Use, Physiology and Genetic Characterisation of Selected Natural Populations of *Adansonia Digitata* in Malawi

by
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Declaration

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Abstract

Adansonia digitata (Baobab) is among the important indigenous fruit tree species identified for domestication and commercialization in southern Africa. Trade in baobab products has recently increased locally, regionally and internationally. A socio-economic study was undertaken in four districts of Malawi including one in an urban setting in order to determine traditional use of the tree and marketing of baobab products. A morphometric and molecular study was done to determine within and between provenance variability, differentiation, genetic control of traits; and gene flow in five populations selected from four silvicultural zones. Reproductive process (mating system, pollinating agents), fruit and seed physiology of *A. digitata* was investigated in a natural population in Malawi. The research showed that revenue accrued per month per person ranged from 2.5 to 715 USD; while cottage industries made between 1100 and 2300 USD; with net profits of about 595 USD per person and 1535 USD from the enterprise. The study further gives an insight in the potential and viability of the baobab industry on the *proviso* that there are enabling government policies that facilitate improved tenure, thereby promoting domestication and commercialization of the Baobab fruit resource. Morphometric traits in fruit traits, seed germination and seedling traits were highly significantly different ($P \leq 0.001$) within and between populations. Mean fruit weight ranged from 125.8 ± 3.25 g to 162.9 ± 3.25 g, seed weight ranged from 38.6 ± 2.5 g to 66 ± 2.01g and pulp weight ranged from 28.7 ± 1.33 g to 41.4 ± 1.33 g. The rich diversity in fruit traits found between and within populations is important for domestication purposes and tree improvement through selection and breeding. Coefficient of variation (CV) in seedling growth variables ranged from 18.6% to 43.6%. Individual narrow sense heritabilities (h^2) ranged from 0.07 to 0.71. Additive Genetic Coefficient of Variation (AGCV) ranged from 3.21% to 14.67%. High and moderate heritability and AGCV values show the potential that baobab can also respond well to tree improvement. In addition, high phenotypic variation found gives an opportunity to effect selection at both provenance and tree to tree level. Molecular results have revealed low mean genetic diversity in most genetic diversity indices; Nei's genetic diversity (h, 0.18±0.03), Shannon Information Index (I, 0.21±0.07), observed number of alleles (na, 1.47±0.10), effective number of alleles (ne, 1.23±0.04) and percentage polymorphic loci (pp, 48%). Low genetic variation could be attributed to the population growing in marginal areas of genetic centre of diversity as well as climatic and anthropogenic factors. Moderate genetic differentiation were observed among populations ($G_{st} = 0.13$) which could be as a result of long-distance gene dispersal having a homogenisation effect. Clustering of individual trees by genetic similarity coefficients, seedling traits and individual seed weight showed two major baobab races: the Mainland and Likoma Island populations. Autogamy (spontaneous self-pollination) lead to fruit development which aborted within the first month showing strong self-incompatibility. Wind pollination resulted into successful fruit development but growth was eventually inferior compared to naturally pollinated fruits probably inferring ineffective pollination (pollen limitation). This proved that the species has a mixed pollination system and not exclusively bat pollinated. The high fruit abortion and pest predation further warrants intensive study that will help to understand the biology and ecology of both the *Adansonia digitata* and the associated pests and diseases.

Opsomming

Adansonia digitata (kremetart) word as een van die belangrikste inheemse bronne van vrugte beskou en is geoormerk vir veredeling en kommersiele produksie in Suidelike Afrika. Handel in produkte vanaf kremetartbome het toegeneem, beide plaaslik en internasionaal. ‘n Sosio-ekonomiese studie was onderneem om die tradisionele gebruike en bemarking van kremetartprodukte in vier distrikte in Malawi, insluitend ‘n stedelike area, te ondersoek. ‘n Morfometriese en molekulêre studie was onderneem om die variasie tussen, sowel as binne verskillended herkomste, die differensiasie, genetiese beheer van eienskappe en geenverspreiding in vyf populasies kremetarte vanuit vier boskultuur sones te bestudeer. Voortplantingstrategieë (paring stelsels, bestuiwingsagente), sowel as vrug en saad fisiologie van *A. digitata* is ondersoek in ‘n natuurlike populasie in Malawi. Die navorsing het getoon dat die inkomste verhaal, per persoon per maand, vanuit *A. digitata* wissel van 2.5 tot 715 USD, terwyl tuisnywerhede tussen 1100 en 2300 USD per maand maak, met ‘n netto wins van ongeveer 595 USD per persoon en 1535 USD vir die tuisnywerhede. Die studie het verder insig verskaf rakende die potensiaal en lewensvatbaarheid van die kremetartindustrie, met die *proviso* dat daar regeringsbeleid is wat verhoogde verblyfsreg fasiliteer en sodoende die aanplanting en kommersialisering van die kremetart hulpbron bevoordeel. Morfometriese verskille in vrugeienskappe, saadontkieming en saailingeienskappe was hoogs statisties verskillend ($P \leq 0.001$) binne en tussen populasies. Die gemiddelde gewig van vrugte het gewissel van 125.8 ± 3.25 g tot 162.9 ± 3.25 g, saad gewig het gewissel van 38.6 ± 2.5 g tot 66 ± 2.01g en pulp gewig tussen 28.7 ± 1.33 g tot 41.4 ± 1.33 g. Die ryk diversiteit van vrug eienskappe is belangrik vir verbetering van bome deur seleksie en teling. Die koeffisient van variasie (CV) in die groei van saailinge het gewissel van 18.6% tot 43.6%. Individuele oorerflikhede (h^2) het gewissel van 0.07 to 0.71. Toevoegende genetiese koeffisient van variasie (AGCV) het gewissel van 3.21% tot 14.67%. Hoë, sowel as gemiddelde, oorerflikheid en AGCV waardes toon aan dat kremetarte goed kan reageer in seleksie en teelingsprogramme. Addisioneel, die hoë fenotipiese variasie verskaf die geleentheid om seleksie toe te pas, beide by die herkoms en individuele boomvlak. Molekulêre resultate toon ‘n lae gemiddelde genetiese diversiteit vir meeste van die genetiese parameters; Nei se genetiese diversiteit (h, 0.18±0.03), die Shannon Informasie Index (I, 0.21±0.07), waargenome aantal allele (na, 1.47±0.10), effektiewe aantal allele (ne, 1.23±0.04) en die aantal polimorfiese lokusse (pp, 48%) was almal gemiddeld tot laag. Die lae genetiese variasie kan moontlik toegeskryf word aan die feit dat die populasie bome groei in marginale areas van die genetiese punt van diversiteit, sowel as aan klimaats en menslike faktore. Matige genetiese differensiasie was gevind tussen populasies ($G_{st} = 0.13$). Dit kan wees as gevolg van langafstand geen verspreiing wat ‘n homogenisasie effek het. Groepering van individuele bome op grond van genetiese similariteits koëffisiënte, saalingeienskappe en individuele saadgewig dui op twee hoof kremetartrasse: die hoofland en die Likoma eiland populasies. Outogamie (spontane selfbestuiwing) lei tot vrugontwikkeling wat binne die eerste maand aborteer, aanduidend van ‘n sterk self-onverenigbaarheid. Windbestuiwing het gelei tot suksesvolle vrugontwikkeling maar groei was minderwaardig in vergelyking met vlêrmuis bestuifde blomme, waarkynlik a.g.v. onvoldoende bestuiwing (stuifmeelbeperking). Dit toon dat die spesie ‘n gemengde bestuiwingsisteem het en nie uitsluitlik deur vlêrmuise bestuif word nie. Die hoë persentasie vrugaborsie en insekpredasie benodig intensiewe studie wat sal help om die biologie en ekologie van beide *A. digitata* en sy geassosieerde peste en siektes te verstaan.

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Dedication

This work is dedicated to my late brothers Gudu and Yesaya and sister Josiphine Yagontha Munthali. May your souls rest in eternal peace.

Table of contents

List of tables

List of figures

1 GENERAL INTRODUCTION

1.1 Background

Adansonia digitata commonly known as baobab, is one of the species of those renewable natural resources that an estimated 80% of Malawians depend on for their subsistence and household income (Malawi Government 2011). Hence sustainable management of forest resources could contribute to enhanced economic growth and poverty reduction. Unfortunately, these resources are degrading at an alarming rate due to unsustainable use largely arising from high population growth rate, poverty, agricultural expansion, inappropriate management practices, low capacity of government enforcement of rules and regulations and weak policies (Malawi Government 2011).

Malawi is a land –locked country with a total area of 119,140 km^2 of which 20,902 km^2 (20 %) is made up of inland waters dominated by Lake Malawi The country lies between latitudes 9^{o}22' and 17^{o}03' and longitude 33^{o}40' and 35^{o}55'E; and is located in Southern-Central Africa, bordered by Tanzania to the north, Zambia to the west and Mozambique to the east and south (Malawi Government 2011). Approximately 27 % of total land area of Malawi is under forest cover (Malawi Government 2011; Mauambeta et al. 2010). The average rainfall ranges from 900 mm to 1200 mm per annum; and the mean annual minimum and maximum temperatures range from 12^{o}C to 32^{o}C. Temperatures can reach up to 38^{o}C in rift valley and lakeshore. Malawi's soils are dominated by the Eutric leptisols (lithosols), the Chromic levisols (latosols) and the Haplic lixisols (Hardcastle 1978; Malawi Government 2005). By 2008, the population of Malawi was estimated at 13.63 million people growing at 2.6% per annum (Malawi Government 2011). The country has a relatively small economy with 2007 Gross Domestic Product (GDP) of US$3.5 billion equivalent to per capita income of about US$230 to 250 with 76% of people earning less than US$2 per day (Malawi Government 2009). In 2008, about 40% of the country's population was poor, with 15% of them considered as ultra poor (Malawi Government 2011). Prudent use of *Adansonia digitata* will help to uplift the economic status of the country and improve the livelihood of many rural communities.

Adansonia digitata is largely part of the Miombo and Mopane woodlands in southern Africa. The most extensive vegetation in southern Africa is the Miombo (or muombo) largely divided into wet and dry miombo (White, 1983; Chidumayo and Gumbo 2010). The woodland is recognized for its species richness and the widespread occurrence of the tree genera *Brachystegia*, *Julbernadia* and *Isoberlinia* (White, 1983; Chirwa et al. 2008). Wet miombo receive rainfall of over 1000 mm per year whereas dry miombo receive less than 1000 mm per year. Wet miombo covers 1.36 million km^2 extending from near the coast in central and northern Angola, through northern Zambia and southern Democratic Repulic of Congo to central and northern Malawi and western Tanzania with small extents in northern Mozambique and Burundi. Dry miombo covers 1.21 million km^2 extending from southeastern Angola, through northern Zambia and Zimbabwe to south, central and northern Mozambique, southern Malawi and much of central and southern Tanzania. The other extensive woodland in southern Africa is the Mopane woodland, covering 380,000 km^2 extending through southern Angola, northern Nambia, northern and eastern Botswana, Zimbabwe, northern South Africa, southern Zambia, southern Malawi and south and central Mozambique receiving rainfall ranging from 400-700 mm per year. The soil characteristics (soil depth, soil texture, nitrogen content, phosphorous content and pH) in the woodlands vary substantially (Chidumayo and Gumbo 2010). *Adansonia digitata* spans across both wet and dry miombo and mopane woodlands in its geographical distribution (Sidibe and Williams 2002). Hence it is most probable that the species has genetically diverged due to adaptation to varying ecological niches.

Despite important contribution of Non Timber Forest Products (NTFPs) to livelihood of thousands of rural households, sustainable utilisation is a challenge and forest degradation is high; estimated at annual deforestation rate of 2.6 to 2.8% (Government of Malawi 2011). Non timber forest products have been traditionally used by rural communities for subsistence and trade (Chirwa et al. 2008; Sidibe and Williams 2002). They are especially important for livelihood security in cash-poor households by ensuring food security, maintaining the traditional balance in people's diets, meeting medicinal needs and as a source of income (Schumann et al. 2010). It is estimated that 39 million people use Miombo woodlands to derive their livelihood (Makonda and Gillah 2007). According to Kalaba et al. (2009), more than 80% of the rural population in sub-Saharan Africa is poor and traditionally depend on natural non-wood goods and services. Kalaba et al. (2009) have reported that indigenous fruit trees from the wild can boost rural annual income by US$300-2000 per house hold. Akinnifesi et al. (2008a) has reported that indigenous fruits from the miombo woodlands are central to the livelihood of both rural and urban dwellers in southern Africa especially during the time of famine and food scarcity. Poorer and more vulnerable households, such as women tend to be more dependent on forestry resources than those households with access to other forms of income (Shackleton et al. 2002). Akinnifesi et al. (2008a) have presented that the Miombo woodlands contain over 200 tree species that bear edible fruits rich in minerals and vitamins and sold for cash income. Studies have also revealed that indigenous fruits contribute about 42% of the natural food basket in southern Africa and between 26 and 50% of households have reported reduced vulnerability during famine by collecting fruits from the Miombo woodlands. Mithofer et al. (2006) in Zimbabwe showed that indigenous fruit collection, consumption and sale reduce income poverty by 33%. The income from fruits usually comes at critical time when income from other sources is low and provide nutrition and food when agricultural labour is high (Akinnifesi et al. 2008a). Assessment on fruit enterprises conducted in Malawi, Zimbabwe and Tanzania showed a profit margin of 15-28% and provided women groups with a 40% of the market share and an average internal rate of return of 34% (Akinnifesi et al. 2008a). Shackleton et al. (2007) have reported net incomes between US$16 and US$160 per week from selling fresh edible NTFPs during the season and US$216 and US$166 per month from palm wine, well above the gross national product per capita of the Democratic Republic of Congo. The authors also reported that sales from the most popular fruit species, amounted to over US$4 million in one year. In southern Africa, *Sclerocarya birrea* fruits are used to make Amarula cream liqueur whilst in Zambia it is used to make Marula wine; in United Kingdom, Afreek beer is made from Amarula and oil extracted from the kernels is valued in South Africa for cosmetic industries (Chirwa et al. 2007). Currently, wine of export quality is produced in Zambia from *Ziziphus mauritian* and about 4000 rural women have been trained in processing of indigenous fruits into juices, jams, and wine in Malawi, Tanzania, Zambia and Zimbabwe (Akinnifesi et al. 2006). Akinnifesi et al. (2008b) reported sales of *Sclerocarya birrea* fruits amounting to US$180,000 annually for a community in South Africa and the Southern African Natural Products Trade Association (Phytotrade) made gross revenue of US$629,5000 from the sale of *Sclerocarya birrea* (US$126,420), *Ximenia caffra* (US$44,120), *Adansonia digitata* (US$22,250) and *Kigelia* spp (US$20,000). Ramaldhani and Schmidt (2008) reported that indigenous fruits were first freely consumed but with time local peasants started business to subsidies their income selling *Uapaka kirkiana*, *Strychnos cocculoides*, *Lannea edulis*, *Ziziphus Mauritian*, *Azanza garkeana*, *Adansonia digitata*, *Vitex mombasse* and *Berchemia discolor*. Then production and market chain grew to include vendors, wholesalers, retailers and consumers in urban areas. However, for a long period indigenous fruits have been neglected due to: lack of information and reliable methods for measuring their contribution to rural economies, livelihoods of communities, and ecological services; low production incentives relating to markets and technologies; bias in favour of large-scale agriculture and conventional forestry. Additionally, colonial interventions also left a profound legacy of neglect of smallholder farm production in favour of estate producers; and a weak interface between private sector actors, researchers and extensionists in tree products (Akinnifesi et al. 2008c). Makonda and Gillah (2007) have also presented that fruit trees that have potential to provide food security and alleviate poverty and malnutrition have been overlooked by science for a very long time and their potential left untapped.

Agroforestry has been identified as one of the strategies to introduce indigenous trees into cropping systems and impact on livelihoods of rural communities in sub-Saharan Africa (Akinnifesi et al. 2008a). For two decades, the World Agroforestry Centre (ICRAF) has spearheaded research and development activities on tree portifolios for the production of edible fruits in its indigenous tree domestication program in southern Africa (Akinnifesi et al. 2008a). Some of the ICRAF tree domestication initiative were focused on *Sclerocarya birrea, Uapaka Kirkiana, Strychnos cocculoides, Vangueria infausta, Parinari curatellifolia, Ziziphus mauritiana, Adansonia digitata, Syzigium cordatum* (Gaertner) and *Vites* species (Akinnifesi et al. 2006). This program has established international provenance trials for *Sclerocarya birrea* (Chirwa et al. 2007) and *Uapaka Kirkiana* (Akinnifesi et al. 2006). However, despite *Adansonia digitata* being one of the important species in the domestication programme, no clear research agenda existed in the region.

1.2 Socio-economic importance of Baobab

Adansonia digitata is one of the most important indigenous fruit trees valued in Africa for food, fiber and medicine (Assogbadjo et al. 2009; Kalinganire et al. 2008; Kyndt et al. 2009; Raebild et al. 2011). Baobab products are sold in informal markets forming important sources of income to many rural communities (Sidibe and Williams 2002). Recently, pulp products and seed oil have been exported to Europe, Canada and the USA (Gruenwald and Galiza 2005; Venter and Witkowski 2011). PhytoTrade collects baobab products especially pulp from Southern Africa for export to international markets (Akinnifesi et al. 2008a). Commercialization of the products means that many households in Africa have their livelihood improved due to injection of income. For example, in Zimbabwe baobab trade (fruit) has increased the income of rural people by 250% (Venter and Withowski 2011). In addition, the authors have reported that income from the sale of baobab fruits in the northern provinces of South Africa benefit over 1500 people for whom it is part of livelihood strategy.

