Kirkhouse Trust

Stress Tolerant Orphan Legumes

MONOGRAPH SERIES



Marama Bean (Tylosema esculentum)





Marama Bean Tylosema esculentum

Stress Tolerant Orphan Legumes

Monograph Series



The Kirkhouse Trust (KT) is a UK-registered charity founded by Sir Edwin M. Southern to fund the improvement of legume crops that are important for food and nutrition security in African countries and India and to promote scientific education. The origins of KT are entwined with the development of Sir Ed's molecular biology company, Oxford Gene Technology (OGT). In 1997, Oxford University assigned Sir Ed's microarray patents to OGT in exchange for 10% of the equity. In 2000, OGT's income began to grow, and KT was registered as a charity and endowed with an initial donation from the company.

KT's funding model aims to address its twin objectives of improving legume crops, which are important for smallholder farming systems in target countries and raising national scientific capacity. KT has a hands-on strategy, with a team of international scientific consultants working closely with the Principal Investigators (PIs) and students they mentor, providing technological backup as needed, and hosting PIs and students for study visits in their laboratories.

The STOL consortium was established in 2018 under the Promoting India-Africa Framework for Strategic Cooperation Initiative in partnership with the Indian Council of Agricultural Research (ICAR), Department of Agricultural Research and Education (DARE), Ministry of Agriculture and Farmers Welfare, New Delhi, India. The programme aims to facilitate the introduction and exchange of stress-tolerant orphan legume varieties among partnering Indian and African institutions and assess the relative response of selected species to the higher levels of abiotic stresses expected because of climate change. Crops have been identified as potentially having a crucial role in adapting to climate change in arid parts of Africa and India, and selected species are likely to become the focus of KT breeding programmes in the medium to longer term.

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- **Cover Photo:** Marama bean filed trials conducted by Namibia University of Science and technology, Namibia with financial support from the Kirkhouse Trust.
- **Credit:** Professor Percy Chimwamurombe, Head of Department: Biology, Chemistry and Physics, Namibia University of Science and Technology, Namibia

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FOREWORD

Roughly 2.5 billion people (30% of the world's population) live in semi-arid regions, and approximately a third of these people depend on agriculture for their food security and livelihood. Crop production in these regions has always faced challenges associated with excess heat, drought, a highly variable climate, land degradation, and a loss of biodiversity, which has been exacerbated in recent times by climate change, limited access to technology, poor market linkages, weak institutions, and lack of national and international partnerships. A possible strategy to cope with



climate change is to switch from the cultivation of current crops to ones which are more drought-hardy. These include several minor legume crops, commonly known as orphan legumes, currently being grown to a limited extent in the drier regions of both Africa and Asia to provide food and nutritional security to households. These species have remained relatively neglected by both researchers and industry because of their limited economic importance in the global market.

To promote these orphan legumes, the Kirkhouse Trust initiated a consortium programme on "Stress Tolerant Orphan Legumes (STOL)" in partnership with several African countries and India. The STOL programme aims to facilitate the introduction and exchange of stress-tolerant orphan legume among partnering Indian and African institutions and assess the relative response of selected species and varieties to the higher levels of biotic and abiotic stresses expected because of climate change.

To facilitate the better understanding and cultivation of these new crops among Indian and African partners the STOL project is supporting the publication of a series of monographs for selected orphan legumes and Marama bean (*Tylosema esculentum*) is one of such crops.

I congratulate the author of this monograph Professor Percy Chimwamurombe, Head of Department: Biology, Chemistry and Physics, Namibia University of Science and Technology, Namibia for compiling and synthesising information to bring out the Marama bean monograph, which the Kirkhouse Trust is pleased to publish as part of the STOL monographs series. I am sure this publication will enlighten the policymakers, scientists, extension personnel, entrepreneurs and farmers for the improved production and consumption of Marama bean across African countries as well as in India.

Edwin Southan

Professor Sir Edwin M. Southern Founder & Trustee of the Kirkhouse Trust







PREFACE

Sub-Saharan Africa has been suffering the brunt of global climate change, in particular experiencing an increased frequency and severity of drought. At the same time, the region is endowed with a number of indigenous plant species which have evolved to tolerate long periods of abiotic stress. As yet, however, little effort has been devoted to apply modern methods of selection and breeding to improve these species.

