EVALUATION OF THE QUALITY CHARACTERISTICS OF THE MARAMA BEAN (*Tylosema esculentum)*, AN UNDERUTILIZED GRAIN AND TUBER PRODUCING LEGUME IN SOUTHERN AFRICA

by

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DECLARATION

I declare that the thesis, submitted to the University of Namibia in fulfillment of the requirements of the degree of Master of Science, is my original work and has not been submitted elsewhere for a degree.

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ABSTRACT

The potential of the marama bean plant, *Tylosema esculentum* (Burch.) Schreiber, a drought-tolerant, bean–bearing legume, native to Southern Africa, was investigated to potentially form part of a food based approach in rural agricultural extension programmes. Nutrient content and sensory attributes of roasted marama beans were determined as well as the potential of the marama plant as fodder for cattle.

Chemical analyses were performed on roasted Namibia and Botswana marama beans to determine the nutritional content thereof. The beans contain high levels of protein, unsaturated fats, phosphorus, calcium, vitamin A, vitamin B_3, vitamin B_6, folic acid, vitamin B_{12}, vitamin E, iron, zinc and iodine. Quantitative descriptive sensory analyses were performed on roasted marama beans from Botswana and Namibia to determine and compare the sensory attributes thereof. The Botswana traditionally roasted, Botswana oven roasted and Namibian oven roasted samples grouped together. The Namibian traditionally roasted marama bean had a significantly more intense burnt, bitter and chemical aroma and flavour and taste.

To assess the value of the marama plant as a fodder for cattle, an experiment was conducted to measure the effect of season, stocking rate and frame size on the diet selection of the marama plant by cattle grazing the veld of the Sandveld Research Farm in the eastern part of Namibia. This study proved that only season had a significant influence on the selection of the marama plant as a feedstuff for cattle. The marama plant is indeed utilized by free-ranging beef cattle, but not preferentially so.

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Introduction and Statement of the Problem

1.1 BACKGROUND

"Hunger and malnutrition are unacceptable in a world that has both the knowledge and the resources to end this human catastrophe". These were part of the opening sentences of the World Declaration on Nutrition produced by the FAO and the World Health Organization (WHO) International Conference on Nutrition (ICN) held in Rome in December 1992.

Around the world it shows that the causes underlying most nutritional problems have not changed very much over the past 50 years. However, the approaches in tackling malnutrition have changed, becoming multi-pronged e.g. combining supplementation programmes with nutritional food fortification and following food-based approaches in extension programmes. Protein-energy malnutrition, vitamin A deficiency, iodine deficiency disorders and nutritional anemias, resulting from iron deficiency or losses, are still the most common serious nutritional problems in almost all countries (Latham, 1997).

1.1.1 Global under-nutrition

Worldwide there are improvements in life expectancy, adult literacy and nutritional status, but the greatest concern is still the fact that approximately 780 million people in developing countries, almost 20% of their combined population, do not have access to enough food to meet their basic daily nutritional needs. More than 2 000 million people, mostly women and children, are deficient in one or more micronutrients. Babies are still born mentally retarded as a result of iodine deficiency. Children die and go blind because of Vitamin A deficiency. Enormous numbers of women and children are severely affected by iron deficiency (Table 1.1).

Table 1.1

Global population at risk of and effected by micronutrient malnutrition (millions)[1]

Region[2]	**Iodine deficiency disorders**		**Vitamin A deficiency**		**Iron deficiency or anemia**
	At risk	**Affected**	**At risk[3]**	**Affected**	
Africa	181	86	31	1.0	206
Americas	168	63	14	0.1	94
Southeast Asia	489	176	123	1.7	616
Europe	141	97	-	-	27
Eastern Mediterranean	173	93	18	0.2	149
Western Pacific[4]	423	141	42	0.1	1058
Total	**1 572**	**655**	**288**	**3.1**	**2 150**

[1] *Latham, 1997*
[2] *WHO regions*
[3] *Preschool children only*
[4] *Including China*

Distressing is the high prevalence and increasing number of under-nourished children under five years of age in Africa, Asia, Latin America and the Caribbean (Table 1.2).

Table 1.2

Global prevalence of underweight[1] children under five years of age[2]

Region	Percentage underweight			Number underweight		
	1980	**1985**	**1990**	**1980**	**1985**	**1990**
Sub-Saharan Africa	28.9	29.9	29.9	19.9	24.1	28.2
Near East / North Africa	172	15.1	13.4	5.0	5.0	4.8
South Asia	63.7	61.1	58.5	89.9	100.1	101.2
Southeast Asia	39.1	34.7	31.3	22.8	21.7	19.9
China	23.8	21.3	21.8	20.5	21.1	23.6
Central America / Caribbean	17.7	15.2	15.4	3.1	2.8	3.0
South America	9.3	8.2	7.7	3.1	2.9	2.8
Global (average Percentage/total number)	**37.8**	**36.1**	**34.3**	**164**	**178**	**184**

[1] *Underweight defined as weight-for-age less than -2 standard deviations from the mean*
[2] *Latham, 1997*

1.1.2 Under-nutrition in Africa

Worldwide, the worst of the underweight situation is in Sub-Saharan Africa. Eastern Sub-Saharan Africa is the sub-region experiencing the largest increases in prevalence and numbers of underweight children (Table 1.3). Northern Africa is the only sub-region where the number of underweight children is decreasing. These negative trends in Africa reflect the deteriorating situation in many Sub-Saharan African countries: the

poverty rate has increased, HIV/AIDS had a huge impact and agricultural productivity has not increased. The focus on child under-nutrition has shifted from Asia to Africa. Although the number of underweight pre-schoolers will remain higher in Asia, the rising prevalence in Africa is alarming (UN SCN, 2004).

Table 1.3

Prevalence of underweight children in under five years of age in Africa[1]

Region	Prevalence (%)				Numbers (million)			
	1990	1995	2000	2005	1990	1995	2000	2005
Eastern Africa	26.7	27.9	29.2	30.6	9.5	10.9	12.8	14.8
Middle Africa	27.8	26.9	26.1	25.3	3.7	4.2	4.7	5.3
Northern Africa	12.3	10.9	9.7	8.6	2.6	2.3	2.1	1.9
Southern Africa	14.0	13.9	13.7	13.6	0.8	0.8	0.8	0.8
Western Africa	27.8	27.5	27.1	26.8	8.8	9.6	10.5	11.7
Africa (Total)	**23.6**	**23.9**	**24.2**	**24.5**	**25.3**	**27.8**	**30.9**	**34.5**

[1] *UN Standing Committee on Nutrition, 2004*

1.1.3. Malnutrition in Southern Africa

In the Southern African region approximately 13.7% of children under the age of five years are stunted (low height for age) and 6.6% under the age of five years are wasted (low weight for height) (UN SCN, 2004). This is an unacceptable situation; in a recent survey in Sekhukhuneland in South Africa the complexity of malnutrition was highlighted with 57% of the women found to be overweight (Schönfeldt *et al.*, 2005) and 0.8 million children in Southern Africa being under-nourished (UN SCN, 2004). In South Africa alone, one in five children suffers from chronic malnutrition. A 1999 Health

Department survey showed that 21.6% of all South African children between the ages of one and nine are stunted because of malnutrition (healtoronto.com, 2005).

1.1.4 Under-nutrition in Namibia

Namibia is a semi-arid developing country with limited rainfall. Almost 80% of all food and feedstuffs are imported, because cultivation of crops is limited, mainly due to variable rainfall and lack of extended agronomic knowledge. The demand for suitable protein and energy sources is not only on the increase for humans, but also for livestock (Mc Donald, 1988). In Namibia the crude protein content of principle grasses varies between 5 and 8% and supplementation is often needed. Protein sources used in supplementary feed are molasses, wheat bran and hominy chop. Cattle in Namibia should receive on average 150 g per day of a protein/energy supplement during the summer season and 300 g per day of protein/energy supplement during the winter season (Pers.comm. Feedmaster, 2005).

The proportion of households living in relative poverty in Namibia was calculated at 38% in 1994 (Namibia 2004 Millennium Developments Goals, 2004). During a Demographic and Health Survey conducted by the Ministry of Health and Social Services in Namibia in 2000, it was determined that 24% of all children under five years of age, were found to be underweight. Five percent of the children under the age of five years is considered to be severely underweight. One quarter of Namibian children under the age of five years is short for their age, or stunted, while eight percent are severely stunted (MOHSS, 2003). Greater proportions of children in rural areas (27%)

were underweight than those (16%) in urban areas (Fig 1.1). The highest proportion of underweight children was found in Ohangwena (35%), Kavango and Omusati (both 28%) and Oshikoto (27%) (El Obeid *et al*., 2001).

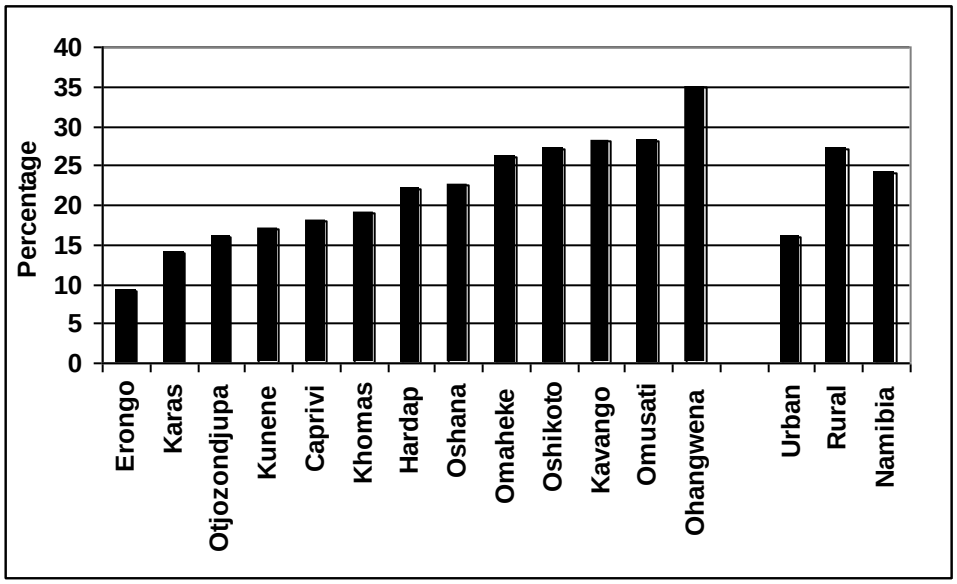

Fig 1.1: Percentages of children under 5 years of age in Namibia severely underweight during 2000 (El Obeid *et al*., 2001)

During the Demographic and Health Survey in Namibia in 2000, it was found that 38% of children under five years of age received a vitamin A supplement in the previous six months. Adequate stores of vitamin A in the body can have an enormous effect on the ability to fight diseases. The percentage of children under five years of age living in households with adequate iodized salt is 57%. This is cause for concern, since iodine is important in the development of cognitive ability. Efforts to promote iodized salt should be focused on the Omaheke region (eastern part of Namibia) especially (MOHSS, 2003).

1.2 MOTIVATION FOR THE STUDY

Throughout history, mankind has used some 3000 plant species for food, but today the world's food basket consists of a mere 20 selected species. The lesser-known food crops which remain outside the scope of science have not been rejected because of any inferiority. They have simply been overlooked because they are native to the tropics, a region generally neglected because the world's research resources are concentrated in the temperate zones (Vietmeyer, 1986). A vast majority of the world's edible plants can however supplement these selected and proliferated species, thereby improving dietary diversity and nutrient intake. Its role in combating malnutrition, both over- and under-nutrition, is mostly unexploited.

Under-nutrition is a direct result of insufficient food intake. Action against children suffering from malnutrition is urgent. Children make up nearly 40% of the world's population (IAACP, 2001). Almost 30 000 children die every day in developing countries and 183 million are under-nourished. The international community has set ambitious goals to at least, by the year 2015, decrease by 50% the proportion of the world's population living in extreme poverty, suffering from under-nutrition and being deprived from having the right to grow up healthy (IAACP, 2001).

This situation presents a challenge to scientists to develop indigenous crops with high nutritional value to be utilized in sustainable agriculture, with the aim to alleviate under-nutrition and poverty. Scientists should design programmes specifically aimed at increasing the underexploited natural resources. This has the possibility of improving

household food security by improving dietary diversity and nutrient intake, resulting in improved nutritional status of potential consumers at village level and exploring the species' role in improved farming systems.

1.2.1 EU Project ICA4-CT-2000-30010

A project was initiated in 2000 by the *European Union International Cooperation with Developing Countries*, with the aim to develop scientific understanding of the marama bean (*Tylosema esculentum* (Burch.) A. Schreib.), an under-utilized grain and tuber producing legume for Southern Africa. The objectives of the project were to:

- Understand the distribution and biology of the marama bean plant, a wild perennial legume of arid areas of Southern Africa, which produces grain (seed) of high protein and oil content, as well as large tubers of high protein and carbohydrate content.

- Assess the suitability of the marama bean plant, which is an under-utilized source of food and animal fodder, as a sustainable, low input, nutritious crop for subsistence agriculture in Southern Africa.

- Develop, based on the knowledge of the plant's biology and field trials, an understanding of the agronomy of the marama bean plant in relation to production and quality requirements of local consumers and producers.

- Analyze the physiology and biochemistry of the marama bean, to understand nitrogen metabolism, carbon assimilation and water relations with respect to environmental conditions.

- Use the information on plant processes to understand the responses and mechanisms of the growth of the marama bean plant in Southern African conditions and to use the information and insights to develop the marama bean plant as a crop for dry areas.

- Disseminate the results from the study of the marama bean plant and use the information to encourage policy makers and advisors to promote the use of the marama bean as a crop.

This particular study is part of the EU Project ICA4-CT-2000-30010 and aims to assess the suitability of the marama bean, an under-utilized source of food and animal fodder, as a sustainable, low input, nutritious legume crop for subsistence agriculture in Southern Africa (Namibia, Botswana and South Africa).

Legumes form an important part of the diet of many individuals. They rank second only to cereals in supplying energy and protein. Legumes supply about the same number of kilojoules as cereals, but they contain two to four times more protein. Also, the amino acid patterns of legumes complement those of cereal grains so that the combination of the two provides dietary protein that is much more efficient (diabetesforum.net, 2005).

For more than 300 million people, an inexpensive bowl of common beans (*Phaseolus vulgaris*) is the centerpiece of the daily diet. Global bean harvest reaches 18 million tons annually and has an estimated value of US$ 11 billion. Nutritionists characterize beans as a nearly perfect food because of its high protein content (contains essential and non-essential amino acids) and high amounts of fiber, carbohydrates and other dietary necessities. A single serving (half a cup) of common beans provides at least half the recommended daily dietary allowance of folic acid; a vitamin B that is especially important for pregnant women. It also supplies 25 to 30% of the recommended levels of iron, 25% of the daily requirement of magnesium and copper, as well as 15% of the potassium and zinc (ciat.cgiar.com, 2001).

The common bean was domesticated more than 7 000 years ago in Mexico and Central America. Scientists believe that dry beans, along with other crops like maize and squash, probably began as weeds in fields planted with cassava and sweet potatoes in Central America (ciat.cgiar.com, 2001). Over the years, farmers have grown a mixture of bean types as a hedge against drought, disease and pest attacks. Latin America is the most important bean producing region, its eight million hectares accounting for almost half of the global output. Because there is no cheaper source of protein, per capita consumption of beans is high in very poor countries, such as Nicaragua (22.5 kg per capita per year) and in poorer regions of higher income countries, such as Northeast Brazil (18.5 kg per capita per year). Dry beans were introduced in sub-Saharan Africa several centuries ago by Portuguese traders. Today this crop is a vital staple on this continent, providing protein for more than 70 million people (ciat.cgiar.com, 2001).

1.2.2 The marama bean – an underutilized crop

The marama bean plant, *Tylosema esculentum* (Burch.) Schreiber is a drought-tolerant, bean–bearing legume, native to the southern regions of Africa. The specific name *esculentum* (= edible) is well chosen, according to Codd (1952), because ”…..not only is the plant sought out by browsing stock and game, but the tubers and seeds were a staple food for the indigenous people and are even today relished by the farming communities in areas where plants grow…….”. The plant grows in open veld, is a perennial legume scrub, with trailing stems which can reach several meters and which arise from a large underground tuber.

The marama bean plant is native to dry areas with little and very variable rainfall. In these dry areas, legumes are of particular importance. In developing countries the cultivation of legumes is the best and quickest way to augment the production of food proteins (National Academy of Sciences, 1979). Many of these underutilized crops, such as the marama bean, occupy huge areas of land and are collected by the poorest communities. The nutritional status of these poor people plays a major role in determining their overall health.

Despite its high nutritional value and the fact that it is highly sought after by humans and animals, the plant remains under-utilized (*An underutilized species is, by definition, one that has not been fully exploited in terms of its potential use as a food and/or non-food product (Azam-Ali, 1996)*). This fact has been stated in the National Academy of Sciences report; ” ……Of all plants described in this book *(Tropical Legumes :*

Resources for the Future), the marama bean is perhaps the least developed in terms of scientific study or plant breeding efforts to improve it " (ECHO, 1999).

Humans, questioned on why they consume marama beans, reported that they have done so over many years and with no apparent side effects (Starcher, 1985). Preliminary studies have indicated that the marama bean is adequate in nutrients for the human diet, but it is recommended that the trypsin inhibitor activity should be inhibited prior to consumption (Bower et al, 1988). Trypsin is an essential enzyme that degrades protein by breaking the peptide bonds between amino acids.

On average the marama seeds contain 30 – 39% protein, which is roughly equal to that of soybeans at 38% protein (Biesele & Murray, 1983). Protein quality of marama beans, i.e. amino acid composition, is slightly better than that for soybeans and is comparable to that of casein or milk protein (Ripperger – Suhler and Longenecker, 1982, cited in Biesele and Murray, 1983). Protein from the marama bean is high in the amino acid lysine (Rachie *et al.*, 1979), but low in the amino acid methionine (Bower *et al*, 1988) like most legumes. Methionine is an essential sulfur containing amino acid and is limiting in a legume based diet.

The oil content of the dry seeds ranges from 36 – 43% and it approaches that of peanuts (Wehmeyer, 1969). The oils of marama beans are reported to contain 31% unsaturated fatty acids, in particular oleic acid and linoleic acid (Ripperger–Suhler and Longenecker 1982, cited in Biesele and Murray, 1983).