The baobab pulp are effective prebiotic ingredients with anti-oxidant and anti-inflammatory properties, that has made it valuable for a variety of applications in beverages used in making soft drinks (cool and hot drinks), milk drinks, ice drinks and natural fruit smoothies (Gruenwald 2009). According to Sacandé et al. (2006), some of the modern baobab industrial products include condiments: functional food, cereal and energy bars, sport and energy drinks, vitamin and mineral supplements, diet supplements, high fiber products, energy strips, cereal additives, yogurt with prebiotic activities, fruit juices, smoothies, ice creams and shakes, powdered shakes and mixtures, cakes and biscuits. Whilst for medicine, anti-constipation preparations, anti-fever products and chewable tablets have been derived. Manufactured cosmetic products include shower and non-shower products (soaps, gels, face-lift creams, body moisturizers, shampoos, hair conditioners). Herbal tea is prepared from baobab seed. Pulp is rich in pectins and have high vitamin C content of 169 mg $100g^{-1}$; tenfold greater than that of orange (Leak 1999). Seed kernels contain 12-15% edible oil with more protein than groundnuts (Leak 1999) and rich in about 20 amino acids, iron, calcium, magnesium, manganese, zinc, sodium and phosphorous

(Sidibe and Williams 2002) (Table 1.1). The young tender leaves of baobab are used green or as dried vegetable, rich in vitamin A and calcium`(Leak 1999; Sidibe and Williams 2002). Baobab pulp and seed in Malawi have also been reported to be rich in nutritional values (Saka et al. 2008) (see Table 1.1). In West Africa the leaves, shoots and fruits are used as fodder (Sacandé et al. 2006).

Table 1. 1 Selected nutritional attributes of different plant parts of baobabs in Malawi and Burkina Faso

Plant part	Energy	Fat	Prot	carb.	Minerals (µgg^{-1})							
	(kJ 100g^{-1})	(%)	(%)	(%)	P	Ca	Mg	Fe	K	Na	VC	VA
Pulp	1480	4.3	3.1	79.4	450	1156	2090	58	28364	188	179.1	21
Seed	-	29.6	28.7	25.4	5.8	456	-	0.5	1186	75.2	-	-
Seed*					6140	3950	3520	18.3		19.6		
Leaves*					3020	20000	5490		1630			

Carb. = Total carbohydrate, Prot = protein, VC = vitamin C, VA = vitamin A, P= phosphorus, C = calcium, Mg = magnesium, Fe = Iron, K = potassium, Na = sodium,

Source: Saka et al. (2008), * Source: Sidibe and Williams (2002

1.3 Problem Statement

Baobab (see Fig.1.1) belong to the family Bombacaceae and the genus *Adansonia* (Assogbadjo et al. 2006). Pocktsy et al. (2009) and Kyndt et al. (2009) have taxonomically grouped *A. digitata* in Malvaceae family. The baobab are comprised of eight species in the genus *Adansonia* with *A. digitata* occurring in hot drier regions of tropical Africa extending from northern South Africa and Namibia to Ethiopia, Sudan and the southern fringes of the Sahara (Gebauer et al. 2002). Detailed distribution map of the species is presented by Sidibe and Williams (2002). *A. digitata* is a deciduous, tropical tree which grows in arid, semi-arid and sub-humid tropical climates (ICUC 2002). The ideal growing environment for baobab is at altitudes of between 450 to 600 m above sea level with rainfall between 300 and 500 mm per annum (Wickens 1982). Gebauer et al. (2002) have reported the species growing in zones with 100 to 1000 mm annual rainfall. Baobab has also been recorded from sea level to at least 1500 m and from areas receiving, annually, as little as 90 mm to as much as 1400 mm; although such distribution has been facilitated by man (Edkins et al. 2007; Wilson 1988). The species tolerates minimum temperature of 5 ^{o}C to maximum temperature of 40 ^{o}C (SCUC 2006). The tree can adapt to various soils types with varying soil moisture (Edkins et al. 2007; Gebauer et al. 2002). The wide ecological adaptation of the species might imply that *Adansonia digitata* has evolved wide genetic diversity in its geographical distribution area. This genetic diversity is presently not well known in southern Africa (Ræbild et al. 2011).

The mating system influences the effective population size and opportunities for gene flow and consequently the genetic diversity and differentiation and ultimately evolution of the species (Eckert et al. 2009). In addition, information on reproductive ecology and factors influencing reproductive success of a species is highly important for managing the species such as sustainable harvesting to maintain the regeneration capability (Soehartono and Newton 2001). Baobab is hermaphrodiet (having both male and female parts in the same flower) with bat pollination as a major pollination agent even though wind and insect pollination have also been mentioned as possible agents (Assogbadjo et al. 2006; Gebauer et al. 2002; Sidibe and Williams 2002). Gibson (2009) has reported that bat pollination does not occur in South Africa and yet baobab in this region fruit (Venter and Witkoski 2011); implying that there are other pollination agents in this locality not yet known. Sidibe and Williams (2002) also reported that sour scent of baobab flowers attract ants, nocturnal moths and species of bollworms that might effect some pollination. This further implies that apart from daytime flower visitors, there are other nocturnal animals effecting pollination in baobab as well. It has also been known that some species such as *Ceiba pentandra* in Bombacaceae family have different mating systems depending on ecological conditions of the sites separated only by about 600 km (Lobo et al. 2005). This shows the danger of generalizing the pollination agents for baobab across its geographical range. Therefore, there is need for domestication programmes in southern Africa to understand the reproductive system of baobab.

Several factors might have structured the baobab genetic structure over time. It is possible mutation, migration and random genetic drift have induced variation in allelic frequencies in tree populations (Yea 2000; Tamarin and Leavit. 1991). Following extensive geographical distribution, it is most likely that baobab populations (individual trees) located in different ecological habitats have differentiated into races or ecotypes through natural selection (Bizoux and Mahy 2007; Sreekumar and Renuka 2006; Zeneli et al. 2005). Habitat fragmentation might have affected baobab genetic diversity due to alteration in the landscape features which tends to reduce gene flow (Pautasso 2009). Reproductive patterns in a population is known to determine the genetic diversity of the future generations (Morgante et al. 1991). Within baobab populations, the proportion of adults that reproduce within a season or even the degree of synchrony in flowering time vary affecting genetic structure of the populations (Sidibe and Williams 2002; Murawski and Hamrick 1991). Furthermore, due to anthropogenic factors, non-random outcrossing on farm lands may also affect the outcome of the baobab mating system; hence altering the genetic outcome of the population (Murawski et al. 1990). In addition, factors such as extent of seed and pollen dispersal, tree density, colonization history, differential mortality and micro-environmental selection have impacted on baobab spatial genetic structuring over time (Kelly et al. 2004). Animals such as elephants, baboons and water disperse baobab fruits (Wickens 1982); likely altering the allelic and genotypic frequencies of populations. In addition, Wilson (1988) has attributed death of many baobab trees due to persistent drought and fungal diseases and elephant damage (Edkins et al. 2007), which have also structured the genetic composition of the populations. Thus current understanding of the degree and pattern of genetic diversity is pertinent for the sustainability of baobab productivity.

There is need to understand the pattern of genetic diversity in baobab across it geographical range (Ræbild et al. 2011; Sidibe and Williams 2002). The understanding of the current diversity status of baobab genetic resource is a prime step for developing efficient domestication, tree improvement and conservation programmes (Sreekumar and Renuka 2006). It is known that tree improvement programmes can greatly affect genetic variability in subsequent forest populations (Pakkad et al. 2008) especially with the use of clonal propagation to reduce precosity of the species ranging from 9 to 125 years (Gebauer et al. 2002; Gruenwald and Galiza 2005; Sacandé et al. 2006; Sidibe and Williams 2002; Venter and Witkowski 2011) to 3 or 4 years (SCUC 2006). Genetic diversity (diversity of genes and genotypes) is important since it is the foundation for genetic change (improvement) and the primary factor allowing populations to adapt to environmental changes (Kitzmiller 1990; Pautasso 2009). Pluess and Stöcklin (2004) have presented that less diverse populations have a reduced ability to buffer the effects of poor environmental conditions or competition, which may be the case with baobab in southern Africa. Loss of genetic variability may lead to species extinction especially in fragmented small populations following genetic drift (Pautasso 2009). Studies in gene flow serve the dual purpose of providing insight into the dispersal processes that shape the genetic structure of the baobab and foraging behavior of the pollinators (Larsen et al. 2009). In order to determine genetic variation, several methods are available: morphological markers, isoenzymes, random amplified polymorphic DNAs (RAPDs), nuclear sequence-tagged sites (STS), cpDNA microsatellites, mtDNA microsatellites, nuclear simple sequence repeats (SSRs or microsatellites) (Maghuly et al. 2006). In the current study, microsatellite markers have been used. Microsatellite markers are short tandem repeats of mono-to tetra-nucleotide repeats, which are assumed to be randomly distributed in the nuclear genome usually such repeats are relatively abundant and have high mutation rates in comparison to other markers, which make them applicable for various types of population studies (Zolgharnein et al. 2010). In addition, nuclear and chloroplast microsatellite markers have found use in determining genetic structure, differentiation and gene flow pattern within and among tree populations (Pakkad et al. 2008). According to Giang et al. (2003), microsatellites as a co-dominant genetic marker enables detection of both homozygotes and heterozygotes that improves the insufficiency of AFLP and could be used for examining mating system. In addition, as few as five or six microsatellite loci can answer many conservation genetic questions (Glaubitz and Moran 2000).

A lot of research on baobab focusing on morphometric and molecular variation has been done in West Africa (Assogbadjo et al. 2006, 2009; Kyndt et al. 2009) which have revealed important information for the management of their populations in the wild and the semi-domesticated trees in the parkland. None of this research has been replicated in the whole of southern Africa even though baobab is at present promoted for growing by small holder farmers. Knowledge in genetic diversity is crucial in the choice of seed source, tree conservation and tree improvement. Furthermore, the knowledge in reproductive biology has so far not covered the whole baobab distribution range (Ræbild et al. 2011; Sidibe and Williams 2002).

The documentation of traditional knowledge on plant use can generate valuable information that can be used for commercialization (Kristensen and Lyke, 2003). Inconsistency of product quality and limited access to markets have been identified as key problems to commercialization of the non-timber forest products (Jama et al. 2008). Traditional knowledge is passed down from generation to generation in verbal form (Lin 2005) which can be lost if not documented. Presently, there is no full ethnobotanical and marketing information on baobab at national and regional level in southern Africa. The information is fundamental for optimizing use of the species for the upliftment of the livelihood of many rural communities. In addition, the knowledge is pertinent for policy makers to draft policies that are conducive for the development of the enterprise.

The current study was undertaken to assess use, reproductive ecology and genetic diversity of baobab in Malawi with an aim of promoting development of novel baobab products for commercialization and establishment of a healthy resource base that should sustainably supply products.

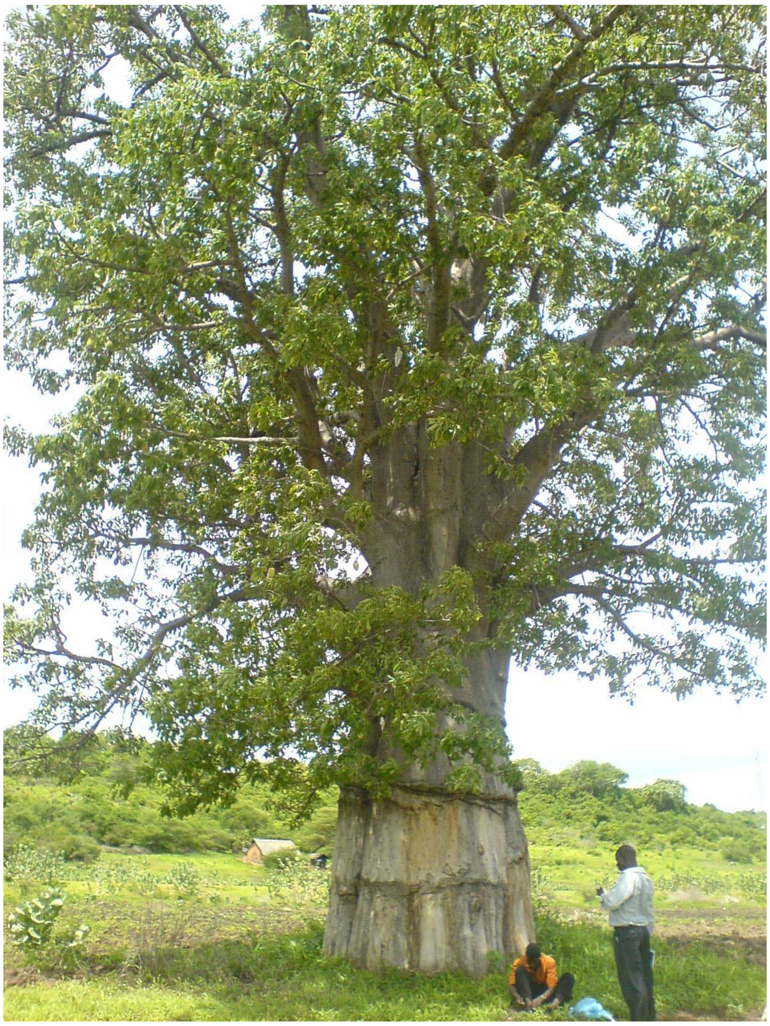

Fig. 1. 1 Baobab tree with signs of bark extraction used for fibre in Karonga

1.4 Conceptual framework of the study

The present study strived to develop an understanding of use, physiology and characterization of *Adansonia digitata* in Malawi. In the diagrammatic concept (Fig.1.2), the base resource is the wild population, the sole supplier of baobab products upon which the livelihood of many Malawians depend. The morphometric (phenotypic) variation is influenced largely by the genotype (both additive and non-additive genetic variances), environment and interaction of genotype and environment. Additive genetic variance is the component that is mostly used in tree breeding manipulation. Thus it has to be determined in perceived successful tree planting programmes. In addition, the knowledge in reproductive biology is important as it influences the gene flow in the population, thus determines the genetic diversity and population differentiation. Furthermore, it affects the level of phenotypic variation of fruits existing between and within wild populations. Any disturbance on the reproductive biology will have a direct effect on many rural livelihoods. Improvement of productivity is important for the resource to sustain the ever increasing population demand. The process manipulates the genetic diversity by concentrating superior genes. This narrows the genetic diversity. Knowledge in the extent and pattern of molecular variation will ensure that there is broad genetic base in germplasm for afforestation operations. Nursery or greenhouse studies have been employed in plantation forestry to quantify genetic variation, population structuring, identifying superior genotypes and estimating the proportion of additive genetic variance (narrow sense heritability) for physiological traits with high precision. This has reduced time lag during classical tree breeding. Modern science has developed molecular study tools which are able to determine the actual allelic and genotypic frequencies in the population. The tools enable correct prediction of genetic diversity, population structuring and mating system in the species. The results find use in proper management of both wild and planted tree populations, conservation and tree improvement strategies. It also helps in species-site matching for optimum tree productivity. Conducive government regulations and policies are required to provide enabling conditions to enhance growth of the enterprise and resource base either through domestication and improved land and tree tenure. The processes are highly interlinked such that it was felt rational in this study to concurrently research on them in five specific studies.