A prime example of an under-utilized Africa legume is the marama bean (*Tylosema esculentum*), a very hardy, drought tolerant, non-nodulating perennial species able to live for as long as 15 years. Various parts of the plant provide a source of starch, protein, essential oils and molecules of pharmaceutical value. The plant not only tolerates heat and drought better than other legumes can, but is also grow in the acidic, phosphorus-deficient soils which are commonplace in the Kalahari desert. Despite these advantages, its area of cultivation is small, reflecting a lack of investment targeting its genetic improvement. A full realisation of its economic potential through the development of stable and high yielding varieties exhibiting enhanced resilience against pests and diseases should serve to boost its popularity and expand its area of cultivation. While some scientific literature exists describing various aspects of marama bean's taxonomy, physiology and agronomy, these reports are scattered and in many cases are rather inaccessible. It is against this background that seeks to collate both genetic and non-genetic aspects of marama bean improvement using both published and unpublished materials.

It is recognized that this monograph will need to be updated as additional literature is generated, but the hope is that it can provide a basic reference document for researchers choosing to work on this unusual legume species.

The author wishes to acknowledge the long term support provided by the Kirkhouse Trust towards improving the status of marama bean, which has included covering the cost of writing and publishing this monograph.

In addition, thanks are due to Drs Prem Mathur and Robert Koebner, acting as consultants to the Kirkhouse Trust, for their critical reviews and constructive comments.

> Professor Percy Chimwamurombe Head of Department: Biology, Chemistry and Physics Namibia University of Science and technology



1. INTRODUCTION

Marama bean (*Tylosema esculentum*), an orphan legume crop, is a wild, tuber-producing, non-nodulating and perennial legume which is well adapted to drought-prone environments. Its seed contains proteins and oils of superior quality to those present in the seed of most cultivated legumes. It produces numerous prostrate stems of up to 3 m in length (Figures 1, 2 and 3) which emerge from a potentially large woody tuberous root (Figures 4 and 5) (Powell, 1987). The seeds, each averaging in weight ~3 g (Figure 6), and tuberous roots (Figure 4) are a source of both nutrition and pharmaceutically active compounds. In the field, phenotypic variation has been observed for internodal length (Figures 7, 8 and 9), the number of seeds set per pod, the number of pods formed per stem, leaf



Fig. 1: A vegetative marama bean patch from Omaheke region, Namibia.



Fig. 3: A large vegetative growth patch of marama bean.



Fig. 2: A marama bean podding vine.



Fig. 4: A young edible marama bean tuberous root.



Fig. 5: A three-year-old woody marama bean tuberous root.



Fig. 6: Mature dry and dark brown marama bean seeds.



Fig. 7: A marama bean plant with a short internodal length (3-4 cm).



Fig. 8: A marama bean plant with a medium internodal length (5-8 cm).



Fig. 9: A marama bean plant with a long internodal length (9-15 cm).

size and colour, stem colour, flowering time and stem die-back time. However, the genetic basis of this variation has yet to be determined.

Marama bean grows in sandy, arid, or semi-arid soils (Figure 10) where few conventional crops can survive. In its native habitat, the plant can withstand summer temperatures reaching up to 50°C and freezing winter temperatures, and the presence of surface water for only eight weeks during the year (Powell, 1987; Bower *et al.*, 1988, Nepolo *et al.*, 2009).

It grows naturally in Namibia and Botswana (Figure 11) in areas where the cultivation of other crops is difficult (Uzabakiriho, 2016). In Namibia its natural



Fig. 10: A marama bean seedling (one-month post germination) growing in red sandy, arid, or semi-arid areas.



Fig. 11: World geographical locations for marama bean cultivation (revised from Omotayo and Aremu, 2021).

distribution is concentrated in Omaheke and Otjozondjupa regions. On an experimental basis, it is presently being grown in Australia, Israel, Kenya, the United States and India.

Marama bean forms an important part of the local diet for the Khoisan and Bantu people native to Southwest Africa, thanks to the high protein content of its seed and the high carbohydrate content of its tuberous roots. The seed contains an estimated 29.4% protein, significantly more than is present in the seed of most legumes.