1.3 STATEMENT OF THE PROBLEM

This study aims to investigate the potential of the marama bean plant (*Tylosema esculentum),* a crop indigenous to the Southern African region, as a food based approach in rural agricultural extension programmes to alleviate under-nutrition amongst the local communities.

This leads to the formulation of the objectives for the study:-

- To determine the nutrient content of the marama bean
- To determine the sensory acceptability and characteristics of the marama bean
- To determine the acceptability of the marama plant as fodder for livestock

1.4 PURPOSE OF THE STUDY

The outcome of this study can play an important role in improving the nutritional status of children suffering from under-nutrition in areas where the marama plant grows in abundance. In Namibia these areas include the Omaheke and Otjozondjupa regions. In Botswana it includes the Maun, Ghanzi and D'kar triangle.

Educational material can be developed for target groups to be incorporated in training and development programmes to increase consumption of the marama bean.

Programmes can also be developed for target groups to improve cultivation and production of the marama bean.

According to the National Academy of Sciences (1979), cattle in Africa “eagerly eat” the leaves and stems of the Marama bean plants, although Watt and Breyer-Brandwijk (1962) reported that; “…….the foliage of this species is apparently not browsed by stock”. The only reported use as an animal supplement is by some native farmers who used them to fatten pigs (Starcher, 1985).

It is anticipated that if livestock prefers the marama plant in abundance, cows may conceive easier (increasing the protein content of their diets), which will result in an improvement in the production of livestock and thus will have an economical benefit in combating poverty.

It is therefore a necessity that the nutritional potential of all plants is exploited, especially underutilized crops with high levels of essential nutrients.

1.5 LAYOUT OF THE THESIS

1.5.1 Literature review as described in Chapter 2

Relevant literature on the quality characteristics of the marama bean is included in this chapter.

1.5.2 Description of the marama plant / tuber / beans as described in Chapter 3

A detailed description and pictures of the marama plant, tuber and beans are given with the relevant literature.

1.5.3 Nutritional content as described in Chapter 4

Chemical analyses were performed on the Namibia and Botswana roasted marama beans and include proximate analyses, fatty acids, vitamins, minerals, dietary fiber, amino acids, iodine, cholesterol and non-structural carbohydrates. Samples were analyzed in duplicate at the Agricultural Research Centre, Irene, Pretoria, the Institute for Soil, Climate and Water, Pretoria and the SGS laboratories, Midrand, South Africa.

1.5.4 Sensory qualities as described in Chapter 5

Quantitative descriptive sensory analyses were performed on marama beans (traditionally roasted and oven roasted) from Namibia and Botswana, to determine and

compare the sensory attributes thereof. An important part of sensory analyses was to show, not only the attributes that consumers like or dislike, but also the most important characteristics which determine the overall acceptability. Ten experienced panelists were asked to compile sensory profiles of the different samples. The sensory attributes to be evaluated, included aroma, flavour, after-taste and mouth-feel characteristics. Evaluations were done at the Agricultural Research Centre, Irene, Pretoria, South Africa. Data was statistically analyzed using the GenStat 2000 computer program.

1.5.5 Animal studies as described in Chapter 6

An experiment was conducted to measure the effect of season, stocking rate and frame size on the diet selection of the marama plant by cattle grazing the veld of the Sandveld Research Farm in the eastern part of Namibia. Eight groups consisting of at least 24 cows were used in the trial. The actual bites taken from the cows from the veld were recorded in 40 minute intervals, were repeated on early mornings and late afternoons per factorial treatment. From the total bites, the percentages bites taken of the marama plant were calculated and statistically analyzed by the general linear model of the SPSS computer programme.

Chemical and biochemical analyses, including proximate analyses, digestibility of organic matter and determination of metabolizable energy content, were conducted at the Agricultural Laboratory, Ministry of Agriculture, Water and Rural Development, Windhoek, Namibia.

1.5.6 Conclusions and Recommendations as described in Chapter 7

This chapter briefly explains the outcome of the study and the recommendations for future research.

1.5.7 References as described in Chapter 8

All references pertaining to the thesis are listed in this Chapter.

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2

Literature review

2.1 GENERAL

The marama bean plant, *Tylosema esculentum* (Burch.) Schreiber, belongs to the Fabaceae (Leguminosae), subfamily Caesalpinioideae (Dubois *et al.*,1995). According to Coetzer and Ross (1977), cited in Dubois et al., there are four species in the genus *Tylosema* namely; *T. esculentum* (Burch) Screiber, *T. fassoglensis* (Kotschy) Torre and Hillc, *T. argentea* (Chiov) Brenan and *T humifusa* (Pichi – Serm and Roti – Michael) Brena, all native to Africa.

It is reported that the marama plant's desirability led to foragers in the Kalahari walking as far as 40 kilometers to collect the marama beans and that some groups live on little else for many months of a year (Tanaka, 1976). Enjoyment of the marama plant is not confined to a small minority of foragers. The first farmers coming from the northern Cape Province of South Africa to the Ghanzi area in Botswana relied heavily for their subsistence on marama beans supplied by their San trading partners (Russell and Russell, 1979).

The Kalahari areas have an abundance of seasonal, edible plants, but perennials that are detectable throughout the year, like the marama plant, are scarce (Keith, 1975)

The marama bean was described in a National Academy of Science (1979) report as a legume crop of considerable potential in arid land agriculture. Several common names have been applied to *Tylosema esculentum.* The most widely used names in African literature seem to be marama or gemsbok bean, while in some American literature morama has been used (Coomber and Coomes, 1950; Watt and Breyer–Brandwijk, 1962) for the same plant. The marama bean is also known as mangetti, braaiboontjie, ombanui / otjipiva / ozombanui (Herero), gami (!Kha-Khu), tsi / tsin (Kung Bushmen), marumama (Thonga), lai / muraki / litammani / rama / tammani (Tswana) and maramma – a perennial legume native to the arid and semi-arid grasslands of Southern Africa (Bousquet,1981–1982; Keegan and Van Staden, 1981)

Bousquet (1983) summarized the work done prior to 1983 and mentioned that the plant has been studied for possible cultivation in the United States at research centers and elsewhere (Bousquet, 1981–1982; Miller, 1981). In the fall of 1980, propagation experiments were conducted at three institutions in Texas to determine the environmental tolerance and potential productivity of the marama bean. The Chihauhuan Desert Research Institute established field test sites. At Texas Tech university germination experiments and greenhouse planting was begun. At Greenhills Agricultural Experiment Station near Dallas, outdoor propagation began in prairie soils. Researchers found that scarification with industrial-strength H_2SO_4 resulted in 82% germination rates. It was found that without supplemental watering, vegetative growth reached a maximum of 20 cm (Miller, 1981). The best results were from the water-enriched plots. Miller reported that seeds planted in a test plot of rich topsoil and given four liters of water daily through a drip irrigation system showed the most growth.

Vegetative growth continued through summer resulting in multiple vines approximately 180 cm in length. The tuber that was uprooted for nutritional analyses weighed just over 2 kg.

Reportedly the native seed sources are declining in Africa, as a combined result of political factors, limited access for researchers and encroachment of non-native ungulates (Biesele and Murray, 1983; National Academy of Sciences, 1979). Available literature states that studies of the marama bean are extremely limited (Vietmeyer 1986).

The seeds have potential in the roasted nut market and the oil, with unsaturated fatty acids, can be used as cooking oil (Biesele and Murray, 1983) or in cosmetics (Coomber and Coomes, 1950).

2.2 NUTRITIONAL CONTENT OF THE MARAMA BEAN

Armateifio and Moholo (1998) concluded their study on the chemical composition of four different legumes consumed in Botswana (Table 2.1) by stating that the cultivation and utilization of the marama bean should be encouraged. The marama bean compared well to other beans likes bambara groundnut, mung and tepary bean.

Production of bambara groundnut is concentrated in the north and east parts of Botswana. Bambara groundnut appears in the market more frequently. The immature seeds are boiled, salted and either consumed on their own or mixed with sweet corn.

Mung and tepary beans are grown in the south and central regions of Botswana. These beans can also be boiled, salted and eaten as a main dish.

According to Armateiffo and Moholo's study, the marama bean contained 34.1% protein compared to bambara groundnut (18.3%), mung (26.4%) and tepary bean (34.7). The marama bean contained very high levels of fat (33.5%) compared to bambara groundnut (6.6%). mung (1.1%) and tepary bean (0.9%). Calcium levels of the marama bean was high (152 mg/100g) compared to bambara groundnut (78 mg/100g), mung (142 mg/kg) and tepary bean (88 mg/kg). Marama bean on the other side was very low in carbohydrate content (24.1%) compared to bambara groundnut (63.5%), mung (59.8%) and tepary bean (63.8%).

Table 2.1

Composition of four legumes consumed in Botswana[e]
(100 g raw sample)

Nutrient g/100g	bambara groundnut	marama bean	mung bean	tepary bean
Ash	4.4 ± 0.14 a	3.7 ± 0.14b	4.3 ± 0.14 a	3.8 ± 0.14 b
Carbohydrate	63.5 ± 0.02 b	24.1 ± 0.02 d	59.8 ±0.02 c	63.8 ± 0.02 a
Crude fat	6.6 ± 0.04 b	33.5 ± 0.04 a	1.1 ± 0.04 c	0.9 ± 0.04 d
Crude fiber	5.2 ± 0.13 b	4.4 ± 0.13 b	4.3 ± 0.13 b	4.9 ± 0.13 a
Crude protein	18.3 ± 0.12 d	34.1 ± 0.12 a	26.4 ± 0.12 b	24.7 ± 0.12 c
Minerals (mg/100g)				
Iron	5.9 ± 0.17 b	4.9 ± 0.17 c	6.8 ± 0.17 a	4.5 ± 0.17c
Potassium	1240 ± 12.8 a	776 ± 12.8 d	1032 ± 12.8 c	1093 ± 12.8 b
Phosphorus	296 ± 19.7 b	397 ± 19.7 a	287 ± 19.7 b	304 ± 19.7 b
Sodium	3.7 ± 0.29 b	4.1 ± 0.29 b	11.8 ± 0.29 a	3.8 ± 0.29 b
Calcium	78 ± 4.7 b	152 ± 4.7a	142 ± 4.7 a	88 ± 4.7 b

[e] *Armateifio and Moholo (1998)*
Means with abc are insignificantly different (P > 0.05)

Bower *et al.* (1988) analyzed marama beans originally harvested from the Kalahari Desert, near Ghanzi, in Botswana and the summarized results of their study as well as the results from previous nutritional studies as presented in Table 2.2.

Table 2.2

Selected nutrients of marama seeds[a]

Nutrients (100 g raw grinded sample)	**Wehmeyer *et al.* (1969)**	**National Academy of Science (1979)**	**Ripperger-Suhler and Longenecker (1982)**	**Bower *et al*. (1988)**
Protein (g/100g)	34.3	29.5	31.6[b]	31.8 ± 1.1
Oil (g/100g)	34.8	42.8	36.1	42.2 ± 1.6
Fatty acids (g/100g)	N.D. [b]	N.D. [b]	N.D. [b]	34.0 ± 1.6
Waxes (g/100g)	N.D.[b]	N.D. [b]	N.D. [b]	8.2 ± 1.9
Carbohydrates[c]	23.1	24.3	23.0	18.9 ± 2.2
Ash (g/100g)	3.2	3.2	2.9	3.2 ± 0.1
Moisture (g/100g)	5.6	N.D. [b]	5.2	3.9 ± 1.0
Energy (MJ)	2.68	2.34	2.27	2.66 ± 0.08

[a] *Values based on an as-received weight of 100 g of mature, viable, de-shelled nuts.*
[b] *No data*
[c] *Obtained by difference*

2.2.1 Protein

Bower *et al.* (1988) compared the nutritional content of two other potential crops of limited cultivation – tepary bean and jojoba bean with the marama bean. Tepary bean is also known as the Texas bean. It originated in Mexico and the plant was taken to

Africa for cultivation. Jojoba is a desert scrub, a plant producing beans which contains up to 50% its weight in oil. It is found in Mexico, California and Arizona.

Nutritional values of the compared crops are close, except that the protein content of the marama bean is much higher. The marama bean containing 32% protein, compares well in total protein to other legumes including lupine (31%), lentils (24%), pea (23%) and phaseolus (22%), although lower than some Soybean varieties with 38 – 40% protein (Boutler, 1977).

The marama bean protein quality is generally superior to most common legume crops, like garden bean and pea (Boutler, 1977). When analyzed for some of the essential and non-essential amino acids, it appeared that the amino acids methionine and cystine are the limiting factors (Table 2.3). In light of this low content of the amino acids cystine and methionine, the marama bean albumin content is higher, and globulin is lower than soybean (Boutler, 1977).

When the marama seed protein is broken down in its constituent proteins, the distribution is different from other legumes. It contains 23.3% albumins, 5% globulins, 15.5% prolamines, 7.7% alkali-soluble glutelins and 0.5% acid-soluble glutelins. Soybean contains 10% albumin and 90% globulin (Bower *et al*., 1988). The presence of an alcohol soluble protein, prolamine, in a legume seed is unusual (Boutler, 1977). It is in the water-soluble and saline-soluble protein fractions that the trypsin inhibitor activity is found.

Table 2.3

Amino acid composition of marama bean protein in percentage

Amino acid (% of protein)	Bousquet (1983)	Bower *et al.* (1988)
Essential amino acids		
Arginine	9.58	5.96
Cystine	*2.01*	*0.78*[a]
Histidine	3.57	2.25
Isoleucine	5.66	3.75
Leusine	7.70	5.58
Lysine	6.51	5.22
Methionine	*1.55*	*0.76 – 1.32*[a]
Phenylalanine	5.81	4.58
Threonine	4.97	2.89
Tyrosine	14.54	11.13
Tryptophan	1.29	1.55
Valine	6.12	4.18
Non-essential amino acids		
Alanine	N. D. [b]	2.98
Aspartic acid	N. D. [b]	10.31
Glutamic acid	N. D. [b]	14.89
Glycine	N. D. [b]	5.36
Proline	N. D. [b]	6.60
Serine	N. D. [b]	5.09
Ammonia	N. D. [b]	1.18
Total	N. D. [b]	95.04%

[a] *Unprotected. Upper limit determined by difference*
[b] *No data*

2.2.2 Fat / Oil

Marama oil, although not commercially available, has a pleasant odour and taste and can therefore be used in the food and cosmetic industries (Engels, 1984). Compared to soybeans, which contain about 17% fat, the amount of oil in marama beans is quite high and is present merely as monosaturated or unsaturated fatty acids.

The oil content of the dry marama seeds ranges from 36 – 43% and it approaches that of peanut (Wehmeyer, 1969). The oils of marama beans are reported to contain 31% unsaturated fatty acids (Ripperger–Suhler and Longenecker, 1982; cited in Biesele and Murray, 1983). Less than 5% of the fatty acids are present as the free fatty acids, the balance is esterified in the oil (Bower *et al.*, 1988) and the oil can be classified amongst the olive group of oils (Ketshajwang *et al*., 1998). The marama bean contains exceptionally high percentages of the polyunsaturated fatty acids, oleic and linolenic acid, as presented in Table 2.3. From a health perspective unsaturated (long chain) fatty acids are usually recommended over saturated fats for human consumption. The long chain fatty acids are more efficient and act faster in the body.

Table 2.4
Composition of fatty acids in marama bean oil in percentage[a]

Fatty acid (%)	Engelter and Wehmeyer (1970)	Bousquet (1983)	Bower et al (1988)
Myristic (C 14:1)	Trace	N.D. [b]	1.3 ± 0.3
Palmitic (C 16:1)	14.1	16.9	13.8 ± 5.0
Palmitoleic (C 16:2)	0.7	1.8	1.7 ± 0.3
Stearic (C18:0)	6.5	10.0	9.7 ± 7.0
Oleic (C18:1)	47.9	34.8	48.5 ± 8.0
Linoleic (C 18:2)	24.6	26.3	19.2 ± 9.5
Linolenic (C18:3)	N.D. [b]	2.3	2.0 ± 1.5
Arachidic (C 20:1)	3.3	3.4	2.8 ± 1.3
Arachidonic (C 20:2)	N.D. [b]	2.1	N.D. [e]

[a] *Data is normalized to 100%. Total fatty acid was 34 g/100g*
[b] *No data*

2.2.3 Minerals

According to analyses done by Bower *et al.* (1988), the marama bean is low in sodium (0.24 g/kg) and chlorine (0.3 g/kg) (Table 2.5).

Table 2.5

Mineral content of defatted marama bean meal

Mineral (amount per kg dry matter)	**Results – Bower *et al.* (1988)**
Sodium (g)	0.24 ± 0.02
Magnesium (g)	5.8 ± 0.1
Silicon (g)	40 ± 20
Phosphorus (g)	6.7 ± 0.4
Sulfur (g)	3.7 ± 0.2
Chlorine (g)	0.3 ± 0.1
Potassium (g)	16.0 ± 1.3
Calcium (g)	4.2 ± 0.2
Manganese (mg)	50 ± 20
Iron (mg)	40 ± 20
Zinc (mg)	100 ± 20

2.2.4 Protease inhibitors

At least five distinct trypsin inhibitors and two elastase inhibitors are present in the marama bean (Starcher et al, 1985). Trypsin is a protease, which belongs to a class of enzymes that degrades protein by breaking the bonds between amino acids. Proteases abound in human cells because cells need the help of different proteases for many functions (National Academy of Sciences, 2003).

Trypsin inhibitors were found in extremely high levels (239 TUI / mg) in the marama bean, almost twice as much as those reported for soybeans (Kakade *et al.*, 1973). Whether these high levels contributed to rats refusing their diet containing marama bean meal, could not be determined. However, roasted bean meal with no antitrypsin activity, was also refused.

The presence of a trypsin inhibitor is common in legumes, typically comprising 5 – 10% of the total protein (Ryan, 1981). According to Bower *et al*. (1988) the trypsin inhibitor activity in the marama bean can be destroyed by heat. Baking at 140 °C decreased the activity in the aqueous protein extracts by 80% and the saline protein extracts by 50%, giving a decrease of 70% in total trypsin inhibitor activity. Boiling fresh extracts from uncooked seed meal for 2 minutes in a microwave oven, reduced the aqueous extract's activity by 90% and the saline extract's activity by 75%. This amounted to an 80% reduction in activity. These findings are in agreement with those of Ripperger-Suhler and Longenecker (1982).

2.3 NUTRITIONAL CONTENT OF THE TUBER

The nutritional content of five month old tubers was reported by Biesele and Murray (1983) as follows: protein, 2.1%; fat, 0.14%; carbohydrate, 4.38%; ash, 0.42% and water, 92.1%.