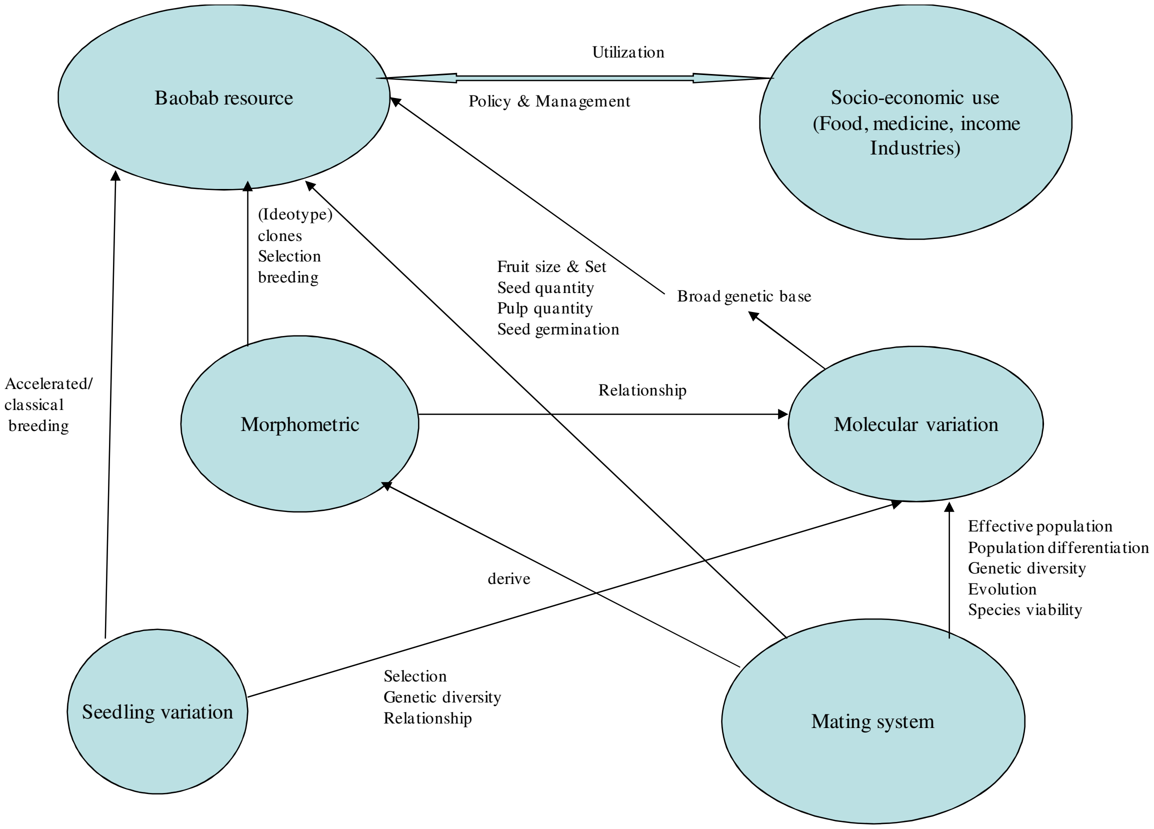

Fig. 1. 2 Linkage of socio-economic, reproductive biology, morphometric and genetic variation of *Adansonia digitata* in Malawi

1.5 Objectives of the study

The overall objective of the study was to develop an understanding of the contribution of baobab tree to the livelihood of rural communities, morphometric and genetic diversity and population differentiation prevailing in the species and its reproductive biology that can help in domestication, commercialization, tree improvement, conservation and tree management that can sustain tree productivity under agroforestry system.

1.6 Specific objectives

Five specific objectives were formulated with related hypotheses.

Specific objective 1: To assess and document traditional knowledge on potential social use of baobab tree and marketing of its products.

Related hypothesis

> Marketing of baobab products has the potential to alleviate poverty of many households in rural communities in Malawi.

Specific objective 2: To assess phenotypic variation in fruit and seed morphology between and within natural populations growing in different ecological zones based on climatic and edaphic factors and investigate possibility of using morphometric traits to delineate populations into distinct races (ecotypes).

Related hypotheses

(i) High morphometric variation exists in fruit and seed traits in five populations occurring in four different silvicultural zones.

(ii) Fruit and individual seed traits can be used to characterize populations from four silvicultural zones into vivid races/ecotypes.

Specific objective 3: To characterize, quantify the genetic variation and estimate the genetic control of *A. digitata* seed germination and seedling characteristics in five natural populations existing in four diverse ecological conditions using nursery study.

Related hypotheses

1. Seedlings traits could be use to determine genetic diversity and population differentiation of the five populations from the four silvicultural zones.

2. Seedling traits are under strong additive gene effects providing possibility of early selection of superior genotypes and achieving high genetic gains.

Specific objective 4: To examine whether the delineation of silvicultural zones have subsequently structured the genetic composition of the trees substantially and also to assess the gene flow among the five populations using microsatellite markers.

Related hypotheses

(i) High genetic diversity prevails in natural populations growing in four different eco-regions important for domestication, tree improvement and conservation.

(ii) Baobab populations are highly genetically structured (differentiated) due to environmental adaptation, geographical isolation and extent of gene flow.

Specific objective 5: To investigate the reproductive biology of baobab especially the mating system and fruit and seed physiology.

Related hypotheses

(i) The Baobab species has mixed mating systems wind, bats, insects and not exclusively bat pollinated.

(ii) Fruit and seed development varies from tree to tree.

1.7 Thesis structure

The thesis is divided into seven chapters. Chapter one describes the background of the study, objectives and hypotheses of the study. The other chapters (Chapter two to six) deal with specific components to address outlined objectives: (2) Socio-Economic value of *Adansonia digitata* L. (Baobab) products in Malawi; (3) Phenotypic variation in fruit and seed morphology of *Adansonia digitata* (Baobab) in five selected wild populations in Malawi; (4) Genetic variation among and within provenances of *Adansonia digitata* (Baobab) in seed germination and seedling growth from selected natural populations in Malawi; (5) Genetic differentiation and diversity of *Adansonia digitata* (Baobab) in Malawi using microsatellite markers; and (6) Reproductive biology of *Adansonia digitata* (Bombacaceae) in Malawi, southern Africa. Chapter 7, the last and final chapter entitled: *Adansonia digitata* (Baobab) as a sustainable health natural resource: Linking reproductive biology, genetic variation and socio-economic use, presents a synthesis of the findings and management options to sustain supply and production of baobab products in southern Africa. Chapters two to six have been submitted to Agroforestry Systems journal since the issues raised in the study mostly address potential domestication and utilisation of the species as a horticultural crop in an integrated way (agroforestry) with other agricultural crops in the parkland.

1.8 References

Akinnifesi FK, Sileshi G, Ajayi OC, Chirwa PW, Kwesiga FR, Harawa R (2008a) Contributions of Agroforestry Research and development to Livelihood of Smallholder Farmers in Southern Africa:2. Fruit, Medicinal, Fuelwood and Fodder Tree Systems. Agricultural Journal 3 (1): 76-88

Akinnifesi FK, Ajayi OC, Sileshi G, Matakala P, Kwesiga FR, Ham C, Kadzere I, Mhango J, Mng'omba, Chilanga T, Mkonda A (2008b) Creating opportunities for Domestication and Commercializing Miombo Indigenous Fruit Trees in Southern Africa. In: Akinnifesi FK, Leakey RRB, Ajayi OC, Sileshi G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds). Indigenous Fruit Trees in the Tropics. Domestication, Utilisation and Commercialization. CABI, Wallingford UK, pp137-170

Akinnifesi FK, Silesh G, Ajayi OC, Tchoundjeu Z (2008c) Accelerated Domestication and Commercialisation of Indigenous Fruit and Nut Trees to Enhance Better Livelihoods in the Tropics: Lessons and Way Forward. In: Akinnifesi FK, Leakey RRB, Ajayi OC, Sileshi G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds). Indigenous Fruit Trees in the Tropics. Domestication, Utilisation and Commercialization. CABI, Wallingford UK, pp392-427

Akinnifesi FK, Kwesiga F, Mhango J, Chilanga T, Mkonda A, Kadu CAC, Kadzere I, Mithofer D, Saka JDK, Sileshi G, Ramadhani T, Dhliwayo P (2006) Towards the development of Miombo Fruit Trees as Commercial Tree Crops in Southern Africa. Forests, Trees and Livelihoods, 16: 103-121

Andersen US, Córdova PJP, Nielsen UB, Kollmann J (2008) Provenance variation in germination and seedling growth of *Abies guatemalensis* Rehder. Forest Ecology and Management 255: 1831-1840

Assogbadjo AE, Kyndt T, Sinsin B, Gheysen G, Van Damme P (2006) Patterns of Genetic and Morphometric Diversity in Baobab (*Adansonia digitata*) Populations Across Different Climatic Zones of Benin (West Africa). Annals of Botany 97: 819-830

Assogbadjo AE, Kyndt T, Chadare FJ, Sinsin B, Gheysen G, Eyog-Matig O, Van Damme P (2009) Genetic fingerprinting using AFLP cannot distinguish traditionally classified baobab morphotypes. Agroforestry Systems, 75: 157-165

Bizoux J, Mahy G (2007) Within-population genetic structure and clonal diversity of a threatened endemic metallophyte, *Viola calaminaria* (Violaceae). American Journal of Botany 94(5): 887-895

Chidumayo EN, Gumbo DJ (eds.) (2010) The Dry Forests and Woodlands of Africa:Managing for products and Services. Mapset Ltd, Gateshead, UK

Chirwa PW, Bwanali RJ, Meke G, Sagona W, Munthali CRY, Mwabumba L (2007). Growth and Phenology of a three-to-four –year –old Sclerocarya birrea international provenance trial in Malawi. Southern Hemisphere Forestry Journal 69(1):49-54

Chirwa PW, Syampungani S, Geldenhuys CJ (2008) The ecology and management of the Miombo woodlands for sustainable livelihoods in southern Africa: the case for non-timber forest products. Southern Forests 70(3): 237-245

Dawson IK, Lengkeek A, Weber JC, Jamnaclass R (2009) Managing genetic variation in tropical trees: Linking knowledge with action in agroforestry ecosystems for improved conservation and enhanced livelihoods. Biodivers Conserv 18: 969-986

Eckert CG, Kalisz S, Geber MA, Sargent R, Elle E, Cheptou P, Goodwillie C, Johnston MO, Kelley JK, Moeller DA, Porcher E, Ree RH, Vallejo-Marlin M, Winn AA (2009) Plant mating system in a changing world. Tree 1151. Doi:10.1016/j.tree.2009.06.013

Edkins MT, Kruger LM, Harris K, Midgley JJ (2007) Baobabs and elephants in Kruger National Park: nowhere to hide. African Journal of Ecology. Doi:10.1111/j.1365-2028.2007.00798.x

Gebauer, J, El-Siddig and Ebert, (2002) Baobab (*Adansonia digitata* L.): a Review on a multipurpose Tree with Promising future in the Sudan. Gartenbauwissenschaft, 67(4): 155-160

Gibson AR (2009) Bats and Their Flowers. http://www.botgard.ucla.edu/html/membgnewsletter/volume4number4/Batsandtheirfl. Site visited on 09/11/2009

Giang LH, Hong PN, Tuan MS, Harada K (2003) Genetic variation of *Avicennia marina* (Forsk) Vierh. (Avicenniaceae) in Vietnam revealed by microsatellite and AFLP markers. Genes Genet. Syst. 78: 399-407

Glaubitz JF, and Moran GF (2000) Genetic Tools: The Use of Biochemical and Molecular Markers. In: Young A, Boshier, D and Boyle T (eds.), Forest Conservation Genetics. Principles and Practice. CSIRO Publishing, United Kingdom

Gruenwald J (2009) Novel botanical ingredients for beverages. Clinics in Dermatology 27: 210-216

Gruenwald J, Galiza MI (2005) *Adansonia digitata* L. Baobab. Market Brief in the European Union for selected natural ingredients derived from native species. The United Nations Conference on BioTrade Facilitation Programme (BTFP)

Hardcastle PD (1978) A preliminary silvicultural classification of Malawi. Forestry Research Record No. 57. Government Printer, Zomba, Malawi.

International Centre for Underutilized Crops (ICUC) (2002) Fruits for the Future. Baobab. Factsheet No.4. March, 2002. ICUC, Southampton, UK

Jama BA, Mohamed AM, Mulatya J, Njui AN (2008) Comparing the “Big Five”: A framework for the sustainable management of indigenous fruit trees in the dry lands of East and Central Africa. Ecological Indicators 8: 170-179

Kalinganire A, Weber JC, Uwamariya A, Kone B (2008) Improving Rural Livelihoods through Domestication of Indigenous Fruit Trees in the Parklands of the Sahel. In: Akinnifesi FK, Leakey RRB, Ajayi OC, Sileshi G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds). Indigenous Fruit Trees in the Tropics. Domestication, Utilisation and Commercialization. CABI, Wallingford UK, pp 186-203

Kaushik N, Kumar K, Kumar S, Kaushik N, Roy S (2007) Genetic variability and divergence studies in seed traits and oil content of Jatropha (*Jatropha curcas* L.) accessions. Biomass and Bioenergy 31: 297-502

Kalaba FK, Chirwa PW, Syampungani S, Ajayi OC (2010) Contribution of agroforestry to biodiversity and livelihoods improvement in rural communities of Southern African regions. In: Teja Tscharntke, Christoph Leuschner, Edzo Veldkamp, Heiko Faust, Edi Guhardja, Arifuddin Bidin (Eds.).Tropical rainforests and agroforests under global change. Springer Berlin, pp 461-476

Kelly BA, Hardy OJ, Bouvet J (2004) Temporal and spatial genetic structure in *Vitellaria paradoxa* (shea tree) in an agroforestry system in southern Mali. Molecular Ecology 13: 1231-1240

Kitzmiller JH (1990) Managing Genetic Diversity in a Tree Improvement Programme. Forest Ecology and Management, 35: 131-149

Kitzmiller JH (1990) Managing Genetic Diversity in a Tree Improvement Programme. Forest Ecology and Management 35: 131-149

Kristensen M, Lykke AM (2003) Informant-Based Valuation of Use and Conservation Preferences of Savanna Trees in Burkina Faso. Economic Botany 57(2): 203-217

Kyndt T, Assogbadjo AE, Hardy, OJ, Kakaïa RG, Sinsin, B, Van Damme P, Gheysen G (2009) Spatial genetic structuring of baobab (*Adansonia digitata*, Malvaceae) in the traditional Agroforestry systems of West Africa. American Journal of Botany 96(5):950-957

Larsen AS, Vaillant A, Verhaegen D, Kjaer ED (2009) Eighteen SSR-primers for tetreploid Adansonia digitata and its relatives. Conservation Genet Resource. Doi 10.1007/s12686-009-9075-y

Leakey RRB (1999) Potential for novel food products from agroforestry trees: a review. Food Chemistry 66:1-4

Lin KW (2005) Ethnobotanical study of medicinal plants used by the Jah Hut people in Malaysia. Indian Journal of Medical Sciences, 59(4):156-161

Lobo JA, Quesada M, Stoner KE (2005) Effects of pollination by bats on the mating system of Ceiba pentandra (Bombacaceae) populations in two Tropical Life Zones in Costa Rica. American Journal of Botany 92(2):370-376

Maghuly F, Pinsker W, Praznik W, Flush S (2006) Genetic diversity in managed subpopulations of Norway spruce [Picea abies (L.) Karst. Forest Ecology and Management 222: 266-271

Malawi Government (2000) Malawi's National Forestry Programme. Priorities For Improving Forestry and Livelihoods. Ministry of Mines, Natural Resources and Environment, Department of Forestry, Lilongwe, Malawi

Malawi Government (2005) National Biodiversity Strategy and Action Plan. The Government Printers, Lilongwe, Malawi

Malawi Government (2009) Malawi Biomass Energy Strategy, Ministry of Mines, Natural Resources and Environment, Lilongwe

Malawi Government (2011) Economic Analysis of Sustainable Natural Resource Use in Malawi: Economic Study. Ministry of Economic Planning and Development, Lilongwe

Makonda FBS, Gillah PR (2007) Balancing Wood and Non –Wood Products in Miombo Woodlands. MITMIOMBO-Management of Indigenous Tree Species for Ecosystem Restoration and Wood Production in Semi-Arid Woodlands in Eastern Africa. Proceedings of the first MITMIOMBO Project Work-shop held in Morogoro, Tanzania, 6th-12th February 2007

Mauambeta DDC, Chitedze D, Mumba R (2010) Status of Forests and Tree Management in Malawi. A position paper prepared for Coordination Union for Rehabilitation of the Environment (CURE)

Mithofer D, Waibel H (2008) Economics of On-farm Production of Indigenous fruits. In: Akinnifesi FK, Leakey RRB, Ajayi OC, Sileshi G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds). Indigenous Fruit Trees in the Tropics. Domestication, Utilisation and Commercialization. CABI, Wallingford, UK, pp 237-272

Morgante M, Vendramin M, Olivier AM (1991) Mating system analysis in *Pinus leucordermis* Aut: detection of self-fertilization in natural populations. Heredity 67: 197-203

Murawski DA and Hamrick JL (1991) The effect of the density of flowering individuals on the mating systems of nine tropical tree species. Heredity: 167-174

Murawski DA, Hamrick JL, Hubbel SP, Foster RB (1990) Mating system of two Bombacaceous trees of a neotropical moist forest. Oecologia 82: 501-506

Pakkad G, Ueno S, Yoshimaru H (2008) Genetic diversity and differentiation of Quercus semiserrata Roxb. In northern Thailand revealed by nuclear and chloroplast microsatellite markers. Forest Ecology and Management 255: 1067-1077

Pautasso M (2009) Geographical genetics and the conservation of forest trees. Perspective in Plant Ecology, Evolution and Systematics 11: 157-189

Plues AR, Stöcklin J (2004) Population genetic diversity of the clonal plant Geum reptans (Rosaceae) in the swiss Alps. American Journal of Botany 91(12): 2013-2021

Pocktsy JL, Lumaret R, Mayne D, Mohamed VA, Abutaba YIM, Sagna M, Raoseta S, Danthu P (2009) Chloroplast DNA phylogeography suggest a West African centre of origin for the baobab, *Adansonia digitata* L (Bombacoideae, Malvaceae). Molecular Ecology 18: 1707-1715

Raebild A, Larsen AS, Jensen JS, Ouedraogo M, De Groote S, Van Damme P, Bayala J, Diallo BO, Kalinganire HAS, Kjaer ED (2011) Advances in domestication of indigenous fruit trees in the West African Sahel. New Forests 41: 297-315

Ramadhani T, Schmidt E (2008) Marketing of Indigenous Fruits in Southern Africa. In: Akinnifesi, F.K, Leakey, R.R.B, Ajay, O.C, Sileshi, Tchoundjeu, Z, Matakala, P and Kwesiga, F.R (eds). Indigenous Fruit Trees in the Tropics. Domestication, Utilisation and Commercialization, CABI, Wallingford UK, pp224-236

Sacandé M, Rønne C, Sanon MD, Joker D (2006) *Adansonia digitata* L. Seed Leaflet No. 109. Hørsholm: Forest & Landscape Denmark

Saka JDK, Kadzere I, Ndabikunze BK, Akinnifesi FK, Tiisekwa BPM (2008) Product Development Nutritional Value, Processing and Utilization of Indigenous Fruits from the Miombo Ecosystem. In: Akinnifesi, F.K, Leakey, R.R.B, Ajay, O.C, Sileshi, Tchoundjeu, Z,

Schumann K, Wittig R, Thiombiano A, Becker U, Hahn K (2010) Impact of land-use type –and bark – and leaf-harvesting on population structure and fruit production of the baobab tree (*Adansonia digitata* L.) in semi-arid Savanna, West Africa. Forest Ecology and Management 260: 2035-2044

SCUC (2006) Baobab Manual, Field Manual for Extension Workers and Farmers, University of Southampton, UK

Sidibe M, Williams JT (2002) Baobab. *Adansonia digitata*. International Centre for Underutilized Crops, Southampton, UK

Shackleton S, Shanley P, Ndoye O (2007) Invisible but viable: recognizing local markets for non-timber forest products. International Forestry Review 9(3): 697-713

Shackleton SE, Shackleton CM, Netshiluvhi BS, Balance A, Fairbanks DHK (2002) Use Patterns and Value of Savanna Resources in Three Rural Villages in South Africa. Economic Botany 56(2): 130-146

Soehartono T, Newton AC (2001) Reproductive ecology of Aquilaria spp. in Indonesia. Forest Ecology and Management 152: 59-71

Tamarin R, Leavit RW (1991) Principles of Genetics (Third Edition). WM.C. Brown publishers, Dubuque

Venter SM, Witkowski ETF (2011) Baobab (*Adansonia digitata* L) fruit production in communal and conservation land-use types in Southern Africa. Forest Ecology and Management 261: 630-639

Wickens GE (1982) The Baobab-Africa's upside-down tree. Kew Bulletin, Vol. 37, No.2: 173-209

Wilson RT (1988) Vital Statistics of the baobab (*Adansonia digitata*). African Journal of Ecology , Volume 26: 197-206

White, E (1983). The Vegetation of Africa. UNESCO, Paris.