2. ORIGINS AND CENTRE OF DIVERSITY OF MARAMA BEAN

Marama bean, sometimes referred to as "green gold," is native to the Kalahari desert in Southwest Africa. It is not grown commercially, but its potential is considerable. Its native habitat experiences regular episodes of both drought and high temperature. In the far west of the Kalahari desert, where the range in annual rainfall is 250-500 mm and precipitation is typically sporadic, the plants experience both flooding and long periods of drought – in particular, years featuring almost no precipitation are relatively common (Cullis *et al.*, 2018). Marama bean plants grow patchily (Figure 12) on very sandy soils containing very low levels of organic matter, nitrogen and phosphorus (Cullis *et al.*, 2018). The average maximum daily temperature during the summer growing season is 32°C (range 28°-37°C) and the light intensity during the afternoon is particularly high (~2,000 µmol m⁻²s⁻¹) (Cullis *et al.*, 2019).



Fig. 12: A patchy marama bean plant growing on the roadside.

3. TAXONOMY

Marama bean belongs to the genus *Tylosema* (Schweinf.) [Torre and Hillc], a member of the *Caesalpinioideae* sub-family of the *Fabaceae*. Members of the genus were previously grouped with those belonging to genus *Bauhinia* (Hao *et al.*, 2003), which harbours more than 500 species of flowering plants widely cultivated as ornamental trees in tropical Asia; but they were later reclassified as a separate clade (Wunderlin, 2010). The *Tylosema* genus comprises four taxonomically accepted species [*T. esculentum* (Burch.) A. Schreib., *T. fassoglense* (Schweinf.) Torre and Hillc., *T. argenteum* (Chiov.) Brenan and *T. humifusum* (Pic. Serm and Roti Mich.) Brenan]. These species are distributed across eastern, central and southern Africa (Castro *et al.*, 2005). *Tylosema* flowers develop up to ten stamens, of which only two are fertile; the stamens are variously shaped and coloured. A lobed non-spathaceous calyx-limb is also present (Castro *et al.*, 2005).

The relationship between the four *Tylosema* species has been investigated using both palynological and molecular approaches, particularly based on chloroplast markers such as the *matK* gene (Banks *et al.*, 2013; Hao *et al.*, 2003, Azani *et al.*, 2017). The separation between *Tylosema* and *Bauhinia* has been confirmed by comparisons between both chloroplast and nuclear genome sequences (Kim and Cullis, 2017). *T. esculentum* and *T. fassoglense*, as well as a putative Angolan species (Castro *et al.*, 2005), may represent a single, genetically diverse species. Marama bean fits well with the little used "ochlospecies" concept first proposed by White (1998), as is the case for many African taxa (Cronk, 1998).

In addition to its usual English name of marama bean, the species is sometimes called the gemsbok bean; in Afrikaans it is variously known as maramaboontjie, elandsboontjie or braaiboontjite, in Tswana as either marama or morama, in Thonga as maramama, in !Kung San as tsi tsin, in Khoi as gami and in Herero as ozombanui (van Wyk and Gericke, 2000).

4. DESCRIPTION OF THE PLANT

The marama bean plant is prostrate, forming a number of vines which creep along the ground. Its leaves are bilobed and show a distinct net venation. Its flowers are bright yellow, and the spherical seeds it sets become dark brown when mature. The internode length is very variable (Figure 7-9).

4.1 Uses of marama bean

Mature marama bean seed is consumed following either roasting, frying or pounding. In addition, the seed is pressed for its oil or used to prepare a non-dairy, nutritious milk (Müseler; 2005). Seed kept within the pod has a long shelf life thanks to its hard testa. The seed provides a source of protein, fibre and healthy fats (Table 1). The species contributes 75% of the total vegetable content of the diet of people belonging to tribes living in the Kalahari desert, who typically roast the seed in hot sand or coal. The roasted seed is either eaten without further processing, or is pounded and boiled with water to make a porridge (Vietmeyer, 1986). Seeds are not usually eaten raw, possibly because of their unpleasant texture and taste. Raw seed also harbours a high content of trypsin inhibitors, which are neutralized by cooking (Powell, 1987; Bower *et al.*, 1988). The flavour of the cooked seed resembles that of almond or cashew. Some farmers use marama bean as a food supplement for fattening pigs (Mataranyika *et al.*, 2020). The oil extracted from the seed has a pleasant and sweet odour and is therefore used by the food and cosmetic industries (Amarteifio and Moholo, 1998).