Chemical and biochemical data of the marama tuber obtained from the Agriculture Laboratory, Windhoek, Namibia, are presented in Table 2.6. The tuber seems to be tasteless to animals (Pers. comm., Sandveld Research Farm, 2003) although it is high in crude protein (8.32%) and crude fiber (9.43%). The young tubers make an excellent vegetable dish for humans when cooked or roasted (Keith *et al*., 1975). Its texture is however fibrous and it becomes bitter when older.

Table 2.6

Selected nutrients of the marama tuber[a]

Nutrients (% of dry matter)	**Results**
Moisture (%)	78.04
Crude Protein (%)	8.32
Calcium (%)	1.72
Phosphorus (%)	0.04
Crude fibre (%)	9.43
Acid Detergent Fibre (%)	14.40
Neutral Detergent Fibre (%)	17.64
Fat (%)	0.19
Ash (%)	1.38
Organic matter digestibility (%)	53.0
Metabolizable energy (MJ/kg)	7.8
Gross Energy (MJ/kg)	15.5

[a] *Agriculture Laboratory, 2002*

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3

Description of the marama plant, tuber and beans

3.1 GENERAL DESCRIPTION

The marama plant (*Tylosema esculentum*) is a scrub with trailing stems, which arises from a large to very large underground tuber. It is a perennial legume that grows in open veld. Numerous annual prostrate runners emerge in spring. The runners can reach a length of six meters and bear y-shaped tendrils. The plant has characteristic, bi-lobed leaves, are glaucous-green and leathery, although soft, reddish-brown when older. The small yellow flowers produce rounded, oblong pods which are at first pale-pink in colour, change to apple-green, then to a dark purplish-pink and finally to brown. Powell (1987) has observed that the yellow flowers were visited by many types of insects, including bees, wasp and butterflies and it is suspected that bees affect cross-pollination.

3.2 OCCURRENCE IN ARID AND SEMI-ARID AREAS

The marama plant is native to dry areas with very limited average seasonal, and often variable rainfall. The plant's occurrence spreads in and around the Kalahari region of

Southern Africa, especially the sand veld regions of Namibia and Botswana, although it also occurs in the Gauteng and elsewhere in South Africa.

A marama plant, once established, will survive a year of no rain by drawing moisture out of deep soil and store the moisture in the tuber (National Academy of Sciences, 1979). The plant also seems to thrive under conditions of excessive moisture. In 1974 a record-breaking 1184 mm of rainfall was recorded at Maun, Botswana, although the rain destroyed the mongongo nut crop, Dobe area gatherers discovered a bumper of marama (Lee, 1979).

Fig 3.1: Marama in grassland – Sandveld Research Farm

3.3 SOIL CHARACTERISTICS

The marama plant grows in sandy soil. The analyses of soils samples from the Sandveld Research Station in the eastern part of Namibia where the marama plant grows in abundance, indicated that the soil is low in nutrients, especially nitrogen. The pH is normal to acid (Table 3.1). The marama plant was never seen growing in other types of soil, although it does grow between limestones (Pers. comm. Sandveld Research Farm, 2004).

Fig 3.2 : Marama plant in sandy soil – Eastern Namibia

Table 3.1

Soil analyses of Sandveld Research Farm in eastern Namibia*

Soil analyses (2001/2002 rainy season)	Sample 1	Sample 2	Sample 3
pH(water)	6.37	7.03	6.70
Electrical Conductivity (µS/cm)	12	16	11
Organic matter (%)	0.3	0.29	0.27
Phosphorus (ppm)	0.05	4.73	0.88
Potassium (ppm)	99	218	95
Calcium (ppm)	350	678	554
Magnesium (ppm)	50	122	98
Sodium (ppm)	15	25	10
Nitrogen (%)	0.019	0.020	0.018
Texture	Sand	Sand	Sand
Cation exchange capacity (cmol/kg)	4.00	4.59	4.99
Exchangeable cations:			
Potassium (cmol/kg)	0.149	0.236	0.157
Calcium (cmol/kg)	0.576	0.515	1.041
Magnesium (cmol/kg)	0.050	0.143	0.110
Sodium (cmol/kg)	1.330	0.000	0.600

**Agriculture Laboratory, MAWRD, 2002*

3.4 TUBER

Below the ground the marama plant produces a nutritive tuber. This tuber ensures the survival of the plant during periods of drought. The tubers also make an excellent vegetable dish and it has been reported that on a dry weight basis they contain as much as 9% protein (Story, 1958).

During experiments done by Keegan *et al.* (1981), it was found that the tubers developed rapidly and that within a period of six months tubers of approximately 72 g fresh weight were produced. These young juicy tubers had a very soft skin and when boiled had a slightly sweet taste. The texture of the boiled tubers was very similar to that of artichokes. The younger, smaller tubers make better eating, especially after being boiled or roasted (Keith *et al.*, 1975).

The National Academy of Sciences (1979) report mentioned the young tubers as a valuable food source. The proper term for the tubers is *sekophane* in Tswana or n//n in !Kung. The tubers have a “sweet pleasant flavor and make a good vegetable dish” (National Academy of Sciences, 1979).

The perennial tuber, which with time becomes fibrous, can weigh as much as 12 kg after 10 years (Watt J M *et al.*, 1962). Botha (pers. comm.) as cited in Keith *et al.* (1975) reported digging up a tuber with a mass of approximately 300 kg. The large tubers are difficult to chew, although they still have a high moisture content. The moisture of the old tubers can be extracted by pounding pieces of it in a suitable container. Larger and presumably older tubers are avoided, because they are tough

and bitter, although this may not be invariably be the case (Biesele and Murray, 1983; Story, 1958). A bit of trivia – reportedly it was the tuber from which water was squeezed in the movie “The Gods must be crazy” (ECHO, 1999).

Fig 3.3: Marama tuber digged out

3.5 STEMS

The long vine-like stems grow continuously throughout the summer months. The marama plant is a creeper rather than a climber. “They hug the ground, presumably avoiding dry winds” (ECHO, 2003). During winter the stems dry back, but the underground tuber produces new stems when it becomes warmer again.

The vines grow primarily in sandy soil, although Keith and Renew (1975) observed that the vines grow in outcrops of limestone. Vierich (1980) reported seeing old plants whose vines cover a radius of 25 meters.

Fig 3.4 : Marama vines growing in different directions

3.6 LEAVES

The marama plant is a relative of the Bauhinia tree, so the leaves have the characteristic butterfly shape of this beautiful tree (Fletcher, 1998). In early summer the leaves are bluish green, but turn to reddish brown during the winter.

Fig 3.5 : Marama leaves on a stem in summer

3.7 FLOWERS

The marama flower has attractive yellow petals**.** The flowers are approximately 4 cm across and are produced in clusters of three to nine, mostly on the distal two-thirds of the stems (Powell, 1987). Although the flowers are not trumpet-, bell-, or tube-shaped as commonly found in heterostylous species, the arrangement of the staminodes and two anthers surrounding the style, helps direct the potential pollinator to the correct position for pollen transfer (Hartley et al. 2002). The marama plant can spread over a large area and therefore pollination is often likely to be from the nearest plant (Levin, 1978). It has been shown that the pollen of the marama plant remains viable under laboratory conditions for seven days (Monoghan, 1993). The lack of pollinators may explain the abundance of flowers in this species and the surplus flowers may thus act as a reward to attract scarce pollinators (Stephenson, 1981).

Fig 3.6 : Marama flowers on stem

3.8 PODS

Young pods of approximately six cm long are light green, but ripen in late autumn, turning into brown woody pods with two or more chestnut – brown seeds inside (National Academy of Science, 1979). The pods usually contain two seeds but can produce as many as six (Watt *et al.*, 1962). Pods have tough, leathery valves that dry and harden as they mature and normally dehisce when dry (Powell, 1987). These fruits yield between 100g and 300g seeds per plant, i.e. between 0.1 tons and 0.3 tons per ha resulting in approximately 50 000 seeds per ha (Lawlor, 2004).

Fig 3.7: Marama pod on stem

3.9 SEEDS (BEANS)

The primary agronomic potential of the marama bean is based upon the high nutrient value of the seeds (Powell, 1987). Hartley *et al.* (2002) however demonstrated that the seed set is very low in this species. This may be a way of the species to adapt to an environment in which rainfall is scarce.

The dark brown seeds, which do not form a very hard coat, weigh approximately 2.4 g upon maturity. Of this, the seed coat contributes about 49% and the embryo about 51% (Keegan and Van Staden, 1981). The seeds are about the size of a thumbnail, with hard woody shells and contain firm, bi-lobbed cream-coloured oily kernels. Normally a pod bears two seeds, but three seeds per pods have been noticed (Pers. comm.. Sandveld Research Farm, 2004). No toxic substances have been detected in the any of the seed components (Watt *et al.*, 1962). The seeds are apparently non-dormant and they germinate within 2-6 days when incubated in a moist substrate (Story, 1958).

Fig 3.8 : Matured marama beans in pod

3.10 CULTIVATION

ECHO (1999) published some hints on the germination of the seeds. Seeds should be kept warm (room temperature). When wetted they swell tremendously. The embryo and endosperm absorb water and then germination starts. To promote water absorption, the outside can be scratched with a file. Germination will not be hastened when dropped into water – it should only be planted in moist soil (neutral to acid, sandy) or potting medium.

3.11 COOKING / ROASTING AND EATING

The young green nuts can be boiled as a green vegetable and are similar to cultivated green peas (National Academy of Sciences, 1979), but this practice is not common (Pers. comm. Namibian and Botswana inhabitants, 2002). The ripe nuts also make a good soup when soaked and prepared in the same way as split-peas and added to bouillon (Keith *et al.*, 1975). Strong trypsin inhibition is found in raw seeds, but this negative nutritional factor is averted by cooking them (Bower *et al*., 1988).

The seeds which are produced in autumn, are never eaten raw by the indigenous population as they are tasteless and of a slimy texture. When roasted they have a nutty flavour comparable to that of cashew nuts (Rachie, 1979). The oil, with 31% unsaturated fatty acids, are sometimes used as a cooking oil by the indigenous people (Biesele and Murray, 1983).

Lee (1979) describes the way the beans are used: Unripe beans may be sun-dried before further processing. A batch of 50 or so beans is roasted in the shell for a few minutes in the hot ashes and sand of the cooking fire. Occasionally, a bean explodes, but without much damage. Slight bursts of steam from the roasting beans indicate that they are ready for eating. The beans are normally cooled down before consumed (Pers. comm. Namibian and Botswana inhabitants, 2002).

Alternatively the beans are traditionally roasted within a rounded cast-iron pot on top of the fire. Beans are roasted for approximately 12 minutes, whilst covered with sandy soil (Pers. comm. Namibia and Botswana Inhabitants, 2002). Beans are only embedded in the pot once the soil has reached a temperature where it burns the fingers when touching.

The beans are removed from the ashes or pot and opened with a single light tap of a rock or stick. Each bean comes apart easily in two halves. Eaten whole, the beans have a rich, strong, nutty flavour. Alternately, the shelled beans may be pounded into a mortar and then mixed with hot water and eaten as soup, porridge or a cocoa-like beverage (Lee, 1979).

Fig 3.9 : Traditional roasting of marama beans

3.12 BROWSING BY LIVESTOCK

Cattle apparently browse on the stems and leaves of the marama bean plant (National Academy of Sciences, 1979). However. no scientific study was so far conducted to proof this.

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4

Nutrient content of the marama bean

4.1 INTRODUCTION TO CHAPTER 4

To help feed an increasing population and to alleviate under-nutrition, to make marginal lands more productive and to meet challenging resource needs, little-known plant species should be investigated. It would build a more stable food supply for drought-stricken Africa and other parts of the world (Vietmeyer, 1986).

The marama bean plant (*Tylosema esculentum*) is a perennial legume which produces beans nutritious for humans. It is native to dry areas with little seasonal rainfall and in these difficult environments, legumes are of particular importance in subsistence agriculture. Legumes can survive on barren sites that are nitrogen–deficient and their residues leave the soil enriched with nitrogen.

Marama beans are seldom eaten raw, because of the slimy and tasteless texture (Rachie, 1979). The beans contain a trypsin inhibitor, but the trypsin inhibitor activity is normally destroyed by heat (Bower *et al*., 1988). After roasting, they have a delicious nutty flavour that has been compared to roasted cashew nuts and are even used in Southern Africa as a substitute for almonds.

The purpose of this study was to determine the nutrient content of the marama beans in order to determine its possible contribution to alleviate under-nutrition in two Southern African regions, namely Namibia and Botswana.

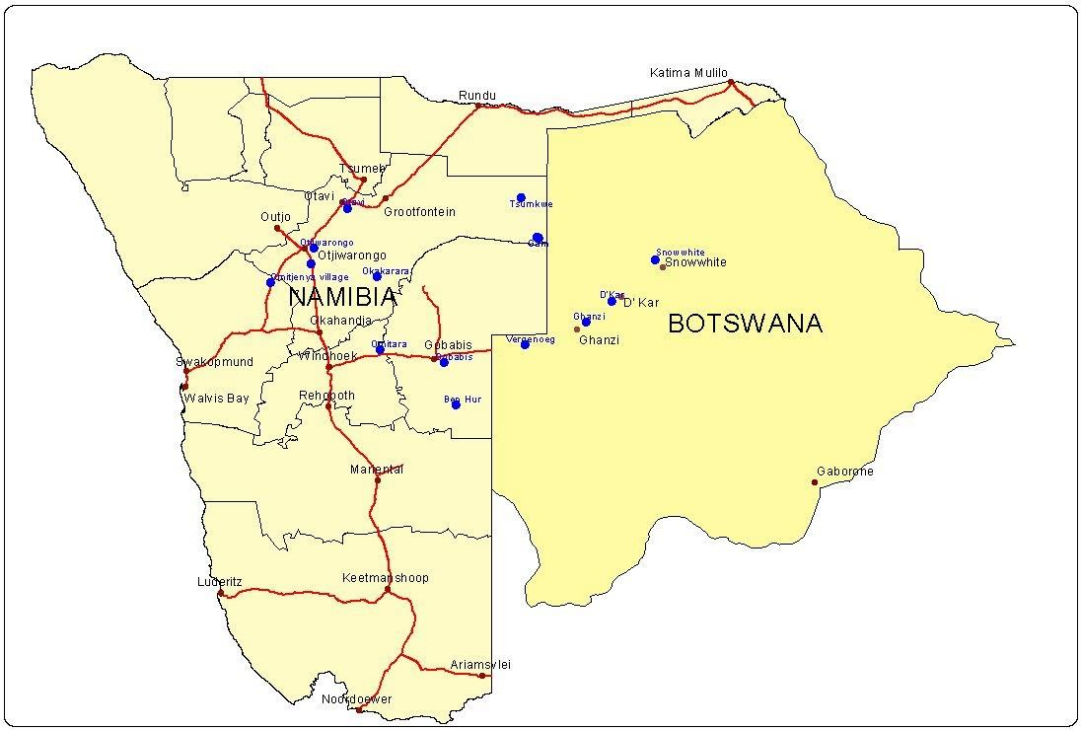
4.2 MATERIALS AND METHODS

4.2.1 Sampling sites

Locations within Namibia and Botswana, where the marama bean plant grows in natural abundance, were chosen as sampling sites for the study of the quality characteristics of the marama bean plant (Fig. 4.1), namely:

- The Sandveld Research Farm area, east of Epukiro, 60 km northeast of Gobabis, in eastern Namibia (22 01’ 04’’ South, 19 08’ 13” East).

- The Ghanzi / D’ kar area in western Botswana (21 25’ 03” South, 21 23’ 28” East / 21 18’ 45’’ South, 21 33’ 41” East).



- ***marama sites***

Fig 4.1

Map of Botswana and Namibia showing the distribution of marama bean plants in Namibia and Botswana.

4.2.2 Sampling

For the chemical analyses sampling was done during the rainy seasons (December, January to March) of three consecutive years, 2001/2002, 2002/2003, 2003/2004. Matured beans were collected from within a radius of 100 km of each site. Collections were made ± two to five kilometers apart. Samples were sealed in paper bags and numbered according to location and date. No young (green) beans were collected, as it was stated by Rachie (1979) that beans were never eaten young and raw (green).

The beans were oven-roasted in the laboratory by roasting them in sand baths filled with ordinary sand that was pre-heated to a temperature of 160 °C at an oven temperature of 180 °C. The beans were roasted in the pre-heated sand for 10 minutes, left over night to cool and then shelled to remove the kernels. The roasting of the beans was done in ten Mielé ovens (model H217) and the temperature of the sand was measured with a hand-model Kane-Mane 1012 type temperature probe (Campbell *et al*., 1980), as part of the standard procedure.

4.2.3 Experimental procedure

A whole spectrum of chemical analyses was conducted on the samples from the first year (2001/2002), in order to determine the high or low abundance of nutrients within the marama beans. Nutrients having values close to the recommended daily intake (Food and Nutrition Board, Institute of Medicine, National Academies) were again analyzed for in the second (2002/2003) and third (2003/2004) year of the study.

Chemical analyses were conducted at the Analytical Laboratories of the ARC (Agricultural Research Centre) Irene, Pretoria; ISCW (Institute for Soil, Climate and Water), Pretoria and SGS (Sociéte Générale de Surveillance), Midrand, South Africa. Accredited methods are accredited by SANAS (South African National Accreditation System) according to ISO 17025.

The content of the shelled nuts of the roasted marama bean samples (approximately five kg) was chopped, mixed and grinded prior to being chemically analyzed, to ensure

an even mixture of the roasted samples. Chemical analyses were conducted in duplicate.

4.2.4. Proximate analyses

Proximate analyses was carried out to determine the percentages of total moisture, fat, protein (N x 6.25 – principle: 16% N in protein) and ash according to the accepted AOAC (1999) methods which will be described briefly.

- **Determination of dry matter (ARC Irene accredited method)**

The dry matter content is the residue expressed in percent by weight, which remains after the drying process. Dry matter is the sample without water. The moisture from a sample is driven off by use of heat. Weight loss is used to calculate dry matter content (AOAC, 1999).

- **Determination of ash (ARC Irene accredited method)**

The total ash is the inorganic matter of a sample. The organic matter of a sample is removed by heating at 550 ^{0}C overnight. The remaining residue is inorganic matter (ash) (AOAC, 1999).