Wright JW (1976) Introduction to Forest Genetics. Academic Press, New York

Yeh FC (2000) Population genetics. In: Young A, Boshier D and Boyle T (eds.), Forest Conservation Genetics. Principles and Practice. CSIRO Publishing, UK

Zeneli G, Kola H, Dida M (2005) Phenotypic variation in native walnut populations of Northern Albinia. Scientia Horticulture 105: 91-100

Zobel B, Talbert J (1984) Applied Forestry Tree Improvement. John Wiley & Sons, New York. U.S.A

Zolgharnein H, Kamyab M, Keyvanshokooh S, Ghasemi A, Nabavi SMB (2010) Genetic Diversity of *Avicennia marina* (Forsk.) Vierh. Populations in the Persian Gulf by Microsatellite Markers. Journal of Fisheries and Aquatic Sciences 5(3): 223-229

2 Socio-economic value of *Adansonia digitata* (Baobab) products in Malawi

Abstract

Adansonia digitata (Baobab) is among the important indigenous fruit tree species identified for domestication and commercialization in southern Africa. Trade in baobab products has recently increased both locally, regionally and internationally. A socio-economic study was undertaken in four districts of Malawi including one in an urban setting in order to determine traditional use of the tree and marketing of baobab products. Targeted and random sampling methods were employed to interview people involved in baobab use and trade using a structured questionnaire. The study revealed significant differences in utilization of tree components and most of the factors influencing the market chain including: type of products sold, type of buyers, average price, and pricing dynamics. The majority of respondents valued fruits as food (75%) and for sale (54.8%). Leaves, bark and roots were largely used for medicinal purposes. Important value addition was production of Mlambe juice (42%), lolly-pop freezes or ice drinks (13.3 %) and jam (2.4%). Revenue accrued per month per person ranged from 2.5 to 715 USD; while cottage industries made between 1100 and 2300 USD; with net profits of about 595 USD per person and 1535 USD from the enterprise. The study showed that baobab tree is critical for rural livelihood despite its poor management. In addition the marketing of *Adansonia* products was predominantly an informal trade with limited commercialization occurring in the form of supermaketisation of 'Mlambe juice' from baobab that is predominantly in central and southern Malawi.

Key words: Baobab; Commercialization; Markets; Indigenous Fruits; Miombo

2.1 Introduction

Miombo woodland ecoregion is vast covering about 2.7 million square km and supporting 39 million people of which 15 million are urban inhabitants (Chirwa et al. 2008). Miombo woodlands are known to have over 75 indigenous edible fruit trees (Chirwa et al. 2008). Estimates of the number of people who rely on non-timber forest products (NTFPs) for at least part of their income could be up to 200 million worldwide (Chirwa et al. 2008). Shackleton et al. (2002) reported that 81% of the households in South Africa use wild fruits. Non-timber forest products provide guaranteed additional income for households that earn much of their income from agriculture and other off-farm sources (Akinnifesi et al. 2008). Kalaba et al. (2010) reported that harvesting of indigenous tree fruits from the wild increase rural annual income by US$300-US$2000 per household. Mithofer and Waibel (2008) reported that indigenous fruit contributes up to 30% of cash to household income. According to Akinnifesi et al. (2006), indigenous fruits contribute about 42% of the natural food basket for home consumption and marketing; enabling 26-50% of households to live above the poverty line during the critical famine periods. Kumar (2008) has further presented wild fruits as conferring important health benefits against malnutrition and improving resilience against many diseases. Saka et al. (2008) reported several marketable products from indigenous fruits such as jam, dried fruit, powder, ice-cream, butter, sweets, local alcohol beverage (kachasu), sweet wine, yoghurt, juice, porridge, coffee, sweet beer (thobwa) and food seasoning. Lowore (2006) reported that micro-enterprise based on forest produce are done either by individuals or small family business, usually starting with little or no capital, producing small quantities of mainly unprocessed goods with little profit. Shackleton et al. (2007) highlighted that trade in NTFP is constrained by factors such as underdevelopment, limited local buying power, inadequate infrastructure, poor exposure and access to markets, weak political power, high transportation costs, communication problems and high illiteracy levels of producers and traders. Furthermore, Chirwa et al. (2008) argue that little consideration has been given to the rate of harvesting and formulation of management systems that can sustain NTFP availability. Notwithstanding the issues highlighted, natural forest resources continue to play an important role in uplifting livelihood of rural communities in southern Africa (Kalaba et al. 2010). Economic restructuring in most developing countries has led to high cost of living forcing rural communities to rely on natural resources (Christian et al. 2008; Kalaba et al. 2010). Presently, miombo fruits are traded locally, regionally and internationally (Akinnifesi et al. 2008) in the form of alcoholic and non-alcoholic beverages, confectionaries, additives for other foods, dried whole fruits, oil and kernel butter. According to Akinnifesi et al. (2008), PhytoTrade Africa in southern Africa reported gross revenue of US$22,250 from the sale of baobab products. Sidibe and Williams (2002) have highlighted formal and informal trade in baobabs in cosmetics, craft products made from the fruit shell, baobab drink products, fresh and dried leaves, fruits and bark fiber products in West Africa. Presently, in Europe and USA, baobab fruit pulp is imported for use in different segments of industries such as food and beverages, botanical remedies and nutraceuticals and natural cosmetics (De Smedt et al. 2011; Gruenwald and Galiza, 2005). By 2003 estimates, the EU imported baobab products worthy 53 million Euros (18,341 tons) and exported 46 million Euros (13,299 tons) (Gruenwald and Galiza, 2005). Apart from pulp, an Italian baobab fruit Company operating in Senegal process leaves and seed into cosmetic products and is considering use of baobab tubers as food, just like carrot (Gruenwald and Galiza, 2005). PhytoTrade Africa in southern Africa also produces baobab seed oil, seed/press cake and baobab briquettes -a firewood substitute made from the fruit shells (Gruenwald and Galiza, 2005). Kristensen and Lykke (2003) in Burkina Faso have reported baobab tree as one of the top eight tree species of high commercial and nutritional value. In Malawi, baobab trees are also retained in the parkland signifying their importance to rural livelihoods. In West African Sahel, *A. digitata* is one of species preferred by farmers because of the nutritional, medicinal and income-generation through selling of predominantly fruit and leaf products (De Smedt et al. 2011; Dhillion and Gustad 2004; Kalinganire et al. 2008; Maranz et al. 2008).

Despite awareness on the contribution of baobab fruit trees for rural livelihood, information on socio-economic contribution in southern Africa is still incomplete (Ramadhani and Schmidt 2008). Indigenous fruit tree domestication, utilisation and commercialization are aimed at reducing poverty of the rural masses in southern Africa (Akinnifesi et al. 2008). However, this drive should be supported with socio-economic research in order to succeed. Additionally, while there is rich indigenous knowledge on use of baobab among the local communities in southern

Africa, this remains undocumented. The current study was therefore done to assess and document traditional knowledge on potential social use of baobab tree and marketing of its products in four districts and one urban area in Malawi.

2.2 Methodology

2.2.1 Socio-economic attributes

In order to establish the traditional knowledge on use of baobab tree, a survey was conducted in April, 2008 in four districts: Karonga, Salima, Mwanza and Chikwawa (Fig. 2.1). The sites were selected based on the silviculture zonation of Malawi (Hardcastle 1978). Secondly, sites selected had large population of trees and reputation of using the trees for livelihood based on information from District Forestry Officers. Purposive sampling was done in the choice of the villages and respondents as there was need to study specific cultural domain with knowledge experts within (Tongco, 2007). Targeted sampling was done at village level where only villages with baobab trees were sampled. Households within villages were randomly sampled. In order to obtain representative data, in each village, the Village Headman was first asked to give the number of registered family households. Interviews were then terminated only after a minimum of 20% households were interviewed in the village. A total of 219 respondents were sampled comprising of 104 males and 115 females. A structured questionnaire was used for interviews covering specific uses of leaves, fruits, bark, roots and wood.

2.2.2 Economic aspects

The data was collected in April and May, 2008 at the peak of fruiting season in three Districts of Chikwawa, Salima and Mwanza, and also Mzuzu City (Fig.2.1). The choice of the sites was based on availability and marketing of baobab products. Purposive sampling was done to collect information on marketing, whereby only respondents processing and found selling baobab products were interviewed (Tongco, 2007). Respondents were chosen purposively as they were too few to be sampled randomly; and not all members of the community had knowledge on the marketing aspects of the species. Places for interviews included roadsides, local markets, premises of cottage industry companies and houses of those making baobab products. The number of respondents interviewed varied from site to site depending on level of trading taking place at the site at that particular time. A total of 83 respondents were interviewed constituting 41 males and 42 females. A structured questionnaire was also used for interviews to investigate baobab products, buyers, price setting and factors affecting price, unit of trade and price, revenue and net profits.

2.2.3 Data analysis

Data were analyzed using SPSS version 11.5. Chi-Square test was carried out to test whether there were differences between sites in social and marketing variables. Cross tabulations were used to calculate percentages to show the magnitude of use and importance for the parameter and categories.

2.3 Results

2.3.1 Social value of baobab

Statistical analysis revealed highly significant differences ($p \geq 0.001$) among sites in the use of leaves, fruits, bark, roots and wood. Leaves were predominantly used as a vegetable, locally known as *therere,* in Chikwawa (52.1%) and Mwanza (73.7%) (Table 2.1); while medicinal use was largely (23.2%) in Karonga. The majority of respondents in Salima (90%) and Karonga (73.9%) did not use leaves. Fruits were mainly used for pulp consumption (72%) and for sale (54%). The bark was greatly used for medicinal purposes in Chikwawa (29.6%) and Karonga (55.4%). In addition, the bark was used for ropes in Mwanza (47.4%), Salima (55%), Chikwawa (31%) and Karonga (23.2%). Many people in Karonga (46.4%) and Chikwawa (56.3%) used roots for medicinal purposes. There was growing utilisation of wood for firewood in Chikwawa (25.4%), Salima (43.3%) and Karonga (21.7%); while in Chikwawa people largely (23.9%) used wood for timber.

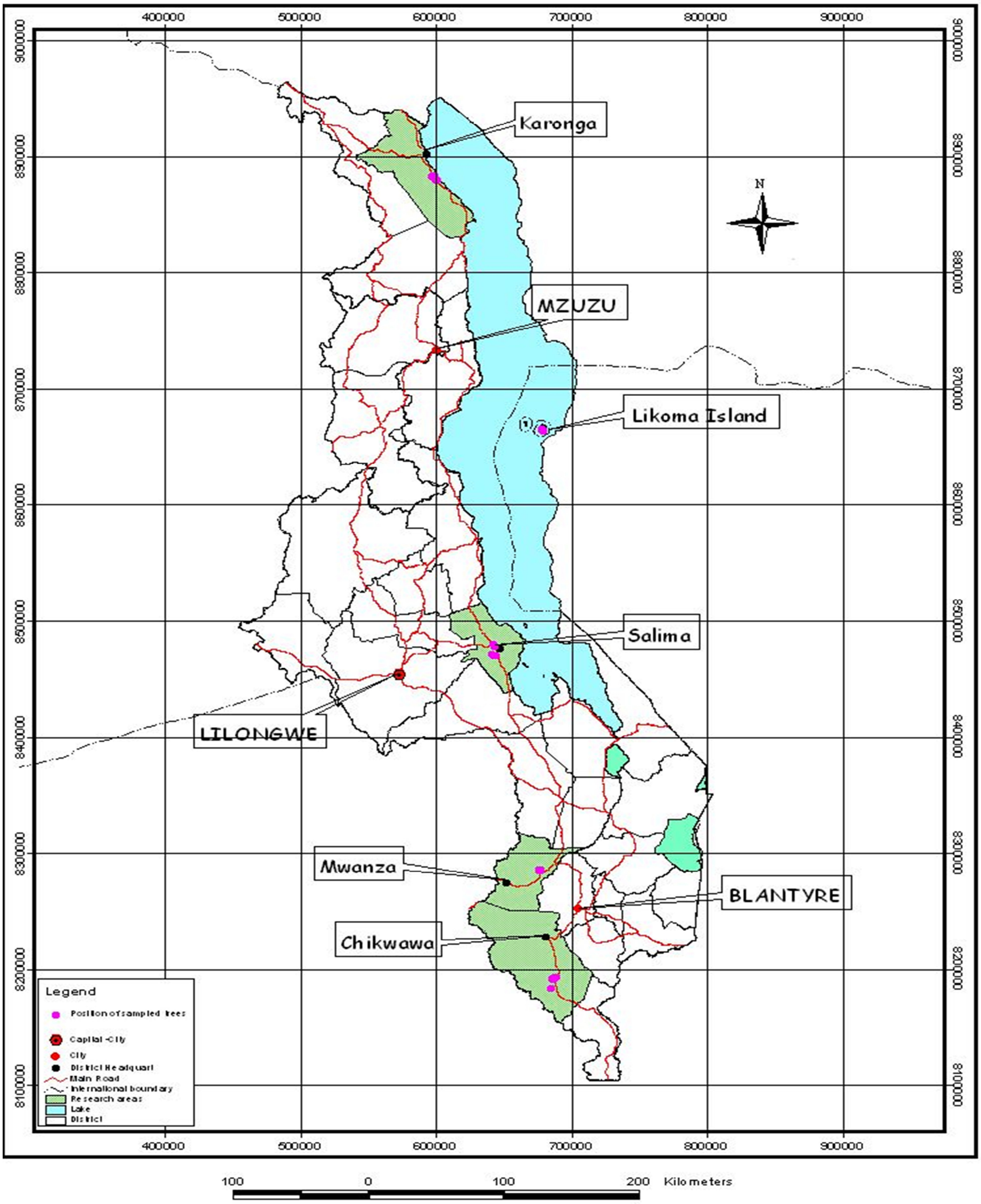

Fig. 2. 1 Map showing location of Karonga, Chikwawa, Mwanza, Salima and Mzuzu study areas and two major cities Blantyre and Lilongwe

Table 2. 1 Frequency of use of leaves, fruit, bark, roots and wood of baobab tree at four sites in Malawi

Parameters categories	Sites				Average Response (N=219)
	Chikwawa (N=71)	Mwanza (N=19)	Salima (N=60)	Karonga (N=69)	
	Percentage of respondents				
Leaves:					
Food (Therere)	52.1	73.7	5	0	24.6
Medicine	11.3	5.3	5	23.2	12.8
Food(Therere)/medicine	21.1	10.5	0	0	7.8
Fodder	0	5.3	0	0	0.5
Manure	1.4	0	0	1.4	0.9
Sprouting indicating onset of rainfall	0	0	0	1	0.5
None*	14.1	0.9	90	73.9	53
Fruit:					
Consumption	46.5	15.8	16.7	37.7	32.9
Sale	40.8	10.5	8.3	0	16.4
Consumption and sale	2.8	31.6	66.7	52.2	38.4
Tea and soda	1.4	5.3	0	0	0.9
Tea/Phala/consumption /sale	0	26.3	0	0	2.3
Coffee/mosquito repellent/juice/soda	0	5.3	0	0	0.5
Consumption/soda	2.8	0	1.7	1.4	1.8
Consumption and adding to milk	0	0	0	4.3	1.4
Consumption/sale/fuel	0	0	5	4.3	2.7
Sale/mosquito repellent	1.4	0	0	0	0.5
None	4.2	5.3	1.7	0	2.3

