Resources for Animal Production in Africa. Proceedings of the Joint Feed Resources Networks Workshop Held in Gaborone, Botswana, 4–8 March 1991.

Marais, E., 1996. Omaungu in Nambia: *Imbrasia belina* (Saturniidae: Lepidoptera) as a commercial resource. In; Gashe, B.A., Mpuchane, S.F., (Eds.), Phane, Proceedings of the Multidisciplinary Symposium on Phane, Held in Gaborone 18 June 1996. Department of Biological Sciences and Kalahari Conservation Society.

McDonald, P., Edwards, R.A., Greenhalgh, J.F.D., Morgan, C.A., 2002. Animal Nutrition, sixth ed. Pearson Prentice-Hall, Essex.

McGuire, M.A., McGuire, M.K., 2000. Conjugated linoleic acid (CLA): a ruminant fatty acid with beneficial effects on human health. Proceedings of American Society of Animal Science 1999 〈http://www.asas.org/jas/symposia/proceedings/0938.pdf〉 (accessed 24/06/2004).

Moleele, N.M., 1998. Encroacher woody plant browse as feed for cattle. Cattle diet composition for three seasons at olifants drift, South-East Botswana. Journal of Arid Environments 40, 255–268.

Mosimanyana, B.M., Kiflewahid, B., 1987. The value of browse as ruminant feed: The case of *Colophaspermum mopane*. The Bulletin of Agricultural Research in Botswana 6, 38–43.

Moruakgomo M.B.W., 1996. Commercial utilization of Botswana's veld products—The economics of phane—the dimensions of phane trade. In: Gashe B.A., Mpuchane, S.F., (Eds.), Phane, Proceedings of the Multidisciplinary Symposium on Phane, Held in Gaborone, Botswana, 18 June 1996. Department of Biological Sciences and Kalahari Conservation Society.

Mpuchane, S.F., Gashe, B.A., Allotey, J., Ditlhogo, M.K., Siame, B.A., Tefera, G., Collison, E.K., Simpanya, M.F., 2001. Phane, its exploitation and conservation in Botswana. Technical Bulletin 6. Phane Research Project, Department of Biological Sciences, University of Botswana.

Ohiokpehai, O., Bulawayo, B.T., Mpotokwane, S., Sekwati, B., Bertinuson, A., 1996. Expanding the use of phane, a nutritionally rich local food. In: Gashe, B.A., Mpuchane, S.F. (Eds.), Phane, Proceedings of the Multidisciplinary Symposium on Phane, Held in Gaborone, 18 June 1996. Department of Biological Sciences and Kalahari Conservation Society.

Ørskov, E.R., Deb Hovell, F.D., Mould, F., 1980. The use of nylon bag technique for the evaluation of feedstuffs. Tropical Animal Production 5, 195–213.

SAS Institute Inc., 1990. SAS User's Guide. Statistics Version 6. SAS Institute Inc., Cary, NC, USA.

Sekhwela, M.B.M., 1989. The nutritive value of Mophane bread–Mophane insect secretion (Maphote or Moaboti). Botswana Notes and Records 20, 151–153.

Siame, A.B., Mpuchane, S.F., Gashe, B.A., Allotey, J., Teferra, G., 1996. Nutritional quality of Mophane worms, *Imbrasia belina* (Westwood), and the microorganisms associated with the worms. In: Gashe, B.A., Mpuchane, S.F. (Eds.), Phane, Proceedings of the Multidisciplinary Symposium on Phane, Held in Gaborone 18 June 1996. Department of Biological Sciences and Kalahari Conservation Society.

Sokal, R.R., Rohlf, F.J., 1969. Biometry. The Principles and Practice of Statistics in Biological Research, second ed. WH Freeman, San Francisco.

Stack, J., 2003. Research Highlights from Mophane Worm and Mophane Woodland Project 〈www.mopane.org/Research%20highlights%202003.pdf〉 (accessed 17/12/2005).

Styles, C.V., 1996. The biological ecology of *Imbrasia belina* (saturniinae, saturniidae) with reference to its behaviour, physiology, distribution, population dynamics, impact within mopane veld and utilisation within South Africa. In: Gashe, B.A., Mpuchane, S.F. (Eds.), Phane, Proceedings of the Multidisciplinary Symposium on Phane, Held in Gaborone, 18 June 1996. Department of Biological Sciences and Kalahari Conservation Society.

van Soest, P.J., Robertson, J.B., Lewis, B.A., 1991. Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. Journal of Dairy Science 74, 3583–3597.

van Voorthuizen, E.G., 1976. The Mophane tree. Botswana Notes and Records 8, 223–261.

Zinzombe, I.M., George, S., 1994. Larval lipid quality of Lepidoptera *Gonimbrasia belina*. Botswana Notes and Records 26, 167–173.