- **Determination of nitrogen (ARC Irene accredited method)**

The method is based on the Dumas Combustion method, which is approved, by the AOAC. The sample is combusted at ± 1100 °C – 1350 °C and 10 cm^3 of the sample gas is analyzed. A thermal conductivity cell detects the difference in thermal conductivity caused by the presence of Nitrogen (LECO Africa). Total protein is calculated N x 6.25 (principle: 16% N in protein).

- **Determination of fat (Soxtec method – ARC Irene accredited method)**

Fat is organic chemical compounds made up of components known as fatty acids and glycerol. Most of the fat is soluble in petroleum ether (PE). The fat in the sample dissolves in the ether at boiling temperature. The ether is evaporated at 90 °C. The fat is left in the beaker. Weight gain is used to calculate fat content. Bounded fat is not soluble in PE and has to be broken down by hydrolysis (Soxtec hydrolyzing system manual).

- **Determination of crude fiber (ARC Irene accredited method)**

Crude fiber is the parts of cell-walls of a plant that can not be digested by the enzymes in the intestine. Crude fiber is determined gravimetrically, after chemical digestion and solubilisation of other compounds present (i.e. protein, starch, and other digestible /

carbohydrates) with diluted sulphuric acid and sodium hydroxide. The fiber mass is then corrected for ash content after ignition (AOAC, 1999).

- **Determination of non-structural carbohydrates (ARC Irene non-accredited method)**

Non structural carbohydrates are analyzed as reducing sugars after complete enzymatic hydrolysis to monosaccharide. This method entails the gelatinization of all the starch to glucose and determination of the glucose content by spectrophotometric measurement (ARC, Pretoria, in-house method).

- **Determination of dietary fiber (ARC Irene accredited method)**

Samples undergo sequential enzymatic digestion by heat α-amylase, protease amyloglycosidase to remove starch and protein. For total dietary fiber (TDF), enzyme digestate is treated with alcohol on precipitate soluble dietary fiber before filtering, and TDF residue is washed with alcohol and acetone, dried and weighed. TDF residue values are corrected for protein, ash and blank (Method 991.43 and 985.29, AOAC, 1999; Prosky *et al*., 1985).

4.2.5 Vitamins

- **Determination of fat soluble vitamins**

Vitamin A / carotene (ARC Irene accredited method): The test material is alkaline saponified and the unsaponifiable matter extracted with ether. An aliquot of the ether extract is evaporated and dissolved in hexane. The analysis is performed on a HPLC with a silica column and hexane as the mobile phase. The carotene is detected with a UV detector at 453 nm. Regression analysis was performed and the carotene calculated (Thompson *et al.*, 1980; Manz and Phillip, 1981; Fox, 1985).

Vitamin D (SGS Midrand non-accredited method) and vitamin E (ARC Irene non-accredited method): Samples are prepared by mixing with hot 50% hydrochloric acid solution and 99.9% ethanol, followed by shaking and liquid – liquid extraction with hexane. The hexane extract is then injected into a HPLC system using a normal phase silica column with isocratic elution and UV detection. The vitamins are then identified on the basis of retention times as compared to standards. Quantification is done by using external standards (Thompson *et al*., 1980; Hulshof, 2002).

- **Determination of water soluble vitamins**

Thiamin **(vitamin B_1)** and riboflavin **(vitamin B_2)** were determined by high performance liquid chromatography and fluorescence detection **(ARC Irene accredited methods)**. After autoclave extraction, samples are derivatized to form thiochrome (a highly fluorescent oxidised product of thiamin). Riboflavin is naturally fluorescent (Wimalasiri and Wills, 1985). Interferences are removed by solid phase extraction on a C_{18} cartridge and the extract analysed with HPLC and reversed phase separation (Sims

and Shoemaker, 1993). The pre-column or postcolumn derivatization of B_1 to thiochrome is normally essential to improve the detection sensitivity in the HPLC analysis (Ollilainin *et al.,* 1993).

Cyanocoblamine **(vitamin B_{12})** levels were determined according to the accepted AOAC method (1999) at the **SGS Midrand Laboratory**.

Niacin **(vitamin B_3)** was determined by High Performance Liquid Chromatography coupled with solid phase extraction cleanup (Chan Mo Cho, 2000) at the **SGS Midrand Laboratory**. Pyridoxine **(vitamin B_6)** was determined according to the Microbiological Method MI 002 in beverage, food, vegetables and fruit products by microbiological assay and folic acid was determined according to the Microbiological Method MI 003 in food by microbiological assay **(SGS Midrand Laboratory)**.

- **Determination of vitamin K (SABS Test house non-accredited method)**

The sample is dispersed in 0.01 N HCl, diluted with ethanol and extracted into n-hexane. The sample is evaporated to dryness and diluted in a mixture of THF / ethanol. The solution is injected onto a reverse phase HPLC column and UV detection at 248 nm (AOAC, 1999).

4.2.6 Minerals / Trace minerals (ISCW Pretoria non-accredited methods)

A suitable mass (0.5g to 1g) of sample is digested with 7ml HNO_3 (conc. nitric acid) and 3ml $HClO_4$ (perchloric acid) at temperatures up to 200 ºC and brought to volume in a 100ml volumetric flask.

Phosphorus (P): The phosphorus concentration in the solution of the digested sample is determined spectrophotometrically as the yellow phospho-vanado–molybdate complex (Cavell, 1955).

Potassium (K) and Sodium (Na): An aliquot of the digest solution is used for determination of K and Na by flame emission spectroscopy (flame photometer) in a LPG-air flame, using Li ($LiNO_3$) as an internal standard. The method has been automated by means of a flow system.

Other minerals **(chlorine (Cl), iodine (I), fluorine (F))** and selected trace-minerals **(iron (Fe), manganese (Mn), zinc (Zn), copper (Cu), chromium (Cr), cobalt (Co), selenium (Se), molybdenum (Mo), lead (Pb), vanadium (V), nickel (Ni), barium (Ba), strontium (Sr), tin (Sn), titanium (Ti))** are analyzed with flame atomic absorption spectrophotometry (AAS) and Air-Acetylene Flame or Nitrous Oxide-Acetylene Flame at different wavelengths depending on the mineral / metal (AOAC, 1999, Zasosi, R. J., 1977, Flame methods Manual for Atomic Absorption).

4.2.7 Amino acids (ARC Irene accredited method)

The method used for the analysis of amino acids involves acid hydrolysis, pre-column derivatisation, seperation by HPLC and detection using a fluorescence detector (Einarsson, S. *et al*. 1983, Cunico *et al.*, unpublished).

- **Tryptophane (ARC Irene accredited method)**

The method used for the analysis of tryptophane involves enzymatic hydrolysis, seperation by HPLC and detection using a fluorescence detector (De Vries *et al*., 1980).

- **Cystine / Cysteine (ARC Irene accredited method)**

The method used for the analysis of cystine/cysteine involves oxidization of cystine/cysteine to cysteic acid, acid hydrolysis, pre column derivatisation, seperation by HPLC and detection using a fluorescence detector (Gehre *et al*., 1985, Cunico *et al*., unpulished; Williams, 1984).

4.2.8 Fatty acids (ARC Irene accredited method)

A gas chromatographic method is used for the determination of fatty acids. The fat extracts are trans-methylated with methanol-potassium hydroxide. Fatty acid methyl esters are extracted with n-hexane and analyzed by gas liquid chromatography (Christopherson, 1969).

4.2.9 Total cholesterol (ARC Irene accredited method)

Total fat is extracted from the sample using the Soxtec extraction. The extract is dried, saponifies, extracted again with hexane and analyzed on a gascromatograph with flame ionization detection (Smuts *et al.*, 1992; Christopherson *et al.*, 1969; Soxtec hydrolyzing system manual).

4.3 RESULTS

4.3.1 Analyses of marama beans from 2001 rainy season

4.3.1.1 Proximate analyses (2001)

According to the proximate analyses (Table 4.1), the marama bean contains 34.71% (34.71 g/100g) protein and is a good source of protein. It compares well to other marama literature with 34.3 % protein (Wehmeyer, 1969). The daily recommended allowance for protein is 19 g per day for children age five, 46 g per day for women age 30 and 56 g per day for men age 30 (National Academy of Sciences, 2001).

The fat content of the Namibia and Botswana bean is similar (39.93% and 40.18% respectively) and compares well to other literature with between 36-43% fat (Wehmeyer,1969). The amount of energy contained in the marama bean is 2.28 MJ/100g, which compares well to the study of Bower *et al.* (1988) with 2.66 MJ/100g.

Table 4.1

Proximate analyses of the Namibia and Botswana marama beans (2001)

Nutrient (% of dry matter)	Namibia bean[1]	Botswana bean[2]	Mean [1,2] & standard deviation
Dry matter	95.93	96.50	96.22 ± 0.40
Ash	3.29	3.08	3.19 ± 0.15
Protein	33.97	35.44	34.71± 1.04
Fat	39.93	40.18	40.06 ± 0.18
TNC*	13.64	14.50	14.07 ± 0.61
Fibre	4.34	3.53	3.94 ± 0.57
Total dietary fibre	50.85	50.77	50.81 ± 0.06
Energy (MJ/100g)	2.26	2.29	2.28 ± 0.02

**TNC = Total non-structural carbohydrates*

4.3.1.2 Analyses of vitamins (2001)

Of the many nutrients that the marama bean contains (Table 4.2), of importance is vitamin A (0.27 mg/100g), essential for proper eyesight. The daily recommended allowance is 0,5 mg per day for children age five, 1.0 mg per day for men, age 30 and 0.8 mg per day for women, age 30 (National Academy of Sciences, 2001). The marama bean contains folic acid (0.14 mg/100g), especially needed by women during pregnancy. The daily recommended allowance is 0.4 mg per day for children age five, 0.7 mg for women age, 30 and 0.9 mg for men, age 30 (National Academy of Sciences, 2001). It contains vitamin B_{12} (0.004 mg/100g), normally only produced in animal tissue. The daily recommended allowance for vitamin B_{12} is 0.001 mg per day for children age 5 and 0.002 mg per day for women and men age 30 (National Academy of Sciences, 2001).

Table 4.2

Vitamins analyses of the Namibia and Botswana marama beans (2001)

Vitamins	Namibia bean[1]	Botswana bean[2]	Mean [1,2] & standard deviation
Vit A (mg/100g)	0.29	0.25	0.27 ± 0.03
Vit B_1 (mg/100g)	0.45	0.3	0.38 ± 0.11
Vit B_2 (mg/100g)	0.048	0.07	0.06 ± 0.02
Vit B_3 (mg/100g)	11.12	7.30	9.21 ± 2.70
Vit B_6 (mg/100g)	1.67	1.74	1.71 ± 0.05
Vit B_{12} (mg/100g)	0.005	0.003	0.004 ± 0.00
Vit D (IU/100g)	126.4	139.32	132.9 ± 9.14
Vit E (mg/100g)	7.02	5.51	6.27 ± 1.07
Vit K (mg/100g)	0.226	0.212	0.22 ± 0.01
Folic acid (mg/100g)	0.188	0.09	0.14 ± 0.07

4.3.1.3 Mineral analyses (2001)

As presented in Table 4.3, the marama bean is high in calcium (241 mg/100g), a mineral essential for maintaining the health of bones and teeth. The recommended daily allowance for calcium is 800 mg per day for children age five, 1000 mg per day for women and men age 30 (National Academy of Sciences, 2001). It contains iodine (0.06 mg/100g), needed for cognitive development. The daily recommended allowance for iodine is 0.09 mg per day for children age five and 0.150 mg per day for women and men age 30. It is adequate in magnesium (274.5 mg/100g), a catalyst needed for many biochemical and physiological processes in the human body. The recommended daily allowance for magnesium is 130 mg per day for children age five, 310 mg per day

for woman age 30 and 310 mg per day for men age 30 (National Academy of Sciences, 2001).

Table 4.3

Mineral analyses of the Namibia and Botswana marama beans (2001)

Mineral	Namibia bean[1]	Botswana bean[2]	Mean [1,2] & standard deviation
Phosphorus (mg/100g)	503	406	454 ± 68.59
Calcium (mg/100g)	274	208	241 ± 46.67
Magnesium (mg/100g)	273	276	274.5 ± 2.12
Potassium (mg/100g)	954	836	895 ± 83.44
Sodium (mg/100g)	64.5	63.0	63.75 ± 1.06
Chloride (mg/100g)	69.4	54.5	61.95 ± 10.54
Fluorine (mg/100g)	3	3	3.00 ± 0.00
Iodine (mg/100g)	0.048	0.064	0.06 ± 0.01

4.3.1.4 Analyses of trace minerals (2001)

Analyses of trace minerals in the marama bean (Table 4.4) show that that it contains iron (3.95 mg/100g), essential to prevent nutritional anemias. The daily recommended allowance for iron is 10 mg per day for children age five, 18 mg per day for women age 30 and 8 mg per day for men age 30 (National Academy of Sciences, 2001). The marama bean contains zinc (6.2 mg/100g), a trace mineral essential for growth and protein metabolism. The daily recommended allowance for zinc is 5 mg per day for children age five, 8 mg per day for women age 30 and 18 mg per day for men age 30 (National Academy of Sciences, 2001).

Table 4.4

Analyses of trace minerals of the Namibia and Botswana marama beans (2001)

Trace minerals	Namibia bean[1]	Botswana bean[2]	Mean [1,2] & standard deviation
Iron (mg/100g)	4.1	3.8	3.95 ± 0.21
Manganese (mg/100g)	1.9	1.8	1.85 ± 0.07
Zinc (mg/100g)	6.4	6.0	6.2 ± 0.28
Copper (mg/100g)	1.18	0.89	1.04 ± 0.21
Chromium (mg/100g)	0.07	0.04	0.06 ± 0.02
Cobalt (mg/100g)	0.02	0.016	0.02 ± 0.00
Selenium (mg/100g)	0.071	0.087	0.08 ± 0.01
Molybdenum (mg/100g)	0.015	0.0167	0.02 ± 0.00
Lead (mg/100g)	0.097	0.148	0.12 ± 0.03
Vanadium (mg/100g)	0.008	0.027	0.02 ± 0.01
Nickel (mg/100g)	0.198	0.328	0.12 ± 0.12
Barium (mg/100g)	0.392	0.783	0.59 ± 0.28
Strontium (mg/100g)	0.272	0.114	0.7 ± 0.61
Tin (mg/100g)	1.59	3.79	2.69 ± 1.56
Titanium (mg/100g)	1.12	2.14	1.63 ± 0.72

4.3.1.5 Fatty acid analyses (2001)

The marama bean contains high levels of the fatty acids oleic acid (42.16% of total fatty acids) and linoleic acid (31.11% of total fatty acids) as presented in Table 4.5. These findings are in agreement with previous studies, showing values of 48.5% oleic acid, 19.2% linoleic acid (Bower *et al*., 1988) and 34.8% oleic acid, 26.3% linoleic acid (Bousquet, 1983). From a health perspective are these long chain unsaturated fats regarded as being healthier than saturated fats.

Table 4.5

Percentage fatty acids of the total fatty acids in the Namibia and Botswana marama beans (2001)

Fatty Acid (% of total fatty acids)	Namibia bean[1]	Botswana bean[2]	Mean [1,2] & standard deviation
C16:0 (Palmitic acid)	13.98	13.62	13.80 ± 0.25
C16:1 (Palmitoleic acid)	0.58	0.53	0.56 ± 0.04
C18:0 (Stearic acid)	7.63	8.47	8.05 ± 0.59
C18:1 (Oleic acid)	38.41	45.91	42.16 ± 5.30
C18:2 (Linoleic acid)	34.89	27.32	31.11 ± 5.35
C18:3 (Linolenic acid)	0.00	0.00	0.00
C20:0 (Arachidic acid)	2.56	2.43	2.50 ± 0.09
C20:1 (Arachidonic acid)	0.39	0.41	0.40 ± 0.01
C22:0 (Behenic acid)	1.56	1.31	1.44 ± 0.18

4.3.1.6 Amino acids (2001)

When analyzed for amino acids, the amino acids methionine and cystine showed to be limiting in the marama bean (Table 4.6). This is in agreement with previous literature on the marama bean, where the amino acids methionine and cystine were also found to be limiting (Bower *et al*., 1988 and Bousquet, 1983). Methionine is one of the nine essential amino acids that cannot be synthesized by the human body. Cystine is regarded as a semi-essential amino acid. The marama bean is thus not a complete protein, due to its limiting amino acids.

Table 4.6

Amino acid analyses of the Namibia and Botswana marama beans (2001)

Amino acid (g per 100g)	Namibia bean[1]	Botswana bean[2]	Mean [1,2] & standard deviation
Arginine	1.462	2.025	1.74 ± 0.4
Serine	0.980	0.956	0.97 ± 0.02
Aspartic acid	2.200	2.393	2.30 ± 0.14
Glutamic acid	3.076	3.270	3.17 ± 0.14
Threonine	1.125	1.132	1.13 ± 0.00
Glycine	1.268	1.407	1.34 ± 0.10
Alanine	1.260	1.086	1.17 ± 0.12
Tyrosine	3.362	3.376	3.37 ± 0.01
Proline	1.349	1.217	1.28 ± 0.09
Methionine	*0.098*	*0.005*	*0.05 ± 0.07*
Valine	1.062	1.148	1.11 ± 0.06
Phenylalanine	1.088	1.114	1.10 ± 0.06
Isoleucine	1.240	1.057	1.15 ± 0.13
Leucine	1.532	1.237	1.38 ± 0.21
Histidine	0.601	0.728	0.66 ± 0.09
Lysine	1.046	1.069	0.56 ± 0.72
Tryptophan	0.700	0.669	0.68 ± 0.02
Cystine	*0.545*	*0.563*	*0.55 ± 0.01*

4.3.2 Comparative analyses of selective nutrients from three rainy seasons (2001, 2002, 2003)

Nutrients were selected according to adequacy in recommended daily dietary allowance for children age five, men age 30 and women age 30. Chemical analyses were thereafter conducted on samples from Namibia and Botswana roasted marama beans from two more consecutive years (2002, 2003).

4.3.2.1 Proximate analyses (2001, 2002, 2003)

Values obtained from the proximate analyses on roasted Namibia and Botswana marama beans from the rainy seasons of 2001, 2002, and 2003 were similar, as presented in Table 4.7.