Table 2.1 continued

Bark:					
Medicine	29.6	0	6.7	55.4	25.6
Sale	7	10.5	0	1.4	3.7
Ropes	31	47.4	55	23.2	36.5
Medicine and sale	1.4	5.3	0	1.4	0.9
Medicine and ropes	1.4	5.3	0	2.9	1.8
Sealing holes in pots	0	0	0	1.4	0.5
Soda	2.8	0	0	0	0.9
None	28.2	31.6	38.3	24.6	30.1
Root:					
Food	5.6	5.3	5	1.4	4.1
Medicine	56.3	10.5	1.7	46.4	34.2
Ropes	2.8	5.3	0	7.2	3.7
Firewood	0	0	0	1.4	0.5
Medicine and ropes	1.4	0	0	1.4	0.9
Food/medicine/ropes	1.4	0	0	0	0.5
Making wheels (Children)	0	36.8	0	0	3.2
None	32.4	42.1	93.3	42	53
Wood:					
Firewood	25.4	5.3	43.3	21.7	27.4
Fencing(Timber)	23.9	0	1.7	0	0.5
Medicine	5.6	0	0	4.3	3.2
Sale	0	5.3	0	0	0.5
Growing spot for edible mushrooms-after falling	0	10.5	11.7	0	4.1
Ropes	0	0	3.3	0	0.9
Firewood/soda	0	0	3.3	0	0.9
None	45.1	78.9	35	72.5	53.9

* none means not using the part

2.3.2 Economic value

The study found that there were significant differences ($p \geq 0.001$) among sites in products sold, type of buyers, factors affecting price over time, unit of trade and price. Price setting and total expenditure were not significantly different between sites (Table 2.2). Trade in raw fruit was most prominent in Mwanza (57.1%) and Mzuzu (60%). Marketing in juice was high in Chikwawa (44%) and Salima (67.6%). Other value addition albeit on minor products, was on lolly-pop freezes, jam and pulp. In general, fruit juice was a predominant product (42.2 %) followed by raw fruits. The results revealed that those who bought to consume predominate in buying baobab fruits in Chikwawa (88%), Salima (88.2%) and Mzuzu (80.8%). The participation of middlemen in baobab trade was most outstanding in Mwanza (42.9%); while retailers and juice producers were active in Chikwawa and Mwanza, respectively. Overall, the most important buyers of baobab products were the final consumers (78.3%). Generally, the majority (81.9%) of sellers set the price for the products accepting minimal haggling (13.3%). The major unit of trade was a sachet of freezes (57.3%) and 50 kg bag (volume) of fruits (25.6%). The study showed that the major factors that influenced price changes partly varied from one locality to another but season (time) of fruit maturity greatly influenced price in Mwanza (69.2%), Salima (97%) and Karonga (50%); while manufacturing of freezes in Chikwawa considerably (68%) pushed up prices. To a lesser extent, quality of fruits and running cost influenced pricing in Chikwawa and Mwanza, respectively.

Table 2. 2 Frequencies of products sold, buyers, factors affecting price and means of transporting and distance to markets at four sites in Malawi

Parameters and categories	Study sites				
	Chikwawa (N =25)	Mwanza (N =14)	Salima (N=34)	Mzuzu (N=10)	Average response (N=83)
	Percentage respondents				
Products sold:					
Raw fruits	28	57.1	26.5	60	36.1
Freezes	16	21.4	0	40	13.3
Juice	44	7.1	67.6	0	42.2
Freezes/Juice	12	0	0	0	3.6
Juice and Jam	0	0	5.9	0	2.4
Pulp	0	14.3	0	0	2.4
Buyers:					
Final consumers	88	35.7	88.2	80	78.3
Agents(Middlemen)	4	42.9	0	20	10.8
Retailers	8	14.3	0	0	4.8
Final consumer/Agents	0	0	11.8	0	4.8
Juice producers	0	7.1	0	0	1.2
Price setting					
Buyer(customer)	4	0	0	0	1.2
Seller	84	57	85.3	100	81.9
haggle	12	28.6	11.8	0	13.3
Village Committee	0	14.3	0	0	2.4
Combination Seller and bargaining	0	0	2.9	0	1.2
Unit of trade :					
Individual fruit	12.5	7.1	2.9	40	11
Sachet of freezes	66.7	21.4	70.6	40	57.3
50 kg bag (volume) of fruits	20.8	64.3	20.6	0	25.6

Table 2 continued					
90 kg bag (volume) of fruits	0	0	0	20	2.4
Dozen fruits	0	0	2.9	0	1.2
500 ml bottle	0	7.1	2.9	0	2.4
Factors affecting price over time:					
Fruit quality	12	0	3	0	4.9
Season	16	69.2	97	50	61.7
Synthesis of freezes	68	23.1	0	20	27.2
Fruit quality/season/freezes	0	0	0	30	3.7
Running costs	0	7.7	0	0	1.2

Exchange rate: 1USD = 140 Malawian Kwacha (MK140.00)

2.3.3 Unit pricing and revenue

The price of a sachet of lolly-pop freezes (ice drink) ranged from MK5 to MK10 depending on size of sachet while a 500 ml bottle of juice was sold at a fixed price of MK80 for both cottage industries. A 50 kg bag (volume) of fruits was sold at a price ranging from MK150 to MK400 but was uniquely high in Mzuzu City at MK2000. For a 90 kg bag, the price ranged from MK400to MK900. On the other hand, the price for a 50 kg bag of pulp ranged from MK1200 to MK1500. Individual traders made revenue ranging from MK300 to MK99,000 per month while total cost incurred varied from zero for those who were just collecting to MK16, 700 for producers and middlemen or long distance traders. Cottage industries made revenue ranging from MK111,715 to MK132,000 per month. The highest net profit for individual traders was about MK82,300 and MK214,900 for cottage industry per month.

2.4 Discussion

2.4.1 Social value

This study revealed that baobab is extremely important for the livelihood of rural Malawians. Every part of the tree is put to use supporting what has been reported by Gebauer et al. (2002) that all parts of baobab tree are utilised. Leaves have been found with multiple uses; the most important roles being use as food and medicine. Young leaves are used as a spinach form of vegetable "therere" and sometimes mixed with okra. Vegetable use was prominent in Chikwawa and Mwanza whilst medicinal use was important in Karonga and Chikwawa. Traditional medicinal knowledge is usually passed down from parents to their siblings (Lin 2005). Some of the medicinal uses included treatments of stomach pains, sexually transmitted diseases, sores and skin rashes. Several authors (Kristensen and Lykke 2003); Leakey 1999; Sidibe et al. 2002; Sidibe et al. 1998; and Wickens 1979) have reported major use of young leaves as cooked spinach and frequently dried, powdered leaves are used for sauces. Leaf products are also sold in formal markets in Mali (Gustad et al. 2004) and Sudan (Gebauer et al. 2002). Leaves have also been reported to be used as condiments and medicine (Nirvana 2000; Kalinganire et al. 2008; ICUC 2002). Sidibe et al. (1998) reported that in Sahel, apart from use as food sauces, dried leaves are marketed in the dry season. The leaves provide vital protein sources which improves protein quality of the diet (Gebauer et al. 2002). Baobab leaves have high amino acid, magnesium and manganese (Sidibe and Williams 2002). Leaves in Malawi are underutilized as food but have potential as source of vitamins, protein and other nutrients for the rural people living in semi-arid areas.

Fruits are at present both source of income and for domestic use. Other important minor uses included roasted and grounded seed used as beverage (coffee), pulp made into porridge "lembezi" usually taken for breakfast. In Karonga, pulp is used as a coagulant in milk. Commercialisation may include minor products as a form of value addition. Fruit pulp has also been reported as food and medicine (Gebauer et al. 2002; Gustad et al. 2004; Gruenwald 2009; Kalinganire et al. 2008; Kristensen and Lykke 2003; Saka et al. 2008; Sidibe and Williams 2002). Roasted seeds are nutritious, rich in protein and fats essential in local diets (Gustad et al. 2004; Kristensen and Lykke, 2003; Sacandé et al. 2006; Sidibe and Williams 2002). Baobab pulp has potential to make commercial wine, thus adding value to the tree and also increasing income generation for rural people (Ham and Akinnifesi 2006). Surprisingly, pulp in Malawi is not used for medicine compared to other countries. With an oil content in seed of 15% (Gustad et al. 2004), seed is used as a source of cooking oil and also exported to international markets for use in cosmetic industries (ICUC 2002; Venter and Witkowski 2011).

Results revealed that the bark and roots were traditionally important largely for medicinal use mainly in Karonga and Chikwawa. The bark fibre was also predominantly used for construction, tethering livestock and craft work. Similar results have been reported by Gebauer et al. (2002) in Sudan where baobab bark produces strong fibre which is used for making mats, bags, hats and finer bark for weaving. Trade in ropes made from bark fibre has also been reported in Mali (Gustad et al. 2004). In addition, bark is also used in fetish traditional medicine for making amulet for children in Mali (Gustad et al. 2004). Wickens (1979) reported infusion of roots used for bathing babies in order to promote smooth skin and fibre used for making fishing nets, sacks and mats. There is an escalation on the use of baobab wood for construction and fuel wood. The use of porous and spongy wood with poor burning properties has been explained due to lack of selectivity and scarcity of the hardwoods (Kristensen and Lykke 2003).

2.4.2 Economic value

Value addition in form of juice production is at present a major baobab preoccupation in the central and southern regions of the country (Table 2.2). The major contributing factor could be the influence of Wildlife Society of Malawi and Ngolowingo cottage companies in the southern and central regions, respectively. These companies have mostly been distributing their products in the southern and central regions. Secondly, it could be due to large tree populations of the species found in the two regions. Thus people have traditionally been using the tree for a long time for income generating activities. However, the local juice manufacturing is mostly done by those with capacity to buy cooler boxes (see Fig.2.2), fridges and owning houses with electricity. In addition, juice makers must also purchase packaging plastics, sugar, colouring and flavouring agents apart from the capital for buying fruits. This agrees with the assertion of Campbell and Byron (1996) that commercialisation of miombo products would work to the advantage of elite groups rather than local communities. Ham and Akinnifesi (2006) reported that community processors preferred production of juice and jam to oil and wine. However, the communities cited the lack of processing equipment, packaging materials, insufficient capital and lack of markets as major limitations faced by rural processing groups. High quality juice is made by the two cottage industries and the products are widely distributed in super markets; but the capacity of these companies is low to satisfy the local demand. One of the major problems is lack of modern technology to mass produce juice. However, both companies indicated that there were plans to add jam production as a form of diversification. Trading in raw fruits is a major preoccupation of middlemen during fruiting season who take the merchandise to urban markets (see Fig.2.4). Shackleton et al. (2007) also reported that production of non-timber forest products for local markets is usually part-time. Unfortunately, the trade is mostly relying on natural trees that cannot be described as a sustainable resource due to lack of natural regeneration and mortality of old trees (Chirwa et al. 2006). The long distance covered by middlemen transporting raw fruits show importance of baobab in the rural economy. Transporting of fruits from rural areas to urban areas and cities supports Shackleton et al. (2007) assertion that growing demand for forest products is as a result of large and growing numbers of rural migrants who create new supply links between the forests and the cities creating burgeoning market demand for low cost forest products. High demand is an opportunity to commercialize baobab products at domestic and national level.

Local people can benefit if high value products made from baobab (Gruenwald and Ganiza 2005; Venter and Witkowski 2011) such as wine, seed oil, root like carrots (see Fig.2.3), and cosmetics could be manufactured locally with government financial support (Ham and Akinnifesi 2006). The advantage with baobab is that both the fruits and pulp have a long shelf life than most other fruits. The major problem observed with this trade, especially selling of ice-drinks, was the overly engagement of children labour in the product marketing; thereby curtailing their education, as they often focus on the trade as alternative income source, thereby altering their priority too early in their life. The other concern is the hygiene in the trade (Akinnifesi et al. 2006) typified by questionable quality of water used for juice making and unsanitary placement of raw fruits on sale usually heaped on the ground. For this reason locally processed fruits are not attractive to the middle class segment of the market.

Fig. 2. 2 Locally made baobab ice-drink (Freezes)

Fig. 2. 3 Roots and leaves of *Adansonia digitata* L seedlings unexploited potential food

The results have revealed that there is no shortage of buyers for baobab products at local and national level (Table 2.2). Middlemen are further linking producers and urban areas (see Fig. 2.4) thereby increasing the value of baobab. PhytoTrade Africa is also a major buyer of baobab fruits from southern Africa mainly for export to EU markets (Gruenwald and Ganiza 2005). However, the operations of this company were not recorded in all the study areas. It may be speculated that middlemen who were found buying in bulk in Salima and Mwanza were ultimately selling to this company. The problems experienced in marketing in Malawi have been pointed out by Shackleton et al. (2007) including communication, inadequate education and levels of organization among producers and traders. Proper organisation in NTFPs trade can lead to high revenue to community organisations (Tchoundjeu, et al. 2008) due to increase in bargaining power and negotiation skills. Teklehaimanot et al. (2008) has also argued that if local farmers are to become effective market players, they have to engage in networking activities and seek strategic alliances in producer-industry partnerships and linking to high value and export markets. Akinnifesi et al. (2008) has also recommended that in order to increase income for rural people, market intervention should give attention to improved access to market information, product development, identification of new markets, certification and labeling. This requires collective action of rural groups to assemble and market produce and policy reforms to improve conditions governing their operations. Hence, baobab fruit collectors also need proper organisation and linkage to established traders and policy reforms to maximize benefits from the forest resource.

Fig. 2. 4 Lorry transporting bags of baobab fruits from Salima to Lilongwe

The study further showed that price setting in baobab trade in Malawi is mainly controlled by the sellers with minimal bargaining between sellers and buyers (Table 2.2). According to Christian et al. (2008) the price set may depend on people's ability to negotiate the price of products and market them, taking advantage of access to market information in global environment. Furthermore, Ham et al. (2008) pointed out that pricing in indigenous fruits should be determined by adding a profit to all costs such as purchasing cost, rental, security, tax, seasonality, fruit taste (sweetness), labour costs, transportation and basic needs of households for immediate cash and the supply of fruits versus demand. In this study, season was the most prominent factor that influenced level of prices in all sites (Table 2.2). The collectors indicated that prices were higher at the beginning of the fruiting season and leveled off at the peak of fruit maturity and picked up again at the end of the season. Similarly, Haq et al. (2008) has reported price dynamics with the simultaneous ripening of all fruits causing glut in the market thereby lowering prices, followed by relative scarcity and higher prices. However, farmers can maximize income by adding value to the fruit and having improved market information (Akinnifesi, et al. 2008). The study also revealed that the price of a sachet of freezes (MK 5-10 (USD 0.036-USD 0.071) was surprisingly uniform across the country. The behaviour of pricing seems monopolistic. It is probable that prices may have been pegged against commercial freezes manufactured by Asian micro-industries. Prices could be deliberately set to out-compete the other types of freezes. Displacement of commercial freezes by *Mlambe* freezes shows potential for commercializing the products. Vendors indicated that the coming in of *Mlambe* juice on the market had resulted in reduced sales of commercially produced ice-drinks. The price for 500 ml bottled *Mlambe* juice from cottage companies was also uniform at MK80 (USD 0.57). It's probable that the Ngolowingo Women Group may have adopted the price from the founders of the product Wildlife Society of Malawi since the cost of producing the product could not be the same. However, with different cost of production, it is not yet known how the uniform pricing is affecting revenue and net profits. Although the revenue for individual traders found in this study ranged from MK350 to MK99,960 (US$2.5-US$714), it is quite substantial for smallholder subsistence farmers. Kasulo et al. (2009) reported that revenue of US$20 per month is quite substantial for rural household in Malawi. Thus, source of extra money no matter how little go long way to the livelihoods of rural community dwellers (Lowore 2006). Net profit in this study ranging from MK83,260.00 (USD 595) and MK208,285 (USD1488) per month is well above the minimum wage in Malawi. In a feasibility study undertaken by Joordan et al (2008), the net profit in the range from 13.8 to 29.8% was reported. The lower net profit was from Tabora in Tanzania, and the highest was for Baobar in Zimbabwe, while Malawi juice concentrate at Magomero was put at 27.7%. Shackleton et al. (2007) in their review also found that individual incomes of some entrepreneurs were obtaining incomes above the minimum wage. In addition, Shackleton et al. (2007) reported that even in USA and Europe, NTFP trading activities are prospering as traditional products continue to be appreciated and favoured by the rich and poor alike. The potential of generating much revenue from baobab is high especially at present when rapid rural to urban migration in Malawi is taking place, creating high demand by the rich and poor alike.