Class	Nutrient	Marama bean	Soybean	Groundnut
Proximate	Ash (%)	3.19	4.50	3.80
	Dry matter (%)	96.22	92	95
	Fat (%)	40.06	25	50
	Moisture (%)	2.67	7	9
	Non-structured carbohydrates (g)	11.85	15	20
	Protein (%)	34.71	45	25

Table 1: Proximate analysis (per 100 g dry matter) of marama bean, soybean and groundnut (Adapted from Omotayo and Aremu, 2021).

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Marama bean is also an important source of various phytochemicals and nutritional components. The seeds contain the three lignans secoisolariciresinol, lariciresinol and pinoresinol (Holse *et al.*, 2010), and are widely used in the functional food, pharmaceutical and nutraceutical industries.

The carbohydrate content reported above has been presented as an estimate, because the samples had been dried before analysis, leaving only a low amount of moisture. The carbohydrate content ranged from 19% to 36% (Müseler, 2005), with the highest calculated percentage being assumed to be anomalous as it deviated from previously determined maximum amounts of carbohydrates. Prior studies have estimated the carbohydrate content of marama bean samples from Botswana, Namibia, and South Africa to lie between 19% and 24% (Mataranyika *et al.*, 2020). Dietary fibre was the most abundant form of carbohydrate present. The mean carbohydrate content was approximately 14 g per 100 g.

Mataranyika et al. (2020) determined that the range in seed protein content of Namibian marama bean accessions lay between 30.1% and 34.8%. Meanwhile, as reported by Müseler (2005), a small-scale analysis of samples harvested between 2001 and 2004 exhibited a higher range of crude protein content (34.0-36.9%). It has been recognised that the climatic conditions prevailing during the sampling season have less impact on nutritional composition than does the seasonal influence on plant growth (Müseler, 2005). This level of protein content is similar to that of soybean seed (mean 37.7%, range 36.9-40.1%), thus marama bean also represents a suitable candidate source for the formulation of nutrient supplements and food alternatives (Mataranyika et al., 2020). Marama bean is superior to other commonly consumed legumes, such as cowpea (crude protein content range 23.2-28.1%) or common bean (20.1 \pm 0.52%). Furthermore, Mataranyika *et al.* (2020) determined that marama bean seed is rich in calcium (241 mg per 100 g), magnesium (274 mg), and zinc (6 mg), all of which minerals are essential for a variety of physiological processes. Additionally, it provides a source of vitamins B6 (1.56 mg) and B12 (0.043 mg), along with trace levels of iodine (0.06 mg), iron (3.95 mg) and copper (1.04 mg) (Müseler, 2005).

T. esculentum tubers can grow to an enormous size – weights of up to 250 kg are not uncommon. Young tubers can be roasted and eaten (van Wyk and Gerick, 2000), but old ones become very fibrous. Tubers have a higher protein content than those of cassava or yam (Müseler, 2005) those Livestock and game in Kalahari have been reported to feed on fresh marama bean foliage (National Academy of Sciences, 1979) but other authors dispute this (Powell, 1987). Perhaps taking advantage of the presence of phenolic compounds in the leaves, natives of the

Kalahari desert are known to crush the leaves into a thick paste which is used to treat wounds and arthritis (Koenen, 2001).

4.2 Mode of reproduction and seed formation

In its natural stands, the marama bean plant takes between 8 and 21 days to germinate provided moisture is available; thereafter, the plant grows vegetatively during the summer. During its vegetative growth phase, an underground tuber begins to develop. This tuber will sustain the plant nutritionally over the winter months after the runners have died back. At the start of the following rainy season, new runners sprout from the tuber. Flowering is initiated in the third year, when the plant is 18-24 months old. The usually yellow flowers (Figure 13) are pollinated by solitary bumble bees.



Fig. 13: Multiple flowers on a marama bean runner.

The cycle of runner growth, flowering and runner die-back repeats itself each year. Mature pods shatter powerfully to facilitate seed dispersal (Figure 14). Young pods start as purplish in colour and later turn green (Figure 15) and finally brown when the seeds are mature.