Table 4.7

Comparison of proximate analyses of Namibia[1] and Botswana[2] marama beans from rainy seasons 2001, 2002, 2003

Nutrient (% of dry matter)	Namibia bean 2001	Namibia bean 2002	Namibia bean 2003	Botswana bean 2001	Botswana bean 2002	Botswana bean 2003	Mean [1,2] & standard deviation
Ash	3.29	3.35	3.51	3.08	3.44	3.49	3.36±0.03
Dry matter	95.93	97.95	97.73	96.50	98.00	97.90	97.4±0.14
Moisture	4.07	2.05	2.27	3.50	2.00	2.10	2.67±0.19
Fat	39.93	37.06	31.54	40.18	33.63	31.93	34.05±1.7
Protein*	33.97	36.94	34.81	35.44	36.14	41.33	36.44±1.69
TNC**	13.64	9.35	10.91	14.50	10.33	12.37	11.85±0.78

**Protein content = N x 6,25*
*** Total non-structural carbohydrate*

4.3.2.2 Nutrients selected according to RDA (2001, 2002, 2003)

The chemical content of the analyses from the different seasons were similar, except for chromium and vitamin B12, which showed higher values during 2002 and 2003 for both the Namibia and Botswana marama beans. The amount of zinc was also lower in 2002 and 2003 than in 2001, for both the Namibia and Botswana marama beans.

Table 4.8

Comparison of RDA selected nutrients of the Namibia[1] and Botswana[2] marama beans from rainy seasons 2001, 2002, 2003

Nutrient	Namibia bean 2001	Namibia bean 2002	Namibia bean 2003	Botswana bean 2001	Botswana bean 2002	Botswana bean 2003	Mean [1,2] & standard deviation
Vit B_6 (mg/100g)	1.670	0.860	1.960	1.740	1.600	1.540	156±0.09
Vit B_{12} (mg/100g)	0.005	0.020	0.010	0.003	0.020	0.020	0.01±0.00
Chromium (mg/100g)	0.07	0.225	0.245	0.04	0.264	0.233	0.18±0.00
Copper (mg/100g)	1.18	1.211	1.20	0.89	0.965	1.342	1.13±0.09
Iron (mg/100g)	4.10	6.470	4.99	3.80	4.90	4.48	4.79±0.56
Zinc (mg/100g)	6.40	4.03	3.87	6.00	4.20	3.63	4.69±0.11
Iodine (mg/100g)	0.048	0.054	0.051	0.064	0.127	0.068	0.07±0.02

4.3.2.3 Fatty acids (2001, 2002, 2003)

The fatty acid composition of the Namibia and Botswana marama beans was similar throughout all the rainy seasons as presented in Table 4.9. On average the beans contained 42.03% (of total fatty acids) oleic acid and 32.12% (of total fatty acids) of linoleic acids.

Table 4.9

Comparison of percentages of fatty acids of the Namibia[1] and Botswana[2] marama beans from rainy seasons 2001, 2002, 2003

Fatty Acid (% of total fatty acids)	Namibia bean (2001)	Namibia bean (2002)	Namibia bean (2003)	Botswana bean (2001)	Botswana bean (2002)	Botswana bean (2003)	Mean [1,2] & standard deviation
C16:0 (Palmitic acid)	13.98	12.44	12.81	13.62	12.96	11.24	12.84±0.33
C16:1 (Palmitoleic acid)	0.58	0.44	0.48	0.53	0.51	0.28	0.47±0.04
C18:0 (Stearic acid)	7.63	8.22	7.32	8.47	7.85	7.64	7.86±0.19
C18:1 (Oleic acid)	38.41	40.88	42.36	45.91	41.30	43.31	42.03±2.09
C18:2 (Linoleic acid)	34.89	33.84	32.02	27.32	32.38	32.48	32.16±2.02
C18:3 (Linolenic acid)	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C20:0 (Arachidic acid)	2.56	2.48	2.54	2.43	2.62	2.54	2.53±0.00
C20:1 (Arachidonic acid)	0.39	0.32	0.38	0.41	0.33	0.38	0.37±0.01
C22:0 (Behenic Acid)	1.56	0.65	1.40	1.31	1.37	1.41	1.28±0.11

***Percentage of total fatty acids in beans*

4.4. DISCUSSION

Marama beans are high in nutrients such as protein, fat, vitamins and minerals needed for the supplementation of diets of persons suffering from under-nutrition (Table 4.10).

Table 4.10

Nutrient content of selected essential nutrients in the Namibia[1] and Botswana[2] marama bean

Nutrient (dry matter basis)	Namibia[1] marama bean	Botswana[2] marama bean	Mean [1,2] & standard deviation
Protein (%)	35.24	37.64	36.44 ± 1.69
Fat (%)	32.84	35.25	34.05 ± 1.70
Iron (mg/100g)	5.2	4.62	4.79 ± 0.56
Iodine (mg/100g)	0.051	0.086	0.07 ± 0.02
Vitamin A (mg/100g)	0.29	0.25	0.27 ± 0.03
Vitamin B_{12}(mg/100g)	0.005	0.003	0.01 ± 0.00
Oleic acid (C18:1) as % of total fatty acids	40.5	43.5	42.03 ± 2.09
Linoleic acid (18:2) as % of total fatty acids	33.6	30.7	32.16 ± 2.02
Cholesterol	None	None	None

Nutrient density is used for assessing the nutritional quality of a food. The higher a food's nutrient density, the better it is as a nutrient source. Comparing nutrient densities estimates the relative nutritional quality. Table 4.10 shows the nutrient densities of selected vitamins and minerals of the marama bean. Values >1 indicates a high nutrient density. It is clear that this is an excellent source of calcium, vitamin A, vitamin B_3, vitamin B_6, folic acid, vitamin B_{12}, vitamin E, iron, zinc and iodine.

Table 4.11

Nutrient densities* of selective nutrients of the marama bean for different life stage groups

Nutrient	Child ** 5 years of age	Men*** 30 years of age	Women**** 30 years of age
Calcium	*0.99*	*1.61*	*1.21*
Vitamin A	*1.79*	*1.44*	*1.37*
Vitamin B_1	0.7	0.68	0.7
Vitamin B_2	0.18	0.19	0.18
Vitamin B_3	*2.54*	*2.59*	*2.48*
Vitamin B_6	*4.7*	*4.16*	*3.95*
Folic acid	*6.13*	*3.71*	*3.12*
Vitamin B_{12}	*42.78*	*34.47*	*26.14*
Vitamin E	*2.96*	*3.34*	*3.17*
Cr	16.98	27.35	20.75
Cu	2.5	2.01	1.53
Fe	*1.59*	*2.55*	*1.29*
Zn	*1.55*	*1.67*	*1.58*
Iodine	*2.52*	*2.44*	*1.85*

* *Dietary References Intakes, National Academy of Sciences, 2001*
** *RDA for energy 7 531.4 kilojoules per day*
*** *RDA For energy 12 133.8 kilojoules per day*
**** *RDA for energy 9205 kilojoules per day*

The Marama bean *(Tylosema esculentum)* can easily be compared to the Bambara groundnut *(Vigna subterranea*) as presented in Table 4.12. The bambara groundnut originated in West Africa and is still cultivated all-over Africa (BAMLINK, 2004). Both the marama bean and the bambara groundnut are under-utilized crops, suitable for hot, dry, marginal soils. Both leguminous crops contain sufficient quantities of protein, fat

and other nutrients (Table 4.11) for human consumption, but is low in the amino acid methionine.

Table 4.12

Nutrient content of the marama bean and bambara groundnut

Nutrient	marama bean	bambara groundnut
Dry matter (%)	96.22	91.32
Ash (%)	3.36	3.26
Fat (%)	34.05	5.78
Prot (%)	36.44	19.94
C18:0 (Stearic acid)*	7.86	4.3
C18:1 (Oleic acid)*	42.03	26.5
C18:2 (Linoleic acid)*	32.16	37.0

***Percentage of total fatty acid content*

4.5 CONCLUSION

The chemical analyses of the Namibia and the Botswana roasted marama beans did not differ. The beans have a protein content of roughly 36% and like other legumes, are rich in the amino acid lysine, but low in the amino acid methionine. The fat content is between 43% and 40% and approaches that of a peanut. The oil is high in the long chain unsaturated fatty acids, oleic acid (C18:1), with 42% of total fatty acids, as well as high in linoleic acid (C18:2) with 32% of the total fatty acids. The marama bean contains no cholesterol. The marama bean can be regarded as an extremely valuable supplemental food source.

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5

Sensory analyses of the marama bean

5.1 INTRODUCTION TO CHAPTER 5

From previous studies (Ramolemana, 2003) it is known that the production of the marama seeds (beans) is *ca*. 0.1 to 0.3 t/ha. Questionnaires were developed and used in a survey amongst target groups within Namibia and Botswana to determine to what extent marama beans are used as food. Mature beans are frequently consumed as a good quality snack or as the main food, mostly roasted.

To determine the sensory attributes of the marama beans, descriptive analyses were performed. Descriptive sensory evaluation addresses the complexity of food systems by taking into account as many of the sensory attributes or notes of the food as possible (Brovelli *et al.*, 1999). The aroma, flavour, after-taste and mouth-feel are the most important sensory attributes normally influencing consumer's purchase behavior (Meilgaard *et al.*, 1991). Results obtained during sensory evaluation can provide valid and reliable information to product manufacturers and entrepreneurs with regard to the product's sensory properties and eventually insight into the positioning the products in relation to other similar products.

The objectives of this experiment were to compare and describe the sensory attributes of the marama bean samples obtained from two locations namely Namibia and Botswana and either traditionally or conventionally (oven) roasted, using sensory analyses (quantitative descriptive analysis). No sensory data on the roasted marama beans is available in other literature.

5.2 SENSORY ANALYSES

Meilgaard *et al.* (1990) stated that the primary function of sensory testing is to conduct valid and reliable tests, providing data on which sound decisions by the customer and consumer can be based. Sensory analyses of foods rely on evaluation through the use of senses (odour, taste, tactile, temperature, pain etc.) According to Jellinek (1985), sensory analyses imply the measuring of differences or classify the quality of a product by using a well-defined scale.

Meilgaard *et al.* (1990) said that various test techniques can be used to test responses of panelists. Of utmost importance is to define exactly what should be measured. According to Meilgaard (1990), through sensory analyses the most cost-effective and efficient method should be found to obtain most of the information. Descriptive tests are used in obtaining detailed descriptions of the texture, flavour and aroma of foods. Descriptive analyses methods involve the detection and description of both the qualitative and quantitative sensory attributes of a product by between five and 100 panelists who were beforehand specifically trained for this purpose. These panelists

must be able to detect and describe the perceived qualitative sensory attributes of a sample (Meilgaard *et al.*, 1990).

Sensory analyses have many applications, i.e. quality control of products, analyses of competitive products, new product development, market research and tests, hedonic tests and factors influencing the odour and flavour, texture, colour of food (Jellinek, 1985).

Jellinek (1985) stated that a test member must have normal olfactory and gustatory sensitivity, which can be improved by training. People of all ages can be test members. Younger persons often have more taste buds, where older persons can concentrate better. Smokers and non-smokers can be test members. Persons who are ill should not be allowed to participate in the testing and the use of smelling cosmetics should be avoided. Disturbances should also be avoided since sensory test require intense concentration.

5.3 MATERIALS AND METHODS

5.3.1 Sampling sites

Locations within Namibia and Botswana, where the marama bean plant grows in natural abundance, were chosen as sampling sites for the study of the quality characteristics of the marama bean plant (Fig. 1.1), namely:

- The Sandveld Research Farm area, east of Epukiro, 60 km northeast of Gobabis, in eastern Namibia (22 01' 04'' South, 19 08' 13" East).
- The Ghanzi / D' kar area in western Botswana (21 25' 03" South, 21 23' 28" East / 21 18' 45'' South, 21 33' 41" East).

5.3.2 Sampling

For the sensory analyses sampling was done during the rainy season (December, January to March) of 2001/2002. Matured beans were collected from within a radius of 100 km of each site. Collections were made ± two to five kilometers apart. Samples were sealed in paper bags and numbered according to location and date. No young (green) beans were collected, as it was stated by Rachie (1979) that beans were seldom or never eaten young (green).

5.3.3 Roasting

Half of the five kg of matured beans collected from the sites in Namibia and Botswana were traditionally roasted by the indigenous people on both the Namibian and Botswana sites visited, as to be able to determine the sensory attributes from the traditional roasting methods. Beans were roasted for approximately 10 - 12 minutes in cast-iron pots, whilst covered with sandy soil. Beans are only embedded in the pot once the soil has reached a temperature where it burns the fingers when touching the soil.

The rest of the beans were oven-roasted in the laboratory. Different temperatures and times were evaluated to obtain the best results. The samples were roasted in sand baths filled with ordinary sand that was pre-heated to a temperature of 160° C at an oven temperature of 180 °C. The beans were roasted in the pre-heated sand for 10 minutes, left overnight to cool and then shelled to remove the kernels. The roasting of the beans was done in ten Mielé ovens (model H217) and the temperature of the sand was measured with a hand-model Kane-Mane 1012 type temperature probe (Campbell *et al*. 1980), as part of the standard procedure.

5.3.4 Experimental design

Approximately 100 traditionally and oven-roasted beans from the two locations in Namibia and Botswana, were shelled. The beans were chopped roughly into 10 mm^2 pieces. The samples were coded as follows: sample 1 = Namibia traditionally roasted (NTR), sample 2 = Botswana traditionally roasted (BTR), sample 3 = Botswana conventionally (oven) roasted (BOR) and sample 4 = Namibia conventionally (oven) roasted (NOR). The marama beans were served according to research guidelines for the sensory evaluation measurements of food products. Care was taken to ensure uniformity of each sample (e.g. amount served etc.) of each replication of the different samples. All samples were randomized to exclude any bias due to the position effect. Each panelist received 30 ml of chopped sample, served in a 50 ml glass beaker, and covered with a three-digit random coded aluminum foil square on a white plastic tray. Fresh carrot rings and water at room temperature were served as palate cleansers in between evaluation sessions.

5.3.5 Training of panelists

Ten experienced panelists of the Sensory Analysis and Human Nutrition Unit of the Agricultural Research Centre (ARC), Irene, Pretoria, South Africa, were selected to participate in the study. During the training sessions, panelists were exposed to different food products related to the food product to be evaluated, for the development of relevant terminology. Panelists were further trained to increase sensitivity and ability to discriminate between specific samples and sensory attributes. Panelists were exposed to different types of leguminous food products, for example raw peanuts, salted peanuts, cocoa, dark chocolate, fresh oil, rancid oil, coffee beans, coffee powder and burnt wood to develop common terminology. Panelists were instructed to give a detailed description of the sensory attributes they perceived.

The training process was guided by a panel leader to ensure that all the attributes generated were relevant to the products to be tested. The identified attributes were grouped as aroma, flavour, after-taste and mouth-feel with a clear definition of each attribute. A nominal rating scale, with one (1) denoting the lowest rating (e.g. no burnt flavour/aroma) and eight (8) denoting the highest rating (e.g. extremely intense burnt flavour/aroma) was constructed and used to evaluate the different samples. Roasted marama bean samples were compared with raw (young, green) marama beans to identify and describe clear definitions for the sensory attributes (Table 5.1).

Table 5.1

List of definitions for sensory attributes of the marama beans

Sensory attributes	Definition of attributes
Aroma	
Roasted Peanut	Aromatic associated with freshly roasted peanuts
Burnt	Aroma associated with over roasted, burnt toast or toasted cereals
Chemical	Aroma characteristic of a closed plastic container, solvents
Peanut Butter	Aromatic associated with peanut butter
Caramel	Sweet aroma, characteristic of heated sugars and some other carbohydrates
Oily	Aromatic associated with oil present in beans
Nutty	Aromatic associated with roasted hazelnuts
Flavour	
Astringent	Puckering/dry taste (or feeling) in the mouth
Burnt (over roasted)	Flavour associated with over roasted, burnt toast or toasted cereals
Bitter	Taste tongue associated with bitter agents, such as caffeine
Chemical	Flavour characteristic of a closed plastic container, solvents
Nutty	Flavour associated with roasted hazelnuts
Woody	Flavour associated with old saw chips, toothpicks, not spicy
Cardboard	A flat aromatic and flavour associated with wet cardboard, dust, a vacuum cleaner bag
Cocoa	Flavour associated with dry cocoa powder
Earthy	Flavour characteristic of damp potting soil, wet compost
Mealy (floury/grainy)	Flavour associated with dry flour
Beany (green)	Flavour present in raw peanuts; nuts that have under-roasted with a characteristic green, beany raw note, freshly broken green beans
Peanuts	Flavour associated with fresh peanuts
Sweet	Flavour associated with materials that have a sweet taste (pleasant sensation)
Caramel	Sweet flavour, characteristic of heated sugars and some other carbohydrates
Coffee	The aroma note associated with freshly percolated coffee
Oily	Flavour associated with oil present in beans
After-taste	
Bitter	After taste on the tongue stimulated by quinine, caffeine and certain other alkaloids, burnt
Burnt	A burnt, harsh after taste, associated with charred sugar –bitter
Coffee	The after taste associated with freshly percolated coffee
Burnt Caramel	A burnt, harsh after taste, associated with charred sugar
Butter (round)	The after taste associated with any of the butter, oily, wax notes
Mouth-feel	
Astringent	Puckering/dry after taste (or feeling) in the mouth

After the four-day period of training, a preliminary study was conducted to familiarize the panel with the product, terminology developed, evaluation form, scoring procedure and to establish the most suitable palate cleanser and amount of sample for evaluation.

Quantitative descriptive analysis was used in order to determine whether differences exist between the four marama bean samples and to determine the direction of the differences. A complete randomized block design was used for evaluation with four replications that were considered the absolute minimum to ensure reliability and validity of results.

5.3.6 Sensory methods and conditions of the test

All samples were evaluated according to the methods described in the Annual Book of ASTM Standards (ASTM, 1989) in the adequate ARC sensory analysis facilities constructed according to ASTM design guidelines for sensory facilities.

The evaluation was conducted over a four-day period consisting of four evaluation sessions with one sample per session per day with 30 minutes between sessions. Panelists evaluated products in separate tasting booths to reduce distraction and panelist interaction and to insure uninterrupted, unbiased, individual responses. Red light was used to mask all possible colour differences.

5.4 STATISTICAL TECHNIQUES

The quantitative descriptive data obtained from the sensory panel were entered on a spreadsheet using Microsoft Excel (2000). Data were statistically analysed by the ARC, Biometry Unit using GenStat 2000 (Dept. Statistics, Genstat Committee, Rothamsted) computer program. The significance of all the sensory attributes measured for each bean sample was tested by means of analysis of variance (ANOVA). The different samples were used as the main effects at a significance level of 95 % ($p \leq 0.05$). If the main effect was significant, Fisher LSD-test was applied to determine the direction of the differences between mean values. Multivariate analysis techniques, PCA (Principle Component Analysis) and CVA (Canonical Variant Analysis) were performed to reduce a large set of variants into a smaller set, which explain most of the variations in the entire data set.