2.5 Conclusion

Commercial processing of indigenous fruit is one of the few comparative advantages that rural communities have in Malawi**.** Multiple social roles and substantial revenue generated from the trade of *Adansonia digitata* presents the importance of the species in its contribution to the livelihoods and generation of income to rural communities. Pulp production is a process that can be used to add value to fruits to fetch high prices for rural communities. In addition, the shell may also be used as firewood. The market chain for baobab trade covers a wider range beyond local collectors signifying the importance of baobab at domestic and national sphere. The potential of commercialization as shown by the vibrant cottage industry with a potential export market calls for the establishment of sustainable resource use by instituting proper management practices including domestication of the species. Poor market mechanism for local players calls for government policies that create an enabling environment to facilitate the marketing to move from the informal to the formal sector and a re-look at both the land and tree tenure issues to encourage true ownership of the resource.

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2.7 Reference

Akinnifesi FK, Sileshi G, Ajayi OC, Tchoundjeu Z (2008). Accelerated Domestication and Commercialization of Indigenous Fruit and nut Trees to Enhance Better Livelihood in the Tropics: Lessons and Way Forward. In: Akinnifesi FK, Leakey RRB, Ajayi OC, Sileshi G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds). Indigenous Fruit Trees in the Tropics. Domestication, Utilisation and Commercialization. CABI, Wallingford, UK, pp392-428.

Brigham T, Chihongo A, Chidumayo E (1996) Trade in Woodland products from the Miombo region. In: Campbell, C (ed). The Miombo in Transition: Woodlands and Welfare in Africa. Centre for International Forestry Research (CIFOR), Bogor, Indonesia

Campbell B, Byron N (1996) Miombo Woodlands and rural livelihoods options and opportunities. In: Campbell, C (ed). The Miombo in Transition: Woodlands and Welfare in Africa. Centre for International Forestry Research (CIFOR), Bogor, Indonesia.

Chirwa M, Chithila V, Kayambazinthu D, Dohse C (2006) Distribution and population Structures of *Adansonia digitata* in Malawi. FRIM, Zomba, Malawi

Chirwa PW, Syampungani S, Geldenhuys CJ (2008) The ecology and management of the miombo woodlands for sustainable livelihoods in southern Africa: the case of non-timber forest products. Southern Forests 70(3): 237-245

Christian MY, Chirwa PW, Ham C (2008) The influence of tourism on the woodcarving treade around Cape Town and implications for forest resources in southern Africa, Development Southern Africa 25 (5): 577-588

De Smedt S, Alaerts K, Kouyaté AM, Van Damme P, Potters G, Samson R (2011) Phenotypic variation of baobab (*Adansonia digitata* L.) fruit traits in Mali. Agroforestry Systems 82: 87-97

Dhillion SS, Gustad G (2004) Local management practices influence the viability of the baobab (Adansonia digitata Linn.) in different land use types, Cinzana, Mali. Agriculture, Ecosystems and Environment 101: 85-103

FAO (1982) *Adansonia digitata* L. Fruit Bearing Forest Trees 34:1-3

Gebauer J, El-Siddig K, Ebert G (2002) Baobab (*Adansonia digitata* L.): a Review on a multipurpose Tree with Promising future in the Sudan. Gartenbauwissenschaft, 67(4): 155-160

Gruenwald J (2009) Novel botanical ingredients for beverages. Clinics in Dermatology 27: 210-216

Gruenwald J, Galiza MI (2005) *Adansonia digitata* L. Baobab. Market Brief in the European Union for selected natural ingredients derived from native species. The United Nations Conference on BioTrade Facilitation Programme (BTFP)

Gustad G, Dhillion SS, Sidibé D (2004) Local use and cultural and economic value of products From Trees in the parklands of the Municipality of Cinzana, Mali. Economic Botany 58 (4) 578-587

Guy GL (1971) The Baobabs: *Adansonia* spp. (Bombacaceae). Journal of the Botanical Society of Southern Africa (57): 31-37

Ham C, Akinnifesi F (2006). Zimbabwe, Zambia, Malawi and Tanzania: priority fruit species and products for tree domestication and commercialization. IK Notes. No. 94. July 2006

Ham C, Akinnifesi FK, Franzel S, Jordan D, Hansmann C, Ajayi OC, de Kock C (2008) Opportunities for Commercialization and Enterprise Development of Indigenous Fruits in Southern Africa. In: Akinnifesi FK, Leakey RRB, Ajayi OC, Sileshi G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds). Indigenous Fruit Trees in the Tropics. Domestication, Utilisation and Commercialization. CABI, Wallingford, UK, pp. 254-272.

Haq N, Bowe C, Dunsiger Z (2008) Challenges to stimulating the Adoption and Impact of Indigenous Fruit Trees in Tropical Agriculture. In: Akinnifesi FK, Leakey RRB, Ajayi OC, Sileshi G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds). Indigenous Fruit Trees in the Tropics. Domestication, Utilisation and Commercialization. CABI, Wallingford UK, pp 50-69

Hardcastle PD (1978) A preliminary silvicultural classification of Malawi. Forestry Research Institute of Malawi (FRIM), Forest Research Record No. 57

International Centre for Underutilized Crops (ICUC) (2002) Fruits for the Future. Baobab. Factsheet No.4. March, 2002. ICUC, Southampton, UK

Kalaba FK, Chirwa PW, Syampungani S, Ajayi OC (2010) Contribution of agroforestry to biodiversity and livelihoods improvement in rural communities of Southern African regions. In: Teja Tscharntke, Christoph Leuschner, Edzo Veldkamp, Heiko Faust, Edi Guhardja, Arifuddin Bidin (Eds.).Tropical rainforests and agroforests under global change. Springer Berlin, pp 461-476

Kalinganire A, Weber JC, Uwamariya A, Kone B (2008) Improving Rural Livelihoods through Domestication of Indigenous Fruit Trees in the Parklands of the Sahel. In: Akinnifesi FK, Leakey RRB, Ajayi OC, Sileshi G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds). Indigenous Fruit Trees in the Tropics. Domestication, Utilisation and Commercialization. CABI, Wallingford UK, pp 186-200

Kasulo V, Mwabumba L, Munthali CRY (2009) A review of edible orchids in Malawi. Journal of Horticulture and Forestry 7: 133-139

Kristensen M, Lykke AM (2003) Informant -Based Valuation of Use and Conservartion Preferences of Savanna Trees in Burkina Faso. Economic Botany 57(2) 203-217

Kumar BM (2008) Homegarden-based Indigenous fruit Tree Production in Peninsular India. In: Akinnifesi FK, Leakey RRB, Ajayi OC, Sileshi G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds). Indigenous Fruit Trees in the Tropics. Domestication, Utilisation and Commercialization. CABI, Wallingford UK, pp 84-99

Leakey RRB (1999) Potential for novel food products from agroforestry trees: a review. Food Chemistry 66:1-4

Lin KW (2005) Ethnobotanical study of medicinal plants used by the Jah Hut people in Malaysia. Indian Journal of Medical Sciences, 59(4):156-161

Maranz S, Niang A, Kalinganire A, Konaté D, Kaya B (2008) Potential to harness superior nutritional qualities of exotic baobabs if local adaptation can be conferred through grafting. Agroforestry Systems, 72:231–239

Mbabu P, Wekesa L (2004) Status of Indigenous Fruits in Kenya. In: Chikamai B, Eyog-Matig, O, Mbogga M (eds.). Review and Appraisal on the Status of Indigenous Fruits in Eastern Africa. Kenya Forestry Research Institute, Nairobi, Kenya. pp 55-58

Mithofer D, Waibel H (2008) Economics of On-farm Production of Indigenous fruits. In: Akinnifesi FK, Leakey RRB, Ajayi OC, Sileshi G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds). Indigenous Fruit Trees in the Tropics. Domestication, Utilisation and Commercialization. CABI, Wallingford, UK, pp 237-272

Nirvana H (2000) Baobab Tree- *Adansonia digitata.* http://www.blueplanetbiomes.org/baobab.htm. Site visited 3/5/2009

Ramadhani, T and Schmidt, E, 2008. Marketing of Indigenous Fruits in Southern Africa. In: Akinnifesi, F.K, Leakey, R.R.B, Ajay, O.C, Sileshi, Tchoundjeu, Z, Matakala, P and Kwesiga, F.R (eds). Indigenous Fruit Trees in the Tropics. Domestication, Utilisation and Commercialization. CABI, Wallingford, UK, pp 224-236

Sacandé, M., Rønne, C., Sanon, M.D. & Joker, D. (2006) Adansonia digitata L. Seed Leaflet No. 109. Hørsholm: Forest & Landscape Denmark

Saka JDK, Kadzere I, Ndabikunze BK, Akinnifesi FK, Tiisekwa BPM (2008) Product Development Nutritional Value, Processing and Utilization of Indigenous Fruits from the Miombo Ecosystem. In: Akinnifesi FK, Leakey RRB, Ajayi OC, Sileshi G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds). Indigenous Fruit Trees in the Tropics. Domestication, Utilisation and Commercialization. CABI, Wallingford, UK, pp 288-309

Saka KJD, Msonthi JD (1994). Nutritional value of edible fruits of indigenous wild trees in Malawi. Forest Ecology and Management 64: 245-248

Shackleton S, Shanley P, Ndoye (2007) Invisible but viable: recognising local markets for non-timber forest products. International Forestry Review 9: 697-712

Shackleton SE, Shackleton CM, Netshiluvhi TR, Geach BS, Balance A, Fairbanks DHK (2002) Use Patterns and Value of Savanna Resources in Three Rural Villages in South Africa. Economic Botany 56(2): 130-146

Sidibe M, Williams JT (2002) Baobab. *Adansonia digitata*. International Centre for Underutilized Crops, Southampton, UK

Sidibe M, Kone M, Schierle J, Frigg M (1998). The baobab tree as a source of vitamins. Agroforestry Today 10 (4): 7-9.

Tchoundjeu Z, Atangana A, Asaah E, Tsobeng A, Facheux C, Foundjeu D, Mbosso C, Degrande A, Sado T, Kanmegne J, Mbile P, Tabuna H, Anegbeh PM, Useni M (2008). Domestication, Utilization and Marketing of Indigenous Fruit Trees in West and Central Africa. In: Akinnifesi FK, Leakey RRB, Ajayi OC, Sileshi G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds). Indigenous Fruit Trees in the Tropics. Domestication, Utilisation and Commercialization. CABI, Wallingford, UK, pp 171-185

Teklehaimanot Z (2008). The Role of Indigenous Fruit Trees in Sustainable Dryland Agriculture in Eastern Africa. In: Akinnifesi FK, Leakey RRB, Ajayi OC, Sileshi G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds). Indigenous Fruit Trees in the Tropics. Domestication, Utilisation and Commercialization. CABI, Wallingford, UK, pp 204-223

Tongco, M.C, 2007. Purposive Sampling as a tool for Informant Selection. Ethanobotany Research & Applications 5:147-158

Venter SM, Witkowski ETF (2011) Baobab (*Adansonia digitata* L) fruit production in communal and conservation land-use types in Southern Africa. Forest Ecology and Management 261: 630-639

Wickens GE (1979) The Uses of the Baobab (*Adansonia digitata* L.) in Afica. In: Kunkel (ed.), Taxonomic aspects of African economic Botany, AETFAT

Wickens GE (1982) The Baobab-Africa's upside-down tree. Kew Bulletin, Vol. 37, No.2: 173-209

3 Phenotypic variation in fruit and seed morphology of *Adansonia digitata* (Baobab) in five selected wild populations in Malawi

Abstract

Adansonia digitata (Baobab) is widely distributed throughout Sub-Saharan Africa stretching from Senegal to Sudan, and from Ethiopia to northern South Africa. Information of phenotypic and genetic variation is a prerequisite for the domestication and improvement of baobab fruits from the wild. A study was done to determine within and between provenances variability in fruit and seed characteristics of five populations selected from four silvicultural zones and assess whether morphometric traits could delineate populations from different zones into land races. Fruits were collected from 59 trees representing a wide geographical range. Six fruit traits and three individual seed traits were assessed. Results showed highly significant differences ($P \leq 0.001$) in fruit, pulp, and seed weight, fruit length and width, number of seed, individual seed weight, seed length and width within and between populations. Mean fruit weight ranged from 125.8 ± 3.25 g to 162.9 ± 3.25 g, seed weight ranged from 38.6 ± 2.5 g to 66 ± 2.01g and pulp weight ranged from 28.7 ± 1.33 g to 41.4 ± 1.33 g. Single seed weight showed pronounced evidence of divergence of populations into ecotypes. The rich diversity found between and within populations is important for domestication purposes and tree improvement through selection and breeding. All populations could be used for seed source but distribution should be consciously done recognizing existence of races.

Keywords: Ecotype ; Domestication; Miombo; Diversity

3.1 Introduction

Adansonia digitata (baobab) belongs to the family Bombacacea (Wickens 1982) and the genus Adansonia (Moiselet 1998). Adansonia is a member of the tribe Adansonieae, or Bombaceae (Sidibe and Williams 2002). Generally in Africa the species is indigenous in drier areas found in West Africa, Sudan, Angola, East Africa, Southern Africa up to northern South Africa covering about 26 countries (Sidibe and Williams 2002; Wickens 1982). The importance of baobab to human livelihood is reviewed by several authors such as Gruenwald and Galiza (2005), Kalinganire et al (2008), Teklehaimanot et al. (2008), FAO (1982), Sidibe and Williams (2002) and ICUC (2002). In addition, baobab is currently a crop of high economic value internationally (De Smedt et al. 2011; Gruenwald and Galiza 2005). Baobab oil, extracted from the seed, is used in the cosmetics industry and is also sold internationally (Gruenwald and Galiza 2005). Baobab fruit pulp has been approved for sale in the EU (2008/575/EC) and USA (GRAS Notice No. GRN 000273) (De Smedt et al. 2011). With a wide range of geographical distribution, certain plant characteristics represent morphotypes (Sidibe and Williams 2002). Zobel and Tarbert (1984) have attributed phenotypic variations in forest trees to three factors: the differing environments in which the trees are growing, the genetic differences among trees and interaction between the tree genotypes and environments where they grow. Across geographical range of baobab, the major phenotypic variation reported include leaf taste, size, form and colour of fruits, seed colour, pulp taste and colour, bark colour, height and width of trees (Sidibe and Williams 2002; Assogbadjo et al. 2006) and fruit traits (Smedt et al. 2011). Presently, domestication of *Uapaka kirkiana* in southern Africa has begun with clonal propagation work, especially in Zambia and Malawi (Akinnifesi et al. 2008b).

With domestication, phenotypic and genetic aspects should be embedded in baobab domestication process to confer tree populations' the ability to keep an appropriate level of genetic diversity to maintain short-term viability and long-term evolutionary potential (Buiteveld et al. 2007). Forest management may affect the genetic resources through drift, mating system related processes, fertility and viability selection related process and migration related processes (Buiteveld et al. 2007). Natural selection has resulted in formation of geographical races (ecotypes) in tree species (Chapman and Reiss 2000) which are in most cases genetically controlled (Zobel and Tarbert 1984) and should be considered in tree domestication, conservation and improvement strategies. Studying phenotypic and genetic variation in heterogeneous environment is key to understanding factors that shape the population structure on which tree domestication, conservation, management and improvement strategies can be modeled (Bizoux and Mahy 2007). Katsvanga et al. (2007) have also reported high variability in fruit traits among sites largely attributed to climatic, edaphic, genetic, and cultural factors. The extent to which individuals develop within the species is also a function of the mechanism of seed dispersal (Chung et al. 2002). In addition, management regime within a given environment may also cause morphological variation especially for traits targeted by artificial selection (Carmona and Casas 2005). Patterns of genetic structuring among populations may be continuous, as in isolation –by-distance models of gene flow or discontinuous as in stepping stone models (Franks et al. 2004). Plant breeders exploit the variability existing in the trees by selecting desirable characteristics for use in domestication and genetic improvement (Katsvanga et al. 2007). According to Dawson et al. (2008), unless a wide genetic base within the species is maintained, trees are vulnerable to inbreeding depression ultimately reducing individual fitness and raises the chances of population and or species extinction.

Presently, there is scarcity of information on phenotypic and genetic structure of baobabs in southern Africa even though the species is extremely important socially and economically (Gruenwald and Galiza 2005; Akinnifesi et al. 2008b). Saka et al. (2008) reported significant differences in physic-chemical and nutritional contents between five provenances of baobab fruits in Malawi. The patterns and extent of fruit variation existing across populations occurring in varying silvicutural zones in Malawi remains unknown. Knowledge of variation in any species is prerequisite in agroforestry (Dawson et al. 2008) and any tree improvement programme (Wright 1976; Zobel and Talbert 1984). Successful domestication requires identification and use of good genetic germplasm. Furthermore, tree improvement and genetic conservation requires understanding of variation (Zobel and Tarbert 1984). The current study was undertaken to determine the pattern of fruit and seed variation occurring between and within five baobab populations selected from different silvicultural zones in Malawi. The major questions addressed included the following: (i) What is the level of phenotypic variation existing between and within natural populations growing in different ecological zones based on climatic and edaphic factors? (ii) Could morphometric traits be used to delineate populations into distinct races (ecotypes)?

3.2 Methodology

3.2.1 Study site and fruit collection

The study populations were selected as per the silvicultural zones (A, Ba, L and J) delineated by Hardcastle (1978) (Table 3.1, Fig.3.1). In addition, Likoma Island population, apart from occurring in silviculture zone L, was included due to its geographical isolation. According to Hardcastle (1978), silviculture zone L has mean annual rainfall (MAR) >1600 mm with predominantly weathered ferralitic soils. Silviculture zone Ba has MAR ranging between 710 to 850 mm and characterized by calcimorphic soils overlaying vertisols. Silviculture zone J has MAR ranging between 1200 to 1600 mm and characterized by ferrallitic soils whilst Silviculture zone A has MAR ranging between 710 to 840 mm with predominantly vertisols.