Fig. 14: A marama bean plant bearing mature pods.



Fig. 15: Green pods of a marama bean plant (2 seeds per pod).

4.3 Interactions with microorganisms

Several bacterial species have been isolated from soil sampled from the rhizosphere of marama bean plants (Kandjimi et al., 2015). All of these could be shown, using a plate assay, as being able to produce ammonia. Although ammonia released by bacteria may be an important source of nitrogen, detailed information is still lacking regarding the ability of marama bean to process ammonia, a form of nitrogen which most plants are not well adapted to tolerate (Weise et al., 2013). The soil microbiome associated with marama bean growing across Namibia is currently being characterized by assaying bacterial 16S and V3-V4 sequences, along with fungal ITS 1 DNA sequences. Some of the endophytes harboured within the seed may contribute to the species' nutritional efficiency, since they have a striking capacity to harness nitrogen into seed protein (Chimwamurombe et al., 2016). The identity of the mycorrhizal fungi, known to support plants' capacity to tolerate drought stress (Rapparini and Peñuelas, 2013), present in the marama bean rhizosphere is also of interest. The enhancement of tolerance of plants to water deficit by mycorrhizae may involve the regulation of drought-induced plant genes, such as those encoding aguaporins, both via the down-regulation of genes encoding plasma membrane aquaporins (Porcel et al., 2006) and/or the enhanced expression of specific aquaporins (Li et al., 2012).

5. GENETIC RESOURCES

Under Namibian conditions, the optimum period to collect seed is between April and early June. Most marama bean seed is collected from wild stands, although a small number of experimental plots have now been established to reduce losses caused by herbivory. The Namibia University of Science and Technology (NUST) set up an initial collection of 391 accessions, later extended to 521 accessions, sampled from diverse wild stands in Namibia (Table 2). Seed of these accessions is stored at room temperature (Figure 16). Natural populations of marama bean are under pressure from both grazing animals and humans, which has prompted investigations of their genetic constitution to develop suitable strategies to both conserve and develop marama bean germplasm (Naomab, 2004). Some progress has been made to characterize variation at the physiological and agronomic level, but more research is still needed to gain an understanding of the species' genetic diversity and the genetic basis of traits relevant for domestication (Travlos *et al.*, 2007).

Seed requests submitted to NUST will need to be accompanied by a formal Material Transfer Agreement.

Location and Population name	Way point	Position	Number of accessions
Ozondema	1	S20 15.921 E18 02.490	25
Ombojondjou	2	S20 18.600 E17 58.525	26
Osire	3	S21 02.031 E17 21.244	96
Otjiwarongo	4	S20 46.092 E16 65.123	50
Omitara	5	S22 21.596 E18 02.476	19
Sandveld	6	S22 01.751 E19 08.009	25
Otjovanatje	7	S20 27.39 31 E16 39.443	20
Omipanda	8	S21 19.355 E20 04.553	31
Post 3/Epukiro	9	S21 39.642 E19 25.092	30
Harnas	10	S21 47.705 E19 19.921	25
Okomumbonde	11	S20 57.000 E18 55.000	44
Total Accessions			391

 Table 2: Geographical positions of the sites of Tylosema esculentum populations in Namibia.



Fig. 16: Harvested marama bean seeds in storage in khaki bags and canvas sacks.

5.1 Use of microsatellites for diversity analysis

Some 80 functional microsatellite primer sets have been developed, using a protocol modified from that described by Zane *et al.* (2002). Initially these were generated with a view to revealing the genetic diversity present in the species. Polymorphism with respect to both amplicon size and band intensity has been observed. The extent of heterozygosity, based on microsatellite genotyping, present within and between 11 populations has been assessed by Nepolo *et al.* (2009). While some of the populations appeared to be genetically quite uniform, others were very heterogeneous. The material includes variants with respect to a number of phenotypic traits (Figures 7-9).

6. BREEDING SYSTEMS

6.1 Breeding objectives

Several breeding objectives have been identified to improve marama bean. One of the leading ones is to increase the seed number per pod from one or two (Figure 16) to at least five. A second priority is reducing the generation time from 18-24 months to 12 months. Among the other traits which need to be targeted are to reduce the tendency of the pods to shatter at maturity, to alter plant habit from its prostrate form to a more erect type and to overcome the plants' selfincompatibility to permit self-fertilization.