5.5 RESULTS AND DISCUSSION

The aroma, flavour, after-taste and mouth-feel attributes for the four marama bean samples are presented in Table 5.2. The mean values for all samples as well as the significant differences ($p \leq 0.05$) are indicated. If the main effect was found to be significant ($p \leq 0.05$) the direction of the differences in the mean values in the same row are described by a, b and/or c (uppercase). A correlation matrix was constructed to show the correlation (positive or negative) between the sensory attributes measured.

Table 5.2
Least square mean values for the sensory analyses of four marama bean samples

Sensory attribute	p-value	l.s.d.	NTR	BTR	BOR	NOR
Aroma						
Roasted Peanut	0.807	0.3519	4.150	4.225	4.275	4.117
Burnt	0.001	0.2815	4.300^{a}	2.825^{b}	2.875^{b}	2.700^{b}
Chemical	0.001	0.3018	3.875^{a}	3.000^{b}	3.100^{b}	2.950^{b}
Peanut Butter	0.001	0.3843	3.200^{b}	4.000^{a}	3.883^{a}	3.950^{a}
Caramel	0.017	0.1734	1.800^{b}	2.025^{a}	2.050^{a}	1.900^{ab}
Oily	0.170	0.2482	2.967	3.250	3.108	3.100
Nutty	0.043	0.2391	3.400^{b}	3.725^{a}	3.675^{a}	3.600^{ab}
Flavour						
Astringent	0.001	0.2809	4.450^{a}	3.675^{b}	3.850^{b}	3.825^{b}
Burnt (over roasted)	0.001	0.2997	5.125^{a}	3.150^{c}	3.500^{b}	3.375^{bc}
Bitter	0.001	0.2893	4.875^{a}	3.750^{b}	3.900^{b}	3.750^{b}
Chemical	0.001	0.2588	3.900^{a}	2.975^{b}	2.950^{b}	2.975^{b}
Nutty	0.021	0.2532	3.425^{b}	3.775^{a}	3.700^{a}	3.775^{a}
Woody	0.011	0.3058	3.075^{a}	2.625^{b}	2.700^{b}	2.625^{b}
Cardboard	0.215	0.2424	2.358	2.150	2.125	2.250
Cocoa	0.001	0.2041	2.750^{a}	2.150^{b}	2.200^{b}	2.100^{b}
Earthy	0.014	0.2599	2.750^{a}	2.625^{a}	2.325^{b}	2.550^{ab}
Mealy (floury/grainy)	0.083	0.2467	3.125	3.450	3.275	3.275
Beanie (green)	0.006	0.2650	2.825^{b}	3.175^{a}^{b}	2.950^{b}	3.250^{a}
Peanuts	0.239	0.2666	3.450	3.725	3.600	3.625
Sweet	0.001	0.1642	1.675^{c}	2.050^{a}	1.850^{b}	1.825^{bc}
Caramel	0.879	0.1904	1.800	1.850	1.875	1.825
Coffee	0.001	0.2450	3.575^{a}	2.725^{b}	2.925^{b}	2.750^{b}
Oily	0.939	0.2035	2.975	2.975	2.933	2.925
After-taste						
Bitter	0.001	0.2956	4.150^{a}	3.250^{b}	3.250^{b}	3.350^{b}
Burnt	0.001	0.2468	4.100^{a}	2.725^{b}	2.750^{b}	2.775^{b}
Coffee	0.001	0.2408	3.075^{a}	2.425^{b}	2.475^{b}	2.375^{b}
Burnt caramel	0.004	0.1666	2.050^{a}	1.825^{b}	1.825^{b}	1.750^{b}
Burnt (round)	0.009	0.2382	2.400^{b}	2.800^{a}	2.700^{a}	2.675^{a}
Mouth-feel						
Astringent	0.001	0.2658	3.950^{a}	3.250^{c}	3.550^{b}	3.425^{b}

Means with different letters (a, b, c or d) are significantly ($p \leq 0,05$) different within rows

5.5.1 Aroma

Seven aroma attributes were identified and evaluated by means of the trained panel.

- **Roasted peanut**. The four marama bean samples showed no significant differences ($p \leq 0.807$) for the roasted peanut attribute.

- **Burnt.** A significant difference ($p \leq 0.001$) was found between the four marama bean samples for the burnt aroma. NTR was found to have a significantly more intense burnt aroma (slightly to moderately intense) and differed significantly from BTR, BOR and NOR. The three samples were rated as having a slight burnt aroma and did not differ significant from each other.

- **Chemical.** A significant difference ($p \leq 0.001$) was found between the four marama bean samples with regard to chemical aroma. NTR had a significantly more intense chemical aroma (traces to slightly bland) compared to the other three samples. NTR, BOR and NOR had a significantly lower chemical aroma and did not differ significantly from each other.

- **Peanut Butter.** The four marama bean samples showed a significant difference ($p \leq 0.001$) with regard to the peanut butter aroma attribute. NTR rated as containing a significantly more intense peanut butter aroma compared to all the other samples. All the remaining samples were rated as having a slight peanut butter aroma.

- **Caramel.** For caramel aroma, significant differences ($p \leq 0.05$) were found between the samples. BTR and BOR were rated as having a significantly higher caramel aroma rating (slightly) compared to NTR that was rated as having a lower (practically none) caramel aroma. NOR was positioned between these two groups of samples with regard to this specific attribute.

- **Oily.** No significant differences ($p \leq 0.170$) were found between the four marama beans samples for the oily aroma. All samples were rated as having a slightly oily aroma.

- **Nutty.** Significant differences ($p \leq 0.05$) were found between the samples for the nutty aroma of the marama beans. BTR and BOR was rated as having a significantly higher nutty aroma rating (slightly) compared to NTR that was rated as having the least nutty aroma. NOR was positioned between these two groups of samples with regard to the nutty aroma (slightly nutty to practically none).

5.5.2 Flavour

- **Astringent.** Significant differences ($p \leq 0.001$) were found between the four marama bean samples for the astringent flavour. NTR was rated as having a significantly more intense (moderatly intense) astringent flavour compared to BTR, BOR and NOR.

- **Burnt.** Significant differences ($p \leq 0.001$) were found between the samples for the burnt flavour attribute. NTR was rated as having a significantly more burnt flavour (moderatly to high) compared to BOR that were rated as having a slightly to moderately burnt flavour, which in turn had a significantly more burnt flavour (slight) than BTR. NOR were positioned between NTR and BOR with regard to this specific attribute.

- **Bitter.** A significant difference ($p \leq 0.001$) was found between the four marama bean samples with regard to the bitter flavour. Sample 1 (Namibian traditionally roasted) was rated as having the most intense bitter flavour compared to all the other samples. BTR, BOR and NOR had a signficanlty lower (slight) bitter aroma and did not differ significantly from each other.

- **Chemical.** Significant differences ($p \leq 0.001$) were found between the four marama bean samples in the chemical flavour attribute. NTR was rated as having a significantly higher chemical flavour compared to the other four samples, which were rated as having a lower chemical flavour (traces to slightly chemical).

- **Nutty.** A significant difference ($p \leq 0.021$) was found between the four marama bean samples with regard to the nutty flavour. BTR, BOR and NOR had a significantly higher nutty flavour compared to NTR that was rated as having the least nutty flavour of all the other samples.

- **Woody.** A significant difference ($p \leq 0.011$) was found between the four marama bean samples with regard to the woody flavour. BTR, BOR and NOR had a significantly lower woody flavour compared to NTR that was rated as having a moderately high woody flavour.

- **Cardboard.** No significant differences ($p = 0.215$) were found between the four marama bean samples for the cardboard flavour.

- **Cocoa.** A significant difference ($p \leq 0.001$) was found between the four marama bean samples with regard to the cocoa flavour. NTR was rated as having the most intense cocoa flavour compared to all the other samples. BTR, BOR and NOR had a signficanlty lower cocoa flavour and did not differ significant from each other.

- **Earthy.** A significant difference ($p \leq 0.05$) was found between the samples for the earthy flavour. NTR and BTR were rated as having a significantly higher earthy flavour (slight) compared to BOR that was rated as having a lower (slightly nutty to practically none) earthy flavour. NOR was positioned between these two groups of samples with regard to an earthy flavour.

- **Mealy (floury/grainy).** No significant differences ($p = 0.083$) were found between the samples with regard to the mealy flavour. All four marama bean samples were rated as having a slight mealy flavour.

- **Beanie (green).** Significant differences ($p \leq 0.006$) were found between the four marama bean samples for a beanie flavour. NOR was rated as having a significantly higher beanie flavour (slightly) compared to NTR and BOR. BTR was positioned in between with regard to beanie flavour.

- **Peanut.** No significant differences ($p = 0.239$) were found between the samples for the peanut flavour. All samples were rated as having a slight to moderate peanut flavour

- **Sweet.** Significant differences ($p \leq 0.001$) were found between the samples for the sweet flavour. BTR was rated as having a significantly higher sweet flavour (slight), compared to BOR that was rated as having traces to a slightly sweet flavour which in turn was more sweet than NTR. NOR was positioned between NTR and BOR with regard to this specific attribute. NTR was rated as the sample with the least sweet flavour of the four samples.

- **Caramel.** No significant differences ($p = 0.879$) were found between the samples for the caramel flavour attribute. All samples were rated as having practically no to traces of a caramel flavour, with NTR rated as having only traces of the caramel flavour, although not significantly so.

- **Coffee.** Significant differences ($p \leq 0.001$) were found between the samples for a characteristics coffee flavour. NTR was rated as having a significantly higher coffee flavour (slight) compared to BTR, BOR and NOR.

- **Oily.** No significant differences ($p = 0.939$) were found between the four marama bean samples for the oily flavour. All samples were rated as having only traces of an oily flavour.

5.5.3 After-taste

All the after taste attributes evaluated, differed significantly between the four marama bean samples.

- **Bitter after-taste.** A significant difference ($p \leq 0.001$) was found between the four marama bean samples for the bitter after-taste. NTR was rated as having a significantly more intense bitter after-taste (moderately) compared to the other samples. BTR, BOR and NOR had significantly less intense bitter after-taste.

- **Burnt after-taste.** Significant differences ($p \leq 0.001$) were found between the four marama bean samples with regard to the burnt after-taste. According to the mean values, NTR was rated as having the most intense burnt after-taste (moderately) compared to BTR, BOR and NOR that were rated significantly lower in burnt after-taste compared to the other samples.

- **Coffee after-taste.** A significant differences ($p \leq 0.001$) were found with regard to the coffee after-taste for the four marama bean samples. NTR had a significantly more intense coffee after-taste compared to BTR, BOR and NOR with a significantly less intense coffee after-taste.

- **Burnt caramel after-taste.** BTR, BOR and NOR were rated as having a significantly ($p \leq 0.004$) less intense burnt caramel after-taste (practically no to traces) compared to NTR that was rated as having a significantly more intense burnt after-taste (slight).

- **Burnt after-taste (round).** Significant differences ($p \leq 0.009$) were found between the four marama bean samples with regard to the burnt (round) after-taste. BTR, BOR and NOR were rated as having a significantly more intense burnt after-taste compared to NTR that was rated to have a less intents burnt (round) after-taste.

5.5.4 Mouth-feel

- **Astringent.** Significant differences ($p \leq 0.001$) were found between the four marama bean samples with regard to the astringent mouth-feel. According to the mean values, NTR was rated as having the most intense astringent mouth-feel (moderately) compared to BOR and NOR which in turn differed significantly from BTR that was rated as the sample with the least astringent mouth-feel compared to the other samples.

5.6 CORRELATIONS

A correlation matrix of all the sensory attributes evaluated for the four marama bean samples were constructed. A fairly strong positive correlation ($r = 0.76$) exists between the chemical aroma and the burnt aroma as evaluated by the sensory panel (Table 5.2).

A positive correlation (r = 0.70) was found between chemical aroma and chemical flavour. This is an indication that the panel could identify the chemical compound in a systematic manner, irrespective if it was an aroma or flavour component. A positive correlation (r = 0.68) was also found between the chemical flavour and the burnt aroma. A positive correlation (r = 0.70) was also found between chemical flavour and burnt flavour. Further more a fairly strong positive correlation (r = 0.75) between the coffee flavour and coffee after taste was found.

5.7 MULTIVARIATE STATISTICAL ANALYSES

5.7.1 Discrimination of the four marama bean samples on aroma, flavour, after taste and mouth feeling attributes using Canonical Variant Analysis (CVA).

A graphical presentation of the grouping of the four marama bean samples is shown in Figure 1. The first canonical variant (CV 1) accounted for 91.60 % of the total variation in the data with a latent root of 1.0581, and the second variant (CV 2), accounted for 6.50 % of the total variation with a latent root of 0.0751. For a canonical variant to have a significant contribution to the variation in the data, a latent root > 1.00 must be indicated. Therefore, according to the results CV 1 accounted for a significant variation in the data. The burnt aroma, flavour and after-taste showed the most important variates according to the CVA (Figure 5.1).

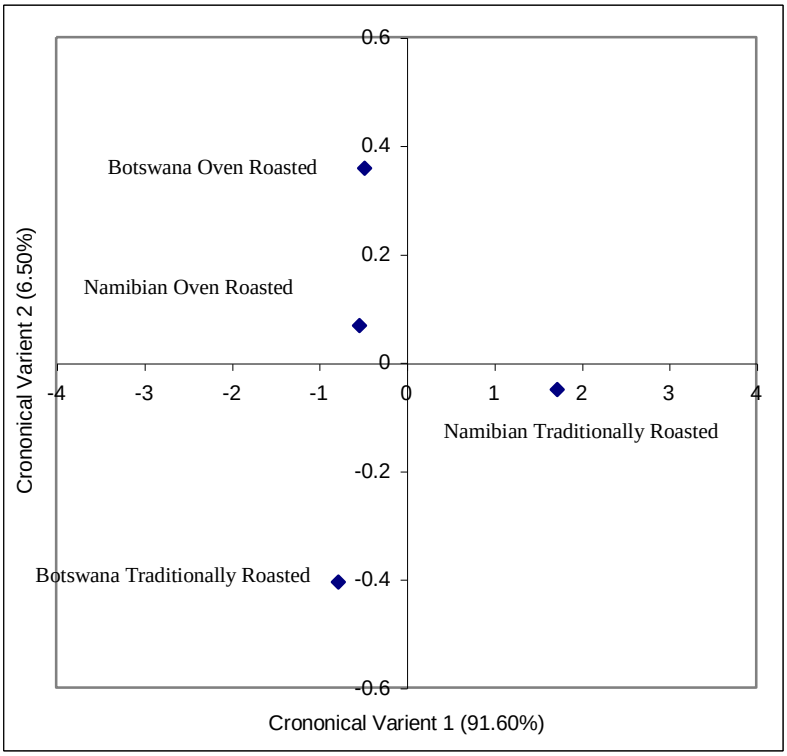

Figure 5.1

Plot of Canonical Variant Analysis mean scores of four different roasted marama bean samples

5.7.2 Discrimination of the four marama bean samples on aroma, flavour, texture, after taste and after-feeling attributes using Principle Component Analysis (PCA)

The main variate discriminating between the four marama bean samples CV1 (horizontal axis) was the burnt flavour (r = 0.912) and burnt after taste (r = 0.840). The burnt flavour is therefore the largest discriminant between the four marama bean samples. Further insight was obtained by plotting the CV-scores in relation to the axis in CV1 (Figure 1). According to the graphical presentation sample NTR contrasted the strongest with BTR, BOR and NOR. Sample NTR had the strongest, most intense burnt flavour and after taste compared to BTR, BOR and NOR indicating as having a slight to moderate burnt flavour, as also found by the ANOVA-analysis.

A graphical presentation of the interrelationships of the sensory attributes of the four samples is shown in Figure 5.2. The principal components in PC 1 and PC 2 accounted for 65.23 % of the total variation in the data. The first principle component (PC 1) accounted for 44.95 % of the total variation in the data with a latent root of 5.844 and the second PC 2 accounted for 20.28 % of the total variation with a latent root of 2.636. The graphical presentation of this specific PCA also gives an indication of the correlation of the sensory attributes measuring the same attribute, for example the correlation of the burnt flavour and after-taste.

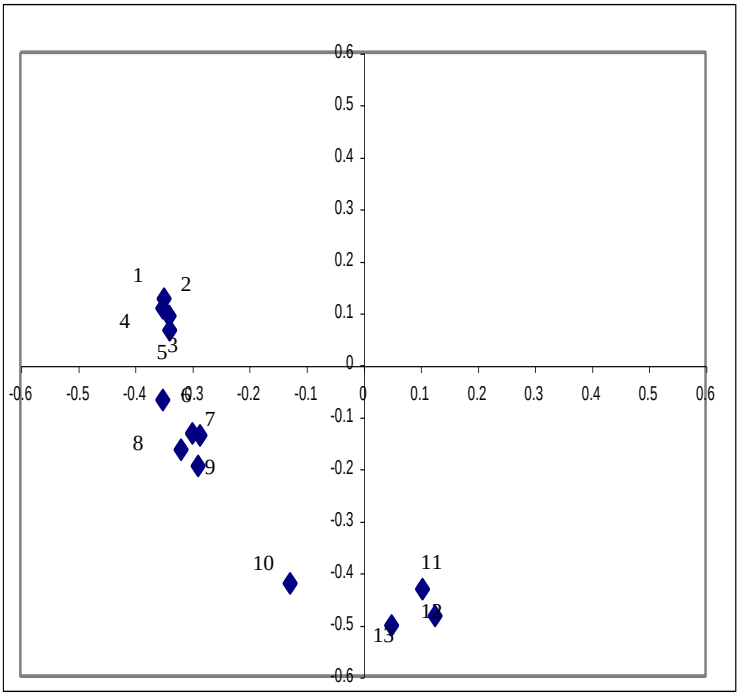

Figure 5.2

Plot of PCA mean scores of four marama bean samples

Aroma burnt (1), flavour chemical (2), flavour burnt (3), aroma chemical (4), after taste burnt (5), flavour astringent (6) flavour bitter(7), after taste bitter (8), mouth feel astringent (9), flacour cocoa (10), aroma peanut (11), aroma oily (12), flavour oily (13).