Fruits were collected from Karonga, Likoma Island, Salima, Mwanza and Chikwawa populations at the peak of fruit season between April and May. Dry fruits were randomly collected from trees spaced at a minimum distance of 100 m to avoid collection from related individuals (Schmidt 2000). Fruits were plucked from the tree and/or collected from the ground. A total of 65 families (separate seedlots from specific mother trees) were collected from the five populations, each consisting of different number of families ranging from 10 to 15 (Table 3.1).

Table 3. 1 Physical description of sites (populations/provenances) and number of trees (families) sampled

Population	Population Code	Silviculture Zone	Average annual Stress Period (weeks)*	Altitude (m)	Mean annual rainfall (mm)	Mean annual temperature (^{0}C)	Soil
Karonga (n=10)	K	L	7” Field capacity: 15 12” Field capacity”: 10	475-1000	>1600	23-25	Ferrisols dominant ; regosols
Salima (n=15)	S	Ba	7” Field capacity:28 12” Field capacity: 26	200-1200	710-850	20-25	Alluvial calcimorphic soils above the vertisols
Mwanza (n=15)	M	J	7” Field capacity:16 12” Field capacity: 11	900-1500	1200-1600	19-21	Sandy ferrallitic
Chikwawa (n=15)	C	A	7” Field capacity:35 12” Field capacity: 35	<200	710-840	>25	Vertisols
Likoma Island (n=10)	L	L	7” Field capacity: 15 12” Field capacity”: 10	475-1000	>1600	23-25	Ferrisols, alluvial calcimorphic, regosols, lithosols

Source: Hardcastle (1978)
* = Information on average annual stress calculations are reported by Hardcastle (1978)

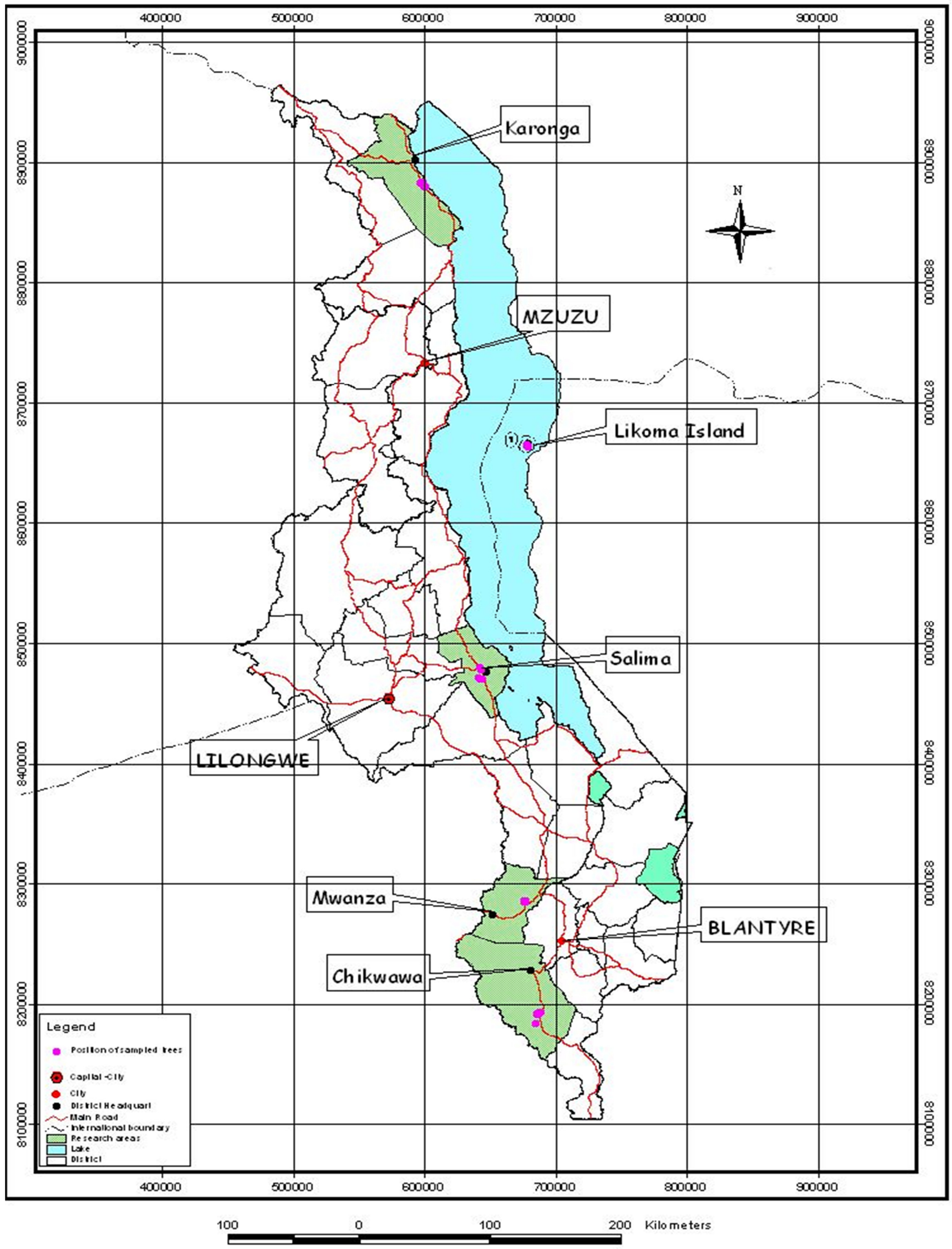

Fig. 3. 1 Map of Malawi showing location of 5 sampled *Adansonia digitata* populations

3.2.2 Fruit assessment, seed characteristics and Data analysis

In total, fruits from 65 trees were characterized. From each tree 20 fruits were used to assess fresh fruit weight, fruit length and width. A sub-sample of ten fruits from each tree (family) was selected randomly to assess seed number, seed weight and pulp weight. A sub-sample of five seeds was randomly picked from each of ten fruits (50 seed per family) to measure individual seed weight, length and width. Fruit length (cm) and width (cm) were measured to the nearest cm and fresh fruits weighed to the nearest g. After opening the fruit, the shell was removed from the shell content (pulp + seeds) and both fruit parts were weighed (to the nearest g). Fruit pulp and seeds were separated by dissolving the dry powdery pulp in water. After washing and drying, pulp weight was calculated by subtraction (Pulp+seed weight - seed weight). Individual seed weight was measured to the nearest hundredth of a gram whilst seed length and width to the nearest mm.

The data on fruit and individual seed variation was analysed using nested analysis. Means were separated using the least significant differences (LSD).

3.3 Results

3.3.1 Fruit variation between provenances

There were highly significant differences ($p \geq 0.001$) in fruit weight, length, width, pulp-seed weight, number of seed, seed weight and pulp weight between populations.

Fruit weight differed significantly among the provenances, with the Mwanza provenance having the heaviest fruits (162.9 ±3.25 g) and Salima having the lowest weight (125.8 ± 3.25 g). Karonga and Chikwawa provenance were not different (Table 3.2). But Chikwawa was superior to Likoma Island and Salima. Coefficient of variation in fruit weight was 40%.

Fruit length was significantly different among provenances, with Mwanza having the longest fruits (16.5 ± 0.16 cm) and Likoma Island having the shortest (11.9 ± 0.27 cm). But Salima was superior to both Chikwawa and Karonga. The coefficient of variation in fruit length was 18.9%. Fruit width varied significantly among the provenances, with width diameter longest in Mwanza (7.6 ± 0.06 cm) and Salima having the shortest (6.8 ± 0.06 cm). Mwanza, Karonga and Likoma Island were not different. Chikwawa was superior to Salima. The coefficient of variation in fruit width was 14.1%. Number of seed varied significantly among the provenences, with Karonga having the largest number of seed (111± 4.36) and Salima with the least (86 ± 3.36). Mwanza and Karonga were not different. Chikwawa and Likoma Island were not also different. The coefficient of variation in number of seed was 42%. Seed weight varied significantly among the provenances, with Mwanza having heaviest seed weight (66 ± 2.01 g) and Likoma Island having the lowest (38.6 ± 2.5 g). Chikwawa and Karonga were not different. Salima was superior to Likoma Island. The coefficient of variation in seed weight was 39.2%. Seed weight constituted on average 39% of whole fruit weight. Pulp weight was significantly different among provenances, with Chikwawa having the heaviest pulp weight (41.4 ± 1.33 g) and Salima having the lowest (28.7± 1.33 g). Mwanza and Chikwawa were not different but superior to Salima. The coefficient of variation in pulp weight was 45.1%. Pulp weight constituted on average 20% of whole fruit weight.

Table 3. 2 Variation in fruit traits among five provenances of *Adansonia digitata*

Provenance	Fruit weight (g)	Fruit length (cm)	Fruit width (cm)	Number of Seed	Seed weight (g)	Pulp weight (g)
Mwanza	162.9 ±3.25a	16.5 ±0.16a	7.6 ±0.06a	108±3.36)a	66.0 ±2.01a	41.4 ±1.33a
Chikwawa	142.9 ±3.31b	13.8 ±0.16c	7.1 ±0.06b	91±3.74b	58.7±2.24b	41.5± 1.48a
Karonga	136.8 ±4.32bc	13.4 ±0.30c	7.5 ±0.08a	111±4.36a	58.5± 2.61b	34.5± 1.73b
Likoma Island	128.8±4.00 c	11.9 ±0.27d	7.5± 0.07a	99±4.18b	38.6 ±2.50d	34.7±1.66b
Salima	125.8 ± 3.25 c	14.4 ±0.16b	6.8 ±0.06c	86 ±3.36c	49.5± 2.01c	28.7± 1.33c
% Fruit wt					39	20
LSD	10.62	0.51	0.19	10.81	6.47	4.29
CV (%)	40	18.9	14.1	42	44.7	45.1

Mean values are followed by the standard error of the mean

Means followed by the same letter are not significantly different ($P \leq 0.001$)

3.3.2 Fruit variation between trees within provenances

Within all provenances, variation in mean fruit weight among trees was highly significant ($P \leq 0.001$) (Fig.3.2). Across all the provenances, mean fruit weight ranged from 61.2 ± 12.6 g in one tree in Likoma Island (L5) to 256.1 ± 12.6 g in another tree in Chikwawa (C15). Seed weight variation was highly significant ($P \leq 0.001$) (Fig. 3) within all the provenances. Across all the provenances seed weight ranged from 6.9 ± 7.8 g in one tree in Likoma Island (L5) to 115.4 ± 7.8 g in another tree in Chikwawa (C10). Pulp weight variation was highly significant ($P \leq 0.001$) (Fig.3.4). Across all the provenances pulp weight ranged from 15.4 ± 5.2 g in one tree in Likoma Island (L5) to 69.1 ± 5.2 g in another tree in Chikwawa (C10).

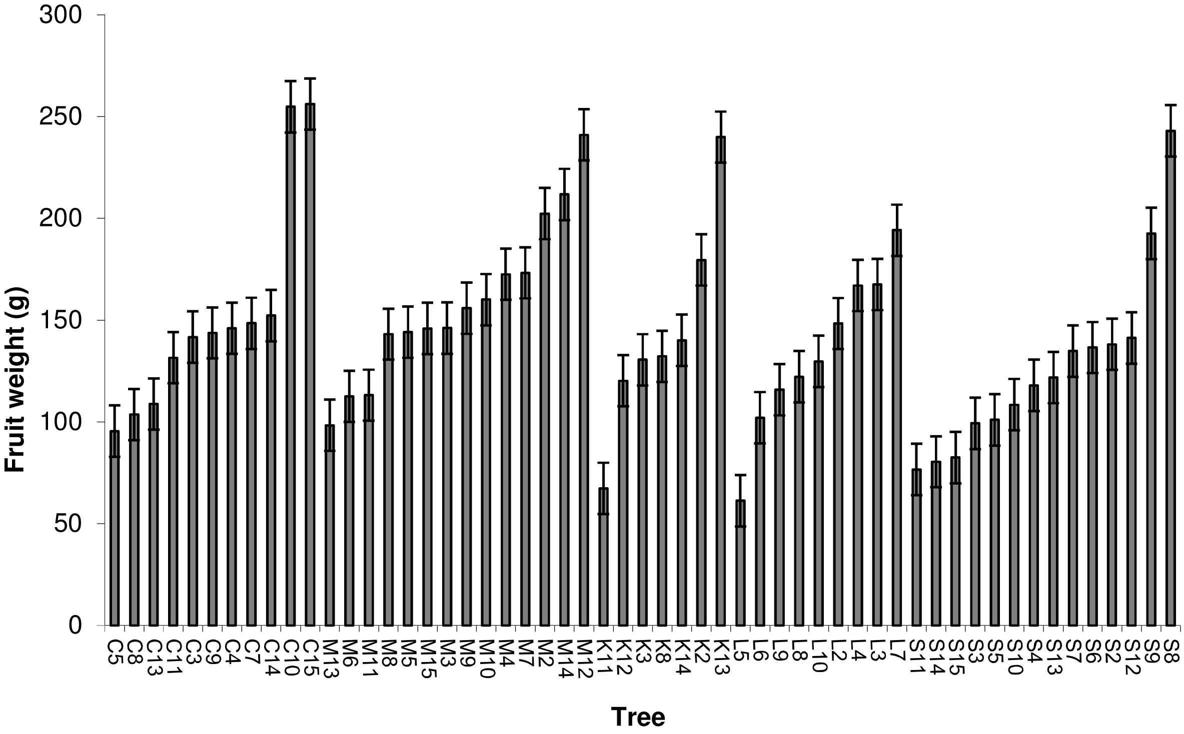

Fig. 3.2 Fruit weight variation within five provenances of *Adansonia digitata* (C=Chikwawa, M=Mwanza, S=Salima, L=Likoma Island and K=Karonga; label of individual trees range from 1 to 15)

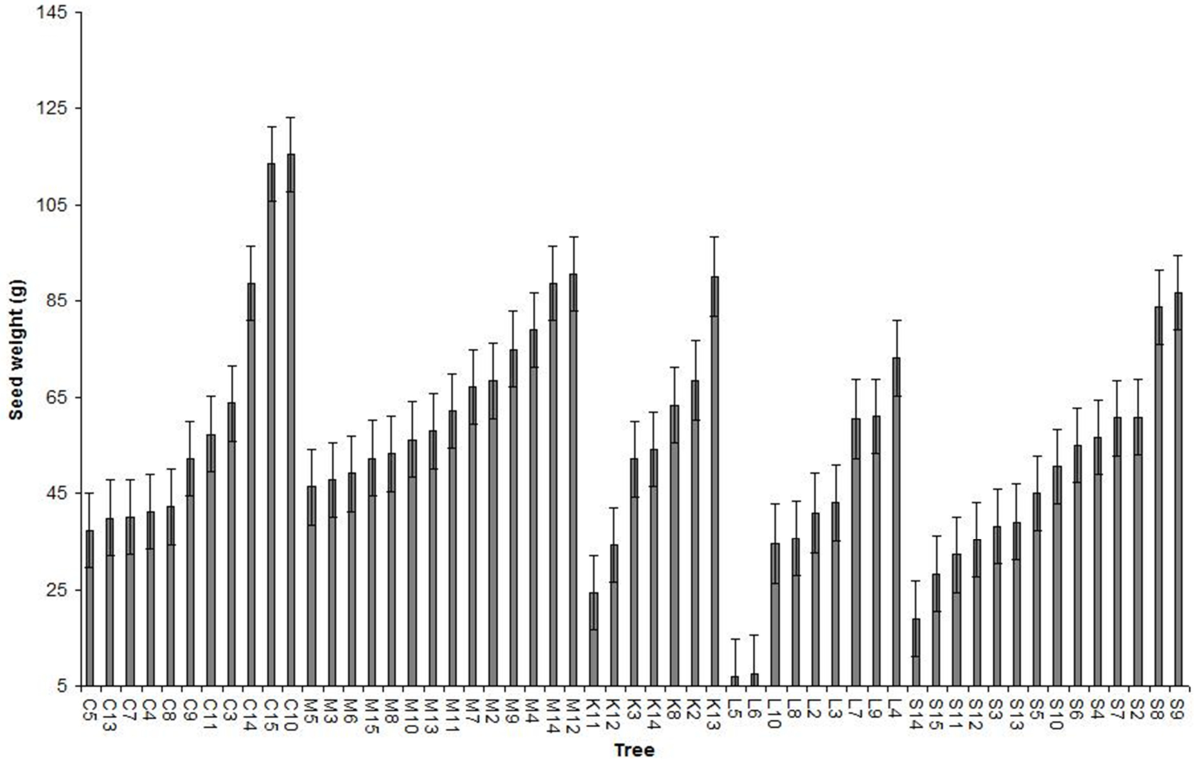

Fig. 3. 3 Seed weight variation within five provenances of *Adansonia digitata* (C=Chikwawa, M=Mwanza, S=Salima, L=Likoma Island and K=Karonga; label of individual trees range from 1 to 15)