While some progress has been made towards characterizing the diversity of marama bean, identifying its pathogens and describing its physiology (Nepolo, 2010), the knowledge base regarding its reproductive biology is as yet insufficient. Extending this will be needed to allow for the elaboration of strategies to improve the genetic potential of currently available germplasm in terms of flowering ability, seed set and ultimately seed yield. Note that although the plant does produce an edible tuber, seed is the most important target for the plant's domestication and improvement. The mode of inheritance of none of the key traits has yet to be elucidated, although, based on what is known from other domesticated legume species, at least some of them are likely to be under major gene control.

6.2 Breeding methodology

The genetic variability and distribution of the Namibian marama bean germplasm has been characterized with a view to establishing a breeding programme. This preliminary assessment has set the stage for a more detailed investigation of the genetic and associated phenotypic variation present. Since the conservation and use of plant genetic resources for domestication depend on identifying the extent of genetic variation available (Monaghan and Halloran, 1996), efforts have been made to document the geographical distribution and diversity of the species within Namibia. The latter has been enabled by the development of a suite of microsatellite assays, which have been applied to amplify genomic DNA extracted from the embryonic axis of germinating seeds.

As with any breeding programme, the initial focus is on available genetic materials (Karademir *et al.*, 2011). A breakthrough in the successful domestication of marama bean can only be attained through such a pre-breeding exercise. At the same time, there is a need to broaden the genetic base of the crop.

7. AGRONOMIC PRACTICES AND ECOLOGY

7.1 Agronomic practices

Despite its potential as a food and cash crop (Amarteifio and Moholo, 1998), very little agronomic research has been conducted on marama bean cultivation. Investigations conducted at NUST have shown that the seed-to-seed cycle is very lengthy. In the first year, four months of vegetative growth end with winter dieback, during which season the plant is quiescent for four months. Re-sprouting commences in the following year. The number of second year runners is typically double that generated in the first year. Cultivators only need to control weeds, rodents and pests, while neither irrigation nor fertiliser application are necessary. During the third season, the plant will typically flower and set seed.

7.2 Crop physiology

The marama bean plant faces several survival barriers in its natural, dry and arid environment. However, they have the ability to grow under climatic conditions not tolerated by crops such as maize or pearl millet (Nepolo, 2010; Mataranyika *et al.*, 2020). It exhibits some typical, along with some rather unique drought avoidance strategies. In response to a reduction in the level of soil moisture, the plant reacts by limiting the elongation of the runners and the formation of new leaves and secondary stems; some of the runners die back completely (Mitchell *et al.*, 2005). In contrast, in the presence of non-limiting soil moisture, many highly branched runners are formed, along with a profusion of leaves (Mitchell *et al.*, 2005). Unusually for a leguminous plant, a substantial tap root develops, which allows the plant to access water a long way down the soil profile (Comas *et al.*, 2013). Since the plant encounters mostly sandy soils, some water remains accessible by the tap root for several months after the rainy season has passed.

A marama bean plant is capable of colonizing a substantial area of the soil surface due to its development of multiple runners (Keegan and van Staden, 1981). Its ability to colonize a substantial area with its broad leaves and runners most likely helps the plant to withstand the dehydration stress imposed by wind-induced evaporation. A further typical drought avoidance trait is represented by the plant's tendency to react promptly to moisture stress by closure of its stomata (Mitchell *et al.*, 2005), a physiological adaptation not generally observed in other legume crop species, which are unable to prevent the development of large

differences in leaf water potential between drought-stressed and non-stressed leaves (Villalobos-Rodriquez and Shibles, 1985). Another adaptation, shared with some other plant species, succeeds in reducing the leaves' exposure to sunlight, particularly under drought conditions, by adjusting the angle between the leaf and the stem (Travlos *et al.*, 2008), thereby reducing the flow of transpiration (van Zanten *et al.*, 2010; Rakocevic *et al.*, 2017).

While many legumes develop a symbiotic relationship with soil bacteria belonging to the genus *Rhizobium* which enables the fixation of atmospheric nitrogen, marama bean does not develop root nodules (Dakora *et al.*, 1999). The plant's water-use efficiency is typical of a C3 plant, and its rubisco kinetics are consistent with its adaptation to hot, drought-prone environments (Mitchell *et al.*, 2005).