The main variates discriminating between the four samples in PC 1 (horizontal axis) was burnt aroma (r = -0.841), chemical flavour (r = 0.848), burnt after taste (r = 0.851) and burnt flavour (r = -0.822). For PC 2 (vertical axis), oily aroma (r = -0.786), and oily flavour (r = -0.627) were found to be the main variates discrimination between the samples. According to the graphical presentation, NTR contrasted the strongest with BTR, BOR and NOR for PC 1. NTR had a higher (more intense) burnt aroma flavour and after taste, as well as chemical flavour compared to BTR, BOR and NOR. Although a tendency is noticed, for oily aroma (r = -0.786) and oily flavour (r = -0.817),

no prevalent differences were indicated for PC2 between the four marama bean samples as confirmed by the ANOVA (Figure 5.2).

5.8 CONCLUSIONS AND RECOMMENDATIONS

Of the seven aroma attributes studied in the marama bean samples, five differed significantly according to the ANOVA, namely burnt, chemical, peanut butter, caramel and nutty aroma. Eleven of the 16 flavour attributes studied showed significant differences between the four samples. The astringent, burnt (oven roasted), bitter, chemical, nutty, woody cocoa, earthy, beany, sweet and coffee flavour attributes discriminated the most between the flavour attributes of the four marama bean samples. All five of the evaluated after-taste attributes (bitter, burnt, coffee, burnt (caramel) and burnt (round)) significantly differed between the four marama bean samples. With regard to mouth-feel, a significant difference was found in the astringent sensation.

According to the sensory profiles, the Namibian traditionally roasted marama bean had a significantly more intense burnt, bitter and chemical aroma and flavour as well as after taste, and a slightly lower peanut butter caramel and nutty aroma, a more intense astringent, burnt (oven roasted), bitter, chemical, woody, cocoa and coffee flavour and as well as a more intense coffee burnt caramel after-taste and a lower burnt after-taste compared to the other three marama bean samples. A more intense astringent flavour and mouth-feel were also detected when compared to the other samples.

The sensory attributes of the Botswana traditionally roasted, Botswana oven roasted and Namibian oven roasted samples, grouped together. They had a significantly lower burnt and chemical aroma and a slightly higher peanut butter aroma. When compared to the Namibian traditionally roasted sample, the Botswana traditionally roasted, Botswana oven roasted and Namibian oven roasted marama bean samples had a significant less intense astringent, bitter, chemical, woody, cocoa and coffee flavour and a slightly higher nutty flavour, a significantly less intense bitter, burnt and coffee after taste as well as lower astringent mouth feel.

According to the results of the CVA and the PCA, burnt aroma, flavour and after-taste were the attributes that showed to be the most important variates discriminating between the four marama bean samples and should therefore be regarded as very important to control in the preparation of this product. The aroma and flavour attributes are important; should it be the objective to market the marama bean as food. It is, therefore recommended that a consumer preference-test on the burnt attributes, (burnt aroma and flavour as well as after taste) be conducted to determine the role of the burnt, bitter flavour in the acceptability of the marama bean as food. The more “dark” aroma and flavour of the Namibian traditionally roasted marama bean should be investigated with regard to consumer acceptability due to the nature of the product as well as the tradition surrounding the preparation thereof.

It is recommended to investigate the techniques of the traditional roasting of the marama beans, due to the obvious influence that it has on the aroma, flavour and after-taste of the product and which might have a negative impact on the acceptance of the

product. The marama bean samples from the different regions (Namibia and Botswana) should be roasted under identical, controlled conditions and be evaluated by the target groups to define whether the target groups prefer the traditionally roasted or oven roasted method.

Future research should incorporate the effects of the sensory attributes on the dietary habits of the children within the target groups (Lawless, 1985) in Namibia and Botswana, since the children are the under-nourished group in these communities.

5.9 REFERENCES TO CHAPTER 5

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6

The value of the marama plant as a fodder for cattle

6.1 INTRODUCTION TO CHAPTER 6

It has been proved by several studies that the marama beans have extensive nutritional potential for humans (Biesiele and Murray, 1983), but little has been done to investigate the value of the marama plant as fodder for livestock.

The marama plant is a rich source of protein and energy in regions where few conventional crops can survive. It grows in some areas that receive up to 800 mm in rainfall and in others where rainfall is so slight and erratic that in some years almost no rain falls at all (ECHO, 1999).

It is generally accepted that cattle depend predominantly on grasses, although utilizing herbaceous dicots and woody plants at certain times and under certain conditions (Forbes, 1995). For example, countless experiments in the USA have indicated that season-long cattle diets average 75% grasses, 15% forbs and 10% shrubs or browse (Merritt *et al.*, 2001).

The demand for energy and protein sources for both animal and human consumption is on the increase and it is likely to continue to do so (McDonald, 1988). Protein is likely to become increasingly scarce and costly. It is therefore a necessity that the nutritional potential of all plants that can possibly be used as food or feed is exploited, especially those under-utilized crops and indigenous plants that are adapted to the soil and climate of the region.

According to the National Academy of Sciences (1979), cattle in Africa “eagerly eat” the leaves and stems of the marama bean plants, although Watt and Breyer-Brandwijk (1962) reported that “the foliage of this species is apparently not browsed by stock”. The only reported use as an animal supplement is by some native farmers who use them to fatten pigs (Starcher, 1985).

One animal study has been reported testing the nutritional value of the marama bean (Ripperger–Suhler, 1983). Young growing rats were fed raw or cooked bean meal, contributing 10% protein to a purified diet. Food consumption was very poor, resulting in an overall weight loss over the four week test period. Mortality was not significant and pancreatic hypertrophy was minimal. Additional studies were conducted searching for anti-nutritional factors that might cause the rats to refuse diets containing marama beans. No hemaglutinins were found using blood samples from rats, rabbits, sheep and humans. On the other hand, trypsin inhibitors were found in extremely high levels, 239 TUI/mg protein which is close to twice that reported for soybeans (Kakade *et al.*, 1973). Whether these high levels of trypsin inhibitor contributed to the rats refusal to

consume the diet, could not be determined. However, diets containing roasted beans, which no longer contained anti-trypsin activity, were also not consumed.

Powell (1983) has initiated studies to test the use of the marama plant as forage under rangeland conditions. The plants are known to tolerate drought, but the main problem is to get them established initially under range conditions characterized by unpredictable rainfall and periodic droughts (Powell, 1987).

6.2 MATERIALS AND METHODS

6.2.1 Experimental field design

The botanical and dietary abundance of the marama plant was determined in the course of a farm-scale, long-term systems trial started at the Sandveld Research Farm in eastern Namibia in 1987, in the central Kalahari, camel thorn tree savanna. The study investigated the effect of four systematically increasing stocking rates of free-ranging beef cattle, as well as two cattle frame sizes, on cattle productivity and veld condition. Since 2001, this systems trial served to elucidate the dietary preferences of cattle during six seasons, viz. three hot-wet (March of 2001, 2002 and 2003), two cold-dry (June-July of 2001 and 2002) and one hot-dry season (October 2002) and data concerning the dietary and botanical abundance of the marama plant was obtained during this period.

At Sandveld, two types of cattle were evaluated, viz. the relatively large-framed Afrikaner x Simmental rotational crossbreed and the small-framed, purebred Sanga. These two widely divergent cattle frame sizes were chosen to elucidate the argument that “big is not always best” (Dickerson, 1978; Els, 1998). Stocking rate was kept relatively constant by fixing the number of cows in a treatment. It increased from “low” (targeted cow mass: 15 kg/ha, equivalent to 30 ha/Large Stock Unit (LSU)) to “low-medium” (25 kg cow mass/ha or 18 ha/LSU) to “medium-high” (35 kg cow mass/ha or 12.9 ha/LSU) and, ultimately, to “high” (45 kg cow mass/ha or 10 ha/LSU). Treatment herds consisted of 18 to 78 cows, depending on the targeted stocking rate, and resembled stocking rates in use by commercial ranchers, at least at the rates. Routine cattle management activities were identical across all eight treatments and included a set programme of preventive health measures, mating, weaning, replacement policy, supplementation and water provision, etc.

Each of these 2 x 4 factorial treatments was allocated six grazing camps of, in total, 689$\pm$4.4 ha. Cowherds were rotated through their allocated grazing area on a fixed cycle of 7-10 days occupation per camp during the hot-wet season and 10-14 days occupation per camp during the two dry seasons (cold-dry and hot-dry). However, the diet selection trial was restricted to only one of the six available camps per treatment, to prevent differences between camps influencing the experiment. The experimental plot (average size: 142$\pm$28.9 ha) was selected from the available six treatment camps to be as similar in soil type (deep red Kalahari sands of the Hutton soil type) and vegetation type (fairly open savanna dominated by perennial grasses and the camel thorn tree,

Acacia erioloba) as possible. Wild herbivores roamed the whole farm freely and no distinction could be made between their impact on plants and that of livestock.

Shortly before the treatment cowherd was put to graze the experimental plot, its botanical composition was determined by 474±72.1 step-points that were placed every 3 m along its diagonal transect. Botanical abundance of all plant species was calculated based on point strikes on their canopy. Multiple plants were recorded at one strike point when it struck one plant growing beneath the canopy of another. Herbaceous yield before grazing was determined by clipping, at ground level, 40 x 1 m^2 equidistant quadrats along the diagonal transect and sorting the yield, at clipping, into 10 different fractions, of which one contained all dicotyledonous plants, of which the marama plant was a part. The biomass produced by this plant was thus not measured separately, only in a group containing all the other herbs of the veld.

While the treatment cowherd was grazing the experimental plot, but still in the first half of their period of occupation, when utilized plants were still clearly recognizable, six cows from each of the eight factorial treatments were selected at random and observed for an uninterrupted period of 10 minutes/cow. All bites taken were counted and all forage plants utilized were identified, to calculate the dietary abundance of each forage plant. Plant parts or organs (including seeds, pods and tuber parts) utilized, were also recorded. This procedure was repeated on two early mornings and two late afternoons per factorial treatment. At these times, cattle, being crepuscular, are feeding most actively (Albright and Arave, 1997). Each time, six cows were randomly selected from

the treatment herd, enabling statistical analysis of the data by simple ANOVA, rather than by repeated measures ANOVA.

The ad- and disadvantages of the various methods available to determine the diet selection of free-ranging cattle have been extensively reviewed on Sandveld Research Farm (Rothauge, 2004). Suffice to accept the statement by Forbes (1995), an eminent expert in this field, that there is no ideal method and that the choice of method eventually depends on the operator and the circumstances of the experiment. At Sandveld Research Farm, the cattle were tame enough that the operator could approach them closely for reliable bite counting and forage identification and the operator had sufficient botanical knowledge to identify forage plants accurately. Given the difficulty of obtaining reliable information from fistulated animals in such a difficult environment, the choice of method was obvious. In addition, direct observation of cattle diet selection avoids the confounding effect that free-ranging wild herbivores, which occur at Sandveld Research Farm in considerable numbers, have on the utilization of forage plants.

To evaluate the effect of cattle frame size on diet selection of cattle, botanical composition of the veld and nutritive value of the diet, all 24 cows within a frame size treatment were pooled. The same was done to the 12 cows belonging to a fixed stocking rate treatment. To establish dietary preference, the dietary abundance of a forage species was compared to its botanical abundance. A ratio larger than 1.0 (dietary abundance : botanical abundance) indicates preference of the particular plant (Petrides, 1975).

6.2.2 Sampling

After termination of grazing the treatment plot, samples from every utilized forage plant species, including the marama plant, were collected. A total of 1017 forage samples were collected, of which 280 (27.5%) were collected in a random manner while 730 (71.7%) were collected in a manner imitating the diet selectivity observed and recorded in cattle while their bites were counted ("imitated" samples). A further 7 samples (0.6%) represented other matter, such as moribund herbaceous matter, lick etc. were also collected. Random samples were only collected from the six ecological indicator grass species and from the total herbaceous bouquet on offer by re-constituting, on a mass-basis, the 10 individual yield fractions (Pers. comm., Rothauge, 2003). The reconstituted random sample is thus a real entity and not an average of other samples. Imitated forage samples were collected from every plant species utilized by cattle in such a manner that the principal forage species were sampled more often than the less important ones. The average grass, woody and dicotyledonous plant sample is, in contrast to the re-constituted random sample, only an arithmetic average of grass, woody and dicotyledonous plant samples, respectively.

These samples were immediately after collection, sealed in plastic, to retain their natural or field moisture content, weighed, dried, ground and subjected to standard chemical analysis to determine their nutritional content, which was presumed to indicate the nutritional content of the selected diet.

6.2.3 Proximate analysis

Crude protein (CP), crude fibre (CF), crude fat (fat), ash, calcium (Ca) and phosphorus (P) content were determined as per AOAC (1990, 1995), Acid detergent fiber (ADF) was determined as per Van Soest et al (1970). Neutral detergent fiber was determined as per Robertson and Van Soest (1981). Acid detergent fiber (ADF) was determined as per Goering and Van Soest (1970). In vitro digestibility of the organic matter (DOM) and metabolizable energy (ME) content were determined as per Menke *et al.* (1979). (Analyses done by the Agriculture Laboratory, Ministry of Agriculture, Water and Rural Development, Windhoek, Namibia by ALASA methods)

6.2.3.1 Moisture / Dry matter

The sample is dried in an open weighing vessel for five hours at 105 °C in a convection oven. Weight loss is used to calculate moisture / dry matter content. (AOAC method 4.1.03 (943.1)).

6.2.3.2 Ash / Organic matter

Organic matter of the sample is removed by heating to a temperature of 550 °C. The remaining residue is the ash. (AOAC method 4.1.10 (942.015)).

6.2.3.3 Crude Fat (Soxlet method)

Ether is heated in a flask until it boils. Cool water condenses the ether vapours and it drops onto the sample placed into an extraction thimble. The fat is extracted by the ether which drops back into the flask. The ether is evaporated from the flask, leaving the fat behind (AOAC method 4.5.01 (920.39)).

6.2.3.4 Crude Protein

The Nitrogen content of the sample is determined by using the Kjeldahl method with Büchi 430 block digestor and a Büchi 322 distillation unit. Samples are digested with concentrated sulphuric acid in which salts are added to increase the boiling point. The nitrogen is converted to ammonia, which reacts with excess of the acid to form ammonium sulphate. This is then neutralized with sodium hydroxide and the ammonia is distilled and collected in an excess of boric acid and is quantitatively determined by titrating the borate formed with hydrochloric acid. N x 6,25 = protein (AOAC method 2.4.03 (955.04)).

6.2.3.5 Crude fiber

Crude fiber is determined gravitimetrically after chemical digestion and solubilisation of other compounds present (i.e. protein, starch and other digestible/solubilisable carbohydrates) with diluted sulphuric acid and sodium hydroxide. The fiber mass is then corrected for ash content after ignition (AOAC method 4.6.01 (962.09)).

6.2.3.6 Acid detergent fiber

The sample is incubated with pepsin under acidic conditions for 24 hours, then extracted with acid detergent solution and the remaining residue is dried and ashed (Goering and Van Soest, 1970).

6.2.3.7 Neutral detergent fiber

The method determines the insoluble portion of a plant consisting of hemicellulose, cellulose, lignin, cutin and silica, by its extraction with neutral detergent solution and alpha-amylase under specific conditions. During extraction, interfering metal ions are removed by complexing with EDTA, proteins are removed with sodium lauryl sulphate, while other nitrogenous matter and non-available proteins are removed with sodium sulphate. Alpha-amylase converts starch to soluble sugars. 2-ethoxyethanol eliminates non-fibrous residues and serves as anti-foam. After the extraction, the residue is dried and ashed (Robertson and Van Soest, 1981).

6.2.4 Minerals

6.2.4.1 Phosphorus

The phosphorus concentration in the solution of digested sample is determined spectrophotometrically as the yellow phospho-vanado-molybdate complex (Cavell, 1955).

6.2.4.2 Calcium

Calcium was determined by Atomic Absorption Flame Spectroscopy (Price, 1972).

6.2.5 In vitro digestibility of organic matter / metabolizable energy content

Digestibility of organic matter was determined with the Hohenheim Gas Test (Menke *et al.*, 1979). The relationship between digestibility in vivo and gas production (carbon dioxide and methane) in vitro, when the plant/feed is incubated with rumen liquor for 24 hours, is used for the estimation of digestibility of organic matter. Metabolizable energy content can be calculated.

6.3 STATISTICAL ANALYSES

Statistical analysis utilized the general linear model of the SPSS computer programme (Bryman and Cramer, 1997), with prior arcsin transformation of all relative abundance data. Relative abundance is typically skewed with only a few high abundances and many low abundances (Zar, 1999).

6.4 RESULTS AND DISCUSSION

6.4.1 Nutrient content of diet selection

Laboratory analysis yielded information on 11 different nutrients within the total diets selection.

Table 6.1

Nutrients of major importance of all random and imitated forage samples collected during the diet selection trial at Sandveld Research Farm[a]

	Random samples	Imitated samples	Statistical parameters
Number	280	730	
Field DM (%)	76.8±15.09	65.1±22.6	$P < 0.01$; $r^2 = 0.06$
CP (%)	4.5±1.63	7.7±3.67	$P < 0.01$; $r^2 = 0.16$
Ca (%)	0.37±0.24	0.70±0.78	$P < 0.01$; $r^2 = 0.05$
P (%)	0.03±0.02	0.05±0.03	$P < 0.01$; $r^2 = 0.09$
CF (%)	37.9±3.21	33.7±7.38	$P < 0.01$; $r^2 = 0.08$
ADF (%)	45.1±3.74	40.8±6.17	$P < 0.01$; $r^2 = 0.11$
NDF (%)	72.7±5.50	64.0±13.55	$P < 0.01$; $r^2 = 0.10$
Fat (%)	1.4±0.37	2.1±1.28	$P < 0.01$; $r^2 = 0.08$
Ash (%)	8.2±2.41	9.3±5.90	$P < 0.01$; $r^2 = 0.01$
DOM (%)	44.9±8.49	50.3±9.23	$P < 0.01$; $r^2 = 0.07$
ME (MJ/kg)	6.2±1.03	7.2±1.17	$P < 0.01$; $r^2 = 0.14$

[a] *Systems trial (Rothauge, 2004)*

The imitated samples consistently had a more advantageous nutrient content than the random samples, as indicated by their higher field moisture (50% difference), protein (71%), calcium (89%), phosphorus (67%), fat (50%), ash (13%) and metabolizable energy content (16%), their greater digestibility (12%) and lower fiber content (9 – 12% difference).