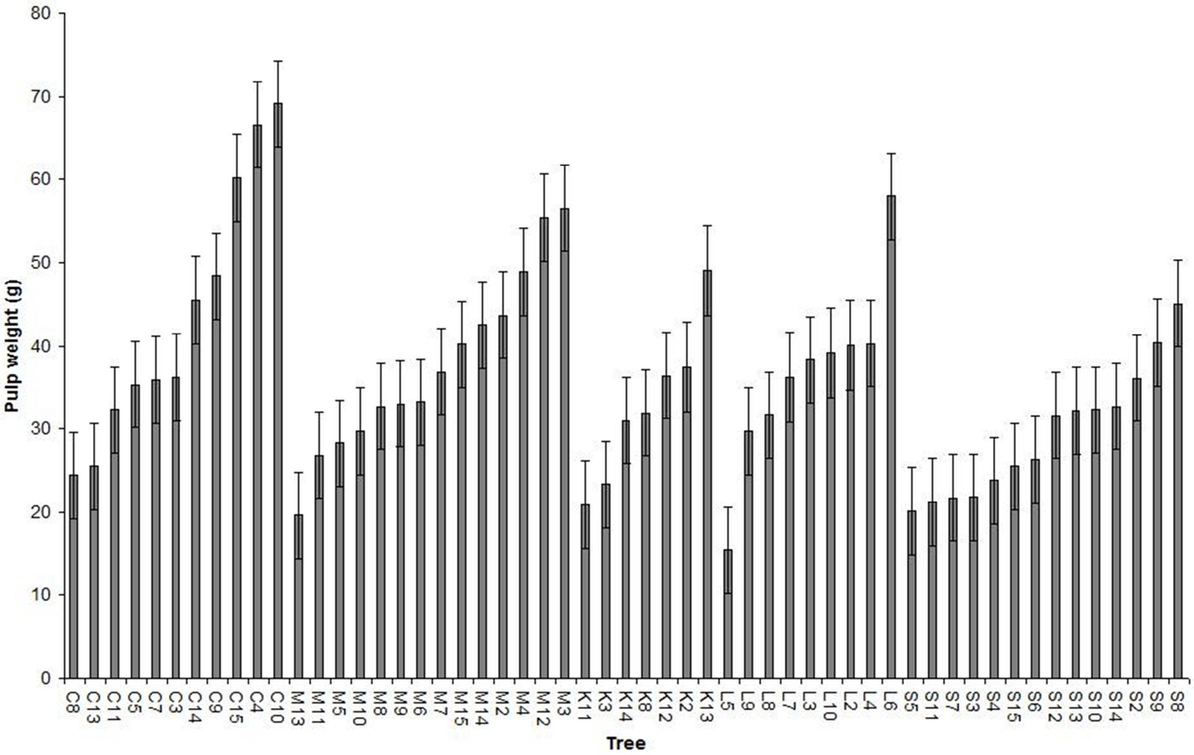

Fig. 3. 4 Pulp weight variation within five provenances of *Adansonia digitata* (C=Chikwawa, M=Mwanza, S=Salima, L=Likoma Island and K=Karonga; label of individual trees range from 1 to 15)

3.3.3 Single seed variation between provenances

Single seed weight was highly significant different (P≤0.001) among provenances, with Chikwawa having the heaviest single seed weight (0.576 ± 0.003 g) and Likoma Island having the lowest weight (0.367 ± 0.003 g) (Table 3.3). Mwanza and Salima were not different. Karonga was superior to Likoma Island. The coefficient of variation in single seed weight was 13.9%. Single seed length was highly significant different (P≤0.001) among provenances, with Chikwawa having the longest seed length (12.5 ± 0.03 mm) and Likoma Island having the shortest (10.9 ±0.04 mm). Mwanza, Karonga and Salima were not different. The coefficient of variation in seed length was 6.1%. Seed width was highly significant different (P≤0.001) among the provenances, with width diameter longest in Chikwawa (10.0 ± 0.02 mm) and Likoma Island having the shortest (8.9 ± 0.03 mm). Karonga and Salima were not different. Mwanza was superior to Karonga and Salima. The coefficient of variation in seed width was 6.5%.

Table 3. 3 Variation in single seed parameters among five provenances of *Adansonia digitata*

Provenance	Seed weight (g)	Seed length (mm)	Seed width (mm)
Mwanza	0.509±0.003b	11.8±0.03b	9.5±0.02b
Chikwawa	0.576±0.003a	12.5±0.03a	10.0±0.02a
Karonga	0.476±0.004c	11.8±0.03b	9.4±0.03c
Likoma Island	0.367±0.003d	10.9±0.04c	8.9±0.03d
Salima	0.502±0.003b	11.7±0.03b	9.4±0.02c
LSD	0.009	0.091	0.077
CV (%)	13.9	6.1	6.5

Mean are followed by the standard error of the mean,

Mean followed by the same letter are not significantly different at ($P \leq 0.001$)

3.4 Discussion

3.4.1 Fruit variation between and within provenances

In order to fulfill the goal of meeting the demands of subsistence farmers and product markets, the knowledge of intraspecific diversity of the baobab trees is fundamental. Current results fill the gap of information for baobab phenotypic diversity for fruit characteristics and individual seed traits for use in the domestication and tree improvement process. The present results have revealed substantial variation in fruit traits (fruit weight, length and width, seed number and weight and pulp weight) between (Table 3.2) and within (Fig.3.2, 3.3, 3.4) the provenances. Considering the best and lowest mean, between provenance variation was of high magnitude: fruit weight 119%, fruit length 37%, fruit width 12%, number of seed 26%, quantity of seed 33% and pulp weight 44%. Extremely high variation existed at family level for example 318% for fruit weight, 1572% seed weight and 476% pulp weight. The findings are important because they suggest the potential of achieving high genetic gains through classical tree breeding and through vegetative propagation. The results support the assertion that use of clones in fruit trees might increase productivity rapidly (Akinnifesi et al. 2008a). The presence of high coefficient of variation (CV= 40-45%) indicates the presence of high environmental impact where as low values (CV=14-19) could be an indication of strong genetic control (Kimmins 1987). In the case of our study, fruit weight, seed weight and pulp weight had high environmental influence. Most qualitative traits such as fruit length and width should have high narrow sense heritability sometimes up to 60% (Akinnifesi et al. 2008b). Therefore, these qualitative traits are easily improved through clonal selection. On the other hand quantitative traits such as fruit weight, seed number and weight and pulp weight should be influenced partly by additive gene effects, environment and interaction of genotype x environment (Zobel and Tarbert 1984). Thus, quantitative traits require classical breeding to achieve high genetic gains (Zobel and Tarbert 1984). The diversity of fruit sizes found in this study has unveiled high polymorphism existing in populations of baobabs in the country. Nearly all the forms of fruits described by Wickens (1982) such as globose, bluntly ovoid, sharply ovoid, large ovoid, shallow sulcate and oblong-cylindrical (Fig. 5) exist within the nation. This implies rich diversity that does not require an infusion of external genetic material for domestication and genetic improvement. The patterns of fruit sizes presently found can not be used to specifically classify populations as ecotypes due to overlapping in multiple comparison tests. Generally all the populations possess fruit sizes that are of economic value and can be used for domestication purposes. However, further research is required to assess the frequencies of different fruit types between and within the sites. Our results indicate viability of increasing baobab fruit productivity through clonal forestry as advocated by ICRAF (Akinnifesi et al. 2008a). However, it should be supported by research on genotype x environmental interaction (clonal plasticity) especially for quantitative traits that have large environmental influence.

The largest component of fruit weight (39%) constituted seed weight. Seed has many economical attributes such as oil for cooking and cosmetic industries (Gruenwald and Galizia 2005; Kalinganire et al. 2008; Teklehaimanot et al. 2008; Venter et al. 2011). Presently the product is not fully utilized in southern Africa, usually considered as a waste by product of juice making. Once promoted, seed weight could be improved through selection that may significantly uplift the life of many rural masses.

In general, sale of fruits is based on size (weight, length, width) (Franzel et al. 2008); with bigger fruits fetching higher prices. Tree breeding may target trees with bigger fruits. It has been reported by Katsvanga et al. (2007) that high fruit diversity attributes among sites could be attributed to climatic, adaphic, genetic and cultural factors. In the case of our sampling sites (Table 3.1), there were huge differences in environmental factors which may be linked to differences in fruit weight related parameters observed. Domestication process involves moving genotypes from one site to another. Presently, it is not known how genotypes would respond once planted in an exotic habitat. Will the fruit quality (shape, size, seed number, seed weight, pulp colour, nutrition and weight) be consistent when seed is moved from one ecological zone to another? In this case, provenance and family evaluation are prerequisite for successful large domestication programmes. Some of the questions to answer in an evaluation programme will be whether there is relationship between altitude, latitude, rainfall, temperature, relative humidity, stress period and edaphic factors on the fruit attributes. Fruit attributes are also influenced by tree management (Katsvanga et al. 2007) such as spacing, pruning, fertilization, irrigation and mulching. Currently it is unknown how baobabs would respond to different silvicultural practices. Further research is required since Zobel and Tarbert 1984 are of the view that to optimize tree productivity a combination of choice of parentage and silvicultural management should be observed. However, Akinnifesi et al. (2008a) reported varying responses to cultural management for miombo species: *Vangueria infausta* performed better with a combination of light dry-season irrigation and manuring whilst combining use of manure, fertilizer and irrigation did not improve yield in *Uapaka kirkiana* and *Sclerocarya birrea*. In Mali, baobab leaf production was extended through irrigation especially for black morphotypes and it was presumed applicable to fruit production too (Kalinganire et al. 2008).

The variation in fruit characteristics for individual trees (Fig.3.2, 3.3, 3.4) show clear trends that the higher the fruit weight the higher the seed weight. The results are supported by Mkonda et al. (2003) who reported strong relationship between seed weight and fruit weight because the sink strength of fruits depends on the numbers and sizes of the seed they contain. In addition, seeds constitute the priority sink as they have big share of substrates than the other fruit tree parts. This could also explain the consistently low pulp weight in most fruits as compared to seed weight.

There were, however, few exceptions where some trees had almost the same level of seed weight and pulp weight. Currently, pulp weight is the most important economic product (Gruenwald and Galiza 2005). Therefore, more economic weighting should be put on this trait in selection index. The results however seem to show that indirect selection of pulp weight based on fruit weight may end up with large error margin because some fruits with high fruit weight are not necessarily having significantly different fruit pulp than relatively smaller fruits as shown by different ranking of trees within provenances in Fig.3.3 and Fig.3.4 in relation to Fig.3.2. Additionally, pulp weight is influenced by environmental factors largely by the amount of rainfall (Katsvanga et al. 2007). This implies that selection of plus trees to improve pulp production should be done following several seasons of data collection of pulp production.

Our results compare favourably with several published literature on baobab phenotypic variations: (Barwick 2004; Carr 1958; De Smedt, et al. 2011; FAO, 1982; Gebauer et al. 2002; Gruenwald and Galiza 2005; Nour et al. 1980). Gruenwald and Galiza (2005) reported that the percentage of fruit pulp varies according to the origin. In southern Africa, fruit pulp constitutes 16.5% of the fruit weight and seed weight 38% whilst in Senegal fruit pulp is about 12%. Gebauer et al. (2002) reported variation in fruit length of 20 to 30 cm and width diameter up to 10 cm. Barwick (2004) reported fruit length of up to 30 cm. Carr (1958) reported range in mean fruit weight of 59.5 g to 88 g in one study and 173.2 g to 252 g in another. Carr (1958) also reported mean percentage seed in whole fruit of 34.9% and 14.3% for pulp weight. Green (1932) reported composition of seed weight of 40% and 15% for pulp. FAO (1982) have also reported baobab fruit size of up to 35 cm length by 8 to 15 cm width. De Smedt et al. (2011) have reported mean pulp yield per fruit of 45 ± 1g and seed production of 71 ± 2g. Our results on pulp weight was higher because it included moisture content of fresh pulp which ranges from 10 to 12% (Gruenwald and Galiza 2005).

Fig. 3. 5 Fruit variation between and within five populations of *Adansonia digitata* in Malawi

3.4.2 Individual seed variation between provenances

Most literature in baobab has not reported variation in individual seed traits. However, the range of seed traits found in this study is similar to values reported by Owen (1999) of seed size measuring 5 mm by 10 mm and weighing on average 0.65 g. Msanga (1998) reported seed dimensions of 12 mm in length and 8 mm broad. The differences in reported results are showing importance of studying variation at regional scale. Low coefficient of variation (6.1-13.9%) between populations in seed trait (Table 3.3) might indicate that the traits are highly genetically controlled (Kimmins 1987). Seed characteristics may also strongly depict species adaptability (Chapman and Reiss 2000). Multiple comparison tests clustered the populations differently depending on seed trait (see Table 3.3). Likoma Island population is consistently classified differently from the other four mainland populations. The contributing factor could be difference in climate especially higher available moisture and short period of moisture stress since it has a maritime climate - influenced by surrounding Lake Malawi. Not surprising, due to low water stress, the seed is the lightest and smallest which may imply that trees do not need to reserve much substrates for the seedlings to survive in the environment (Katsvanga et al. 2007). Similarly, rainfall pattern of Karonga and Salima populations are partly influenced by Lake Malawi; and as such their seed characteristics are intermediate. The other two populations, Chikwawa and Mwanza, are both in land populations but lie in different topographic regions influenced by different climatic regimes (see Table 3.1). Chikwawa has extremely harsh climate such as high temperature and long moisture stress. Thus, the seed are heavy and large due to stored reserves (substrates) presumably adequate for the seedlings to survive well; unlike Mwanza where the population is in a wetter place but still experience relatively prolonged stress period. Thus, the population has the second heaviest and largest seed. The differentiation in seed traits is suggesting that populations with time may have evolved differently due to different ecological conditions (Kimmins 1987). These results are similar to the findings on *Pterocurpus angolensis* (Munthali 1999) where it was reported that rainfall strongly influenced seed size. In this case, populations receiving heavy rainfall had lightest seed while those from low rainfall areas had heaviest seed. These results are supported by earlier reports on miombo tree species (Munyanziza 1996) which are said to react to stress by developing large seed with large cotyledons that feed developing seedlings. The same phenomenon has been shown in this study; Chikwawa and Mwanza provenances have heavy seed because they experience severe water stress as compared to the rest of the populations. Thus, it can be argued that rainfall regime may have partitioned baobab populations into ecotypes in Malawi. Overlapping of the provenances could be due to gradual differentiation or introgression as a result of continuous variation in some subpopulations (Frank et al. 2004). For example most of the populations occur in the Lake Shore areas without many barriers. Current results also mirror findings of Mwase et al. (2006) who found variation in seed weight in *Uapaka kirkiana* provenances and attributed divergence in seed traits due to differences in water availability with changes in elevation. Ngulube et al. (1996) attributed variation in seed size in *Uapaka kirkiana* due to genetic and environmental effects too. Present results of seed weight may imply that the trait could be used to predict genetic structuring between populations and distinguish the populations from different ecological zones.

3.5 Conclusion

Large between and within provenance phenotypic diversity found in this study indicates great potential for improving fruit quality. The fruit variation between provenances ranging from 12% to 119% and between tree to tree variation 318% to 1572% is wide enough for domestication initiatives and further demonstrate substantial genetic gains that could be achieved through selection and breeding. The variation found can be quickly harnessed in tree improvement through vegetative propagation that can also accelerate fruit precosity from more than ten years to four years (Kalinganire et al. 2008). The breeding strategy may also use sexual procedure to increase genetic diversity. Combined selection of provenance and tree to tree (family) variation could be used to attain high genetic gains. However since baobab is a fruit tree, sexual reproduction may result in gene segregation which will eventually affect fruit quality. Research should be initiated to understand inheritance of fruit traits and information generated will be used for designing tree breeding strategies. Following high diversity in all provenances, all populations can be used for selection depending on the trait being selected for. Overlapping in clustering of populations in fruit characteristics does not provide an easy way of delineating populations as distinct gene pools. However, Mwanza and Salima were consistently different from other populations probably showing that they are unique ecotypes. The other three populations were generally intermediate which may imply that they are genetically similar. However, fruit characteristics are greatly affected by environmental factors, cultural factors and additive gene effects (Kasvanga et al. 2007). Further studies are required to understand plasticity of traits of economic importance such as fruit pulp and nutritional aspects. In addition, horticultural studies should be done to test the effect of cultivation on fruit attributes especially following clonal selection. It has been argued that during the development of germplasm networks, proper consideration must be given to geographic scale because significant mal-adaptation and/or out-breeding depression are possible if germplasm exchange occurs over too large a geographic area (Dawson et al. 2008). In the present study, individual seed weight also seemed to strongly suggest populations of Chikwawa, Karonga and Likoma Island are unique ecotypes while Mwanza and Salima are similar. The question is whether the seed for these populations can be adapted (distributed and planted) in all the silvicultural zones. The present study has demonstrated importance of studying variation in fruit trees. Baobab occurs widely in southern Africa, but it is still poorly researched in population genetics. It will be important to extensively investigate the spatial genetic structuring in the species in the region so that the information could be used in tree domestication, conservation, management and improvement. In addition, further molecular studies should be done to assess genetic diversity at large scale (regional) and fine scale (within countries) to complement on the current study. So far the results are showing all provenances have substantial fruit diversity that may be put to use in domestication and tree improvement strategies.

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3.7 References

Akinnifesi