7.3 Major diseases and insect pests

Little attention has been paid to date to documenting the pests and pathogens of marama bean. Using ITS sequencing, Uzabakiriho (2016) has reported the presence on marama bean leaf and pod tissues of Phoma spp. and Alternaria tenuissima. The latter species, like its related species A. alternata, is a potential alternariol- and tenuazonic acid-producing microorganism. According to Takundwa et al. (2014), it is also possible to identify the presence on marama bean leaf lesions of Penicillium brevicompactum, Rhizopus stolonifer, P. olsonii, Fusarium chlamydosporum, F. equiseti and A. solani. As yet, there is no evidence for the presence of any mycotoxin-producing species. The marama bean plant produces appreciable amounts of various secondary metabolites, including polyphenols (Shelembe, 2012). Some of these may be involved in its defence against fungal attack (Mayer et al., 2014). Shelembe (2012) have noted the presence of both vanillic acid and gallic acid (inter alia), known to inhibit the synthesis of aflatoxin by A. flavus (Mayer et al., 2014). The polyphenol chlorogenic acid is documented to have a negative influence on the expression of the alternariol synthesis (Wojciechowska et al., 2014).

8. PRODUCTION AREA

Marama bean plants are widely distributed across Namibia, but occur patchily. The Namibian locations where the plants currently grow have been documented using GPS technology and recorded into a database by Nepolo *et al.* (2009). These include Ozondema, Ombujondjou, Osire and Otjiwarongo in the Otjozondjupa region; Omitara in the Khomas region; and Sandveld, Otjovanatje, Omipanda, Post 3, Harnas and Okomombonde in the Omaheke region. The initial set of 391 accessions (later increased to 521) was initially sampled from each of the locations as presented in Table 2.

9. CURRENT RESEARCH PRIORITIES

Even though marama bean remains a largely non-domesticated plant, the nutritional composition of its seed is of interest in view of the global pressure to reduce the reliance of the human diet on animal products. The current research priorities for marama bean include the selection of the best-performing accessions and the determination of which phenotypic attributes are associated with productivity. Little is known as yet concerning the range of phenotypic variation which exists in nature or what could represent good agronomic practice. Limited knowledge remains the major barrier to domestication. There may in future be scope to breed varieties which can be used to develop high value food products such as butter and non-animal milk products.

10. RESEARCH CONTACTS AND RESEARCH CENTRES

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11. SUMMARY AND CONCLUSIONS

This monograph has compiled most of the existing knowledge regarding marama bean. It has also exposed the existing gaps which need to be filled to advance the development of marama bean as a cash crop and as an alternative protein source crop of the future in a climate change-prone world. The breeding of superior, better performing genotypes is still work-in-progress, which requires financial and technical support and attention.

Developing countries need to use under-appreciated native plants to improve their overall food security, health, and economic viability. Marama bean is popularly called "green gold" thanks to the value of both its above-ground (seed) and belowground (tuber) organs. The nutritional value of its seed is comparable to that of the leading legume species groundnut and soybean. The seed also contains ample quantities of secondary metabolites, notably phenolic acids, phytosterols and flavonoids, while the tubers contain behenic acid, grifonilides and carbohydrates. Only sparse descriptions are available in the scientific literature regarding the nutritional, medicinal and economic value of the plant, primarily because of a lack of clear research objectives and the limited resources devoted specifically to this indigenous plant.

Innovative approaches are needed, including the use of molecular techniques, to accelerate the selection of domestication-associated traits. An improvement in the plant's amenability to formal cropping will surely boost the commercial and economic potential of marama bean and its contribution to a sustainable food production and nutrition.

For marama bean, the way forward is to intensify the effort devoted to its domestication. The reality of climate change and its negative effects on most parts of the world, particularly in sub-Saharan Africa, calls for the development of drought-tolerant legumes. In addition, it will be important to intensify, alongside with domestication efforts, the development of value-added marama bean products. The leading reason for domesticating marama bean is that the ecological niche occupied by the plant is not suitable for the cultivation of either soyabean or groundnut, as these species are incapable of tolerating extreme drought conditions.

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