Table 6.2

Content of major nutrients of random and imitated (in *brackets*) samples of the principal forage species of free-ranging beef cattle at the Sandveld Research Farm*

Species	Number	CP (%)	NDF (%)	DOM (%)
Schmidtia pappophoroides	48 *(58)*	4.4±1.47 *(5.6±1.98)*	69.8±3.93 *(70.3±3.08)*	50.3±7.67 *(55.5±8.39)*
Anthephora pubescens	30 *(73)*	5.9±2.04 *(7.3±2.64)*	65.7±3.85 *65.6±3.09*	53.2±9.25 *55.7±8.40*
Eragrostis lehmanniana/ E. trichophora	0 *(76)*	*(6.1±1.82)*	*(73.5±2.57)*	*(48.1±7.32)*
Stipagrostis uniplumis	48 *(59)*	4.0±1.09 *(5.2±1.50)*	75.7±3.99 *(75.1±2.62)*	40.1±5.80 *(45.2±8.07)*
Melinis repens repens	0 *(54)*	*(6.0±2.03)*	*(70.2±3.90)*	*(50.1±7.09)*
Eragrostis rigidior	48 *(65)*	4.1±1.10 *(5.7±2.28$^{)}$*	75.2±4.11 *(73.9±2.66$^{)}$*	40.9±5.68 *(44.3±7.73$^{)}$*
Grewia flava/ G. flavescens	0 *(16)*	*(15.4±3.12)*	*(42.2±3.80)*	*(46.3±10.06)*
Tarchonanthus camphoratus	0 *(15)*	*(10.1±2.06)*	*(47.6±5.58)*	*(47.7±5.58)*
Acacia mellifera	0 *(11)*	*(10.7±1.35)*	*(30.2±4.36)*	*(42.3±12.52)*
Terminalia sericea	0 *(12)*	*(7.2±1.72)*	*(48.0±9.37)*	*(41.4±3.24)*
Nidorella resedifolia	0 *(29)*	*(10.2±2.59)*	*(40.9±11.49)*	*(50.1±6.64)*
Hermannia tomentosa	0 *(8)*	*(12.9±2.67)*	*(46.7±4.01)*	*(55.9±8.84)*

(Different superscripts indicate significant differences between means in a cell, i.e. within a species and nutrient. If superscripts are in capital letters, $P < 0.01$ and if superscripts are in small letters, $P < 0.05$.)
*Systems trial (Rothauge, 2004)

The nutritional properties of the random as well as the imitated samples differed between the different species of grass ($P < 0.01$; except for the field dry matter (DM) content of random samples, for which $P > 0.05$), as well as between different species of woody plants ($P < 0.01$) and different species of dicotyledonous herbs and forbs ($P <$

0.05; except for their field DM and acid detergent fiber (ADF) content, for which P > 0.05). As far as the grasses are concerned, the major nutritional difference between random and imitated samples was in their crude protein (CP) content and digestibility, with neutral detergent fiber (NDF) content being less sensitive to manner of sampling. The CP content of the principal grasses, as utilized by cattle (imitated), varied roughly from 5 to nearly 8%. The CP content of the principal browse forages and dicots was twice as high than that of grasses. Despite the much lower NDF content of the woody forages, their digestibility was still lower than that of the grasses, while the dicots had a lower NDF content but higher digestibility than grasses.

6.4.2 Botanical and dietary abundance of the marama plant

Over all treatments and seasons of the diet selection trial, dicotyledonous plants contributed 15.0$\pm$10.95 g dry matter/m^2 to the total herbaceous yield of 172.1$\pm$39.51 g dry matter/m^2, or 8.7$\pm$5.70%. The marama plant contributed noticeably to dicotyledonous yield, but its yield was not quantified separately. The marama plant comprised 5.6$\pm$2.43% of all plants in the treatment plots (Table 6.3) and varied significantly with season of the year ($p \leq 0.01$, $R^2 = 0.81$), but its botanical abundance was not influenced significantly by cattle frame size ($p = 0.80$, $R^2 = 0.28$) and stocking rate of cattle ($p = 0.44$, $R^2 = 0.28$). It made up only a very small part of the diet of cattle, comprising 0.5$\pm$1.08% of all bites taken (Table 6.3). The season of the year had a significant effect on when cattle selected it ($p \leq 0.01$, $R^2 = 0.55$), but dietary abundance was not influenced by cattle frame size ($p = 0.93$, $R^2 = 0.13$) and stocking rate of cattle ($p = 0.40$, $R^2 = 0.13$).

Table 6.3

Relative abundance (%) and standard deviation of the marama plant in the natural vegetation and the diet of cattle at Sandveld Research Farm

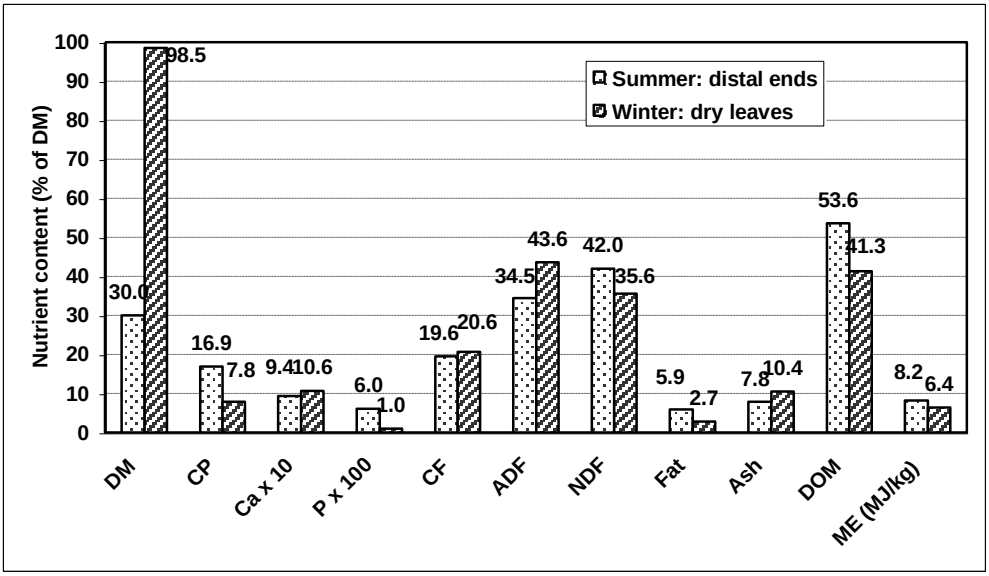
TREATMENT	Botanical abundance (%)	Dietary abundance (%)
Over all treatments	5.60±2.43	0.54±1.08
Large-framed cattle	5.57±2.699	0.45±0.69
Small-framed cattle	5.63±2.20	0.64±1.38
Low stocking rate	6.44±3.10	0.53±1.48
Low-medium stocking rate	5.57±2.19	0.23±0.60
Medium-high stocking rate	5.46±1.82	0.86±1.31
High stocking rate	4.93±2.50	0.56±0.72
Hot-wet season	6.83±1.79	0.98±1.38
Cold-dry season	5.14±2.56	0.15±0.36
Hot-dry season	2.83±0.80	0.00

The veld at Sandveld Research Farm was in good condition over all treatments, with grasses comprising at least 69% of all plants and more than 99% of all grasses were perennial. Of course, veld condition varied with treatment, but had not yet proceeded over the threshold towards bush-encroached veld dominated by annuals, or even bare veld, at the highest stocking rate treatment. The marama plant made up a sizeable proportion of the plants, more or less equal to the botanical abundance of a relatively common grass like *Eragrostis lehmanniana and E. trichophora*. Its abundance was not

influenced statistically by stocking rate treatment, but it did appear to become less abundant as stocking rate increased, decreasing by about 31%. This tendency cannot be explained by its abundance in the diet of cattle, because it comprised only 0.5% of cattle diet, varying without tendency between stocking rate treatments. Its preference rating was very low throughout all treatments and seasons, around 0.10, indicating that cattle did not seek it out for consumption. In fact, the low preference rating indicates the opposite, viz. that cattle refrained from eating it, failing to take a bite even if they came across the marama plant during their foraging forays. It did appear as though the small-framed Sanga cattle selected it 42% more readily than the large-framed Afrikaner x Simmental crossbreeds, but this tendency was not significant, mainly due to the tremendous variation in its selection between the eight factorial treatments. In fact, the standard deviation of the dietary abundance of the marama plant exceeded its average abundance in cattle diets in all treatments and seasons, indicating an extremely high variability in its contribution to cattle diet due, probably, to taste differences between individual cows or micro site effects on individual plants, rather than systematic treatment effect. It was noticed anecdotally, that porcupines utilized the plant well, often digging up its enormous underground tubers and leaving them half-eaten, exposed to the elements in a hole, from which treatment the plant seemed to be able to recover very quickly.

The marama plant was, however, significantly more abundant in the veld during the hot-wet, vegetative growing season (Table 6.3), when it was sprouting new leaves from its prostrate vines, than during the cold-dry or hot-dry dormant season, when it was loosing most or all of its leaves, due to its semi-deciduous nature. As a result, it was

selected by cattle significantly more often during its growing than during its dormant season (Table 6.3). This happened because cattle usually selected only individual, young leaves from the distal ends of the prostrate vines for consumption, sometimes taking the distal portion of the vine as well, but generally avoided mature leaves and the vine itself. In the dormant season, cattle occasionally took dry, brown leaves from the vines, avoiding leaves that had already fallen off. At no stage did cattle take the tubers, or part of tubers, even if they had been completely or partially exposed by porcupines. Similarly, cattle were never seen to take flowering parts, or the fruit (seedpod) or the seed itself.



(DM = dry matter, CP = crude protein, Ca = calcium, P = phosphorus, CF = crude fiber, ADF = acid detergent fiber, NDF = neutral detergent fiber, DOM = digestibility of organic matter, ME = metabolizable energy)

Figure 6.1

The nutritive value (%) of those organs of the marama plant that were selected by free-ranging cattle during the hot-wet season (summer) and the cold-dry season (winter).

The marama plant contributed disproportionably much to the nutrition of free-ranging cattle despite its very low abundance in their diet, especially in terms of its high crude Protein content and, in summer, also its high digestibility. Distal ends of vines, including young, fresh leaves, contained about four times as much crude protein as grass (crude protein contents in the range of 5 – 8%), the staple feed of cattle, contained at the same time and place. It is unlikely that perennial, non-woody herbs contain a lot of tannins, so that it can be assumed that most of this protein was also available to the ruminant digestive tract. Even the crude protein content of dry leaves, in winter, was twice as high as that of dormant grasses in winter, although their digestibility was poor. The digestibility of young, distal vines in summer was only slightly higher than that of actively growing grasses. However, the crude fat content of distal vines was about four times higher than that of grasses at the same stage. The analysis of crude fat includes lipids used as energy components as well as aromatic compounds that determine the taste of a plant. In the case of the marama plant, the high fat content of the distal, young vines (Fig 6.1) may indicate a high level of unpalatable aromatic oils, causing avoidance by foraging cattle, the cattle did not avoid it completely, but some took a bite and went on to the other fodder. It is suspected that when the leaves are old and leathery, it has more fibers and the water content is not that much to make up despite contributing to exceptionally high metabolizable energy content reminiscent of that of concentrate feed supplements. The Crude Fiber content of the selected organs were low irrespective of season, indicating the non-woody growth form of this dicot, while its high ash content was likely due to soil pollution of the prostrate vines.

6.5 CONCLUSION

This study indicates that the marama plant is indeed utilized by free-ranging beef cattle, but not preferentially and forms only a very small part of their diet. Dietary abundance is sensitive to seasonal effects, probably because the plant is at different growth stages in different seasons. However, increasing the stocking rate did not entice cattle to consume more of this plant, although it appeared to decrease its botanical abundance, indicating that the plant may be sensitive to defoliation or pruning of its vines. It appears that different cattle types may select the plant more often than others, but it remains a minor dietary component only. The biggest value of the marama plant may be in the large amount of available crude protein it contains, especially during its growing season, typical of leguminous fodder plants. It is also possible that its high crude fat content contains unpalatable aromatic oils that discourage its utilization by foraging cattle. Considering its low preference value by cattle, the plant does not warrant further investigation to turn it into a cultivated fodder plant, despite its apparent hardiness and high yield.

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7

General conclusions and recommendations

The EU project (ICA4-CT-2000-30010 Final Report, 2004) revealed the following scientific understandings of the marama bean plant (*Tylosema esculentum*):

The marama bean plant is widespread with large populations in Namibia, Botswana and smaller populations in the Gauteng area of South Africa. It grows at altitudes between 1 000 and 1 500 m, in summer with 300 to 700 mm rainfall and at a minimum temperature above 15 ºC and a maximum *ca*. 33 ºC. It is dormant in winter, with re-growing from the tuber in spring. Marama grows in well-drained, fine, generally calcareous sands, but also in regions of harder calcareous conglomerates, at pH 6 to 8, with very little organic matter or nitrate and where available phosphate is small. Growth is in open sand veld and in open grass and bush savanna. It grows more vigorously in strong sun than in shade. People in the areas comment little on pests and diseases of the marama, although casual observations showed insect damages.

The questionnaire in Namibia and Botswana showed that the marama is widely and highly regarded as food. Seed is frequently consumed (but tubers much less so) as food, generally roasted when mature. Marama beans are collected in substantial quantities from the wild to eat and are frequently sold in markets. Unregulated

collections may damage populations. People in the area do not grow the plant, but consider it to be a good crop and wish to grow it, but need the know-how. A market exists for the seed, both national and international, but lack of crop with adequate yields prevents exploitation.

There is genetic variation between individuals within populations but no differences between populations. Seed have no physiological dormancy and germinate rapidly after long-term storage. The large store of nitrogen enables rapid early growth for some weeks. Growth from tubers is vigorous and uses nitrogen stored as protein as well as accumulated carbohydrates.

Plants were grown from seed in field trials in Botswana and some flowered in the first year. Seed production is variable, suggesting that selection for higher yielding strains is possible. This agrees with the biodiversity studies. Production is *ca.* 0.1 to 0.3 t/ha and is dependent on adequate rainfall.

Studies in laboratory and veld using gas-exchange methods showed that the marama plant has C3 photosynthesis. The enzyme ribulose bisphospate carboxylase-oxygenase, responsible for assimilating CO_2 in the leaves, has characteristics for hot, dry conditions. The marama plant has bi-lobed leaves, which open in the cooler, moist mornings, but fold together as temperature rises. They also solar-track, presenting the smallest angle to the sun around noon. Stomata also open early and close later in the day, thus optimizing conditions for photosynthesis. Drought induces earlier leave folding and stomatal closure, but elative water content do not decrease greatly.

Vegetative growth is slowed down by water stress and leaves abscise quickly. These features, as well as the dependence of seed formation on rain, show that the marama plant is a “drought-avoiding” species, surviving by storing nutrients and water in the underground tuber.

Studies of nitrogen assimilation with ^{15}Nitrogen and biochemical analyses show that the marama bean plant does not form root nodules with symbiotic bacteria, thus does not assimilate gaseous nitrogen. It rather uses the extensive, deep root stem to remove nitrate from the very poor soil and accumulate it in a large proportion in the tuber. Afronomic trials show that growth from the marama bean plant is not stimulated by application of nitrogen fertilizer, indicating large efficiency in acquiring nitrogen. Similarly, phosphate fertilizers do not stimulate growth, suggesting a large efficiency in acquisition and use of phosphate. .

This study contributed to the EU project by revealing the following scientific understandings:

The chemical analyses between the Namibia and the Botswana roasted marama beans did not differ. The marama bean can be regarded as an extremely valuable supplemental food source. It has a protein content of roughly 36% and like other legumes, is rich in the amino acid lysine, but low in the amino acid methionine. The fat content is between 34% and 40% and approaches that of a peanut. The oil is high in the long chain unsaturated fatty acids, oleic acid (C18:1), with 42% of total fatty acids, as well as high in linoleic acid (C18:2) with 32% of the total fatty acids. Of the many

other nutrients that the marama bean contains, of importance is that it contains folic acid (0.14mg/100g), needed during pregnancy; iodine (0.06 mg/100g), essential for cognitive development; iron (4.79 mg/100g), essential to prevent anemia; vitamin A (0.27 mg/100g), needed for proper eyesight and even vitamin B_{12} (0.01 mg/100g), which is normally only produced in animal tissue.

The Botswana traditionally roasted, Botswana oven roasted and Namibian oven roasted marama samples were proven to have more or less the same sensory attributes. They had a significantly lower burnt and chemical aroma and a slightly higher peanut butter aroma than the Namibian traditionally roasted marama bean. The Namibian oven roasted marama bean samples had significant less intense astringent, bitter, chemical, woody, cocoa and coffee flavour and a slightly higher nutty flavour, a significantly less intense bitter, burnt and coffee after-taste, as well as lower astringent mouth-feel when compared to the Namibian traditionally roasted sample, the Botswana traditionally roasted sample and the Botswana oven roasted sample as indicated in Figure 5.2.

It is recommended to investigate the techniques of the traditional roasting of the marama beans, due to the obvious influence that it has on the aroma, flavour and after-taste of the product and which might have a negative impact on the acceptance of the product. The marama bean samples from the different regions (Namibia and Botswana) should be roasted under identical, controlled conditions and be evaluated by the target groups to define whether the target groups prefer the traditionally roasted or oven roasted method.

This study indicates that the marama plant is indeed utilized by free-ranging beef cattle, but not preferentially so and forms only a very small part of their diet. Dietary abundance is sensitive to seasonal effects, probably because the plant is at different growth stages in different seasons. However, increasing the stocking rate did not entice cattle to consume more of this plant, although it appeared to decrease its botanical abundance, indicating that the plant may be sensitive to defoliation or pruning of its vines. It appears that different cattle types may select the plant more often than others, but it remains a minor dietary component only. The biggest value of the marama plant may be in the large amount of available crude protein it contains, especially during its growing season, typical of leguminous fodder plants. It is also possible that its high crude fat content contains unpalatable aromatic oils that discourage its utilization by foraging cattle. Considering its low preference value by cattle, the plant does not warrant further investigation to turn it into a cultivated fodder plant, despite its apparent hardiness and high yield.

In conclusion : The marama bean plant *(Tylosema esculentum)* is a valuable food, it can be grown readily if better yielding plants are selected. This miraculous plant can be included in programmes specifically aimed at improving household food security and in programmes aimed to alleviate under-nutrition in Southern Africa.

AD FINIS VOLUMINIS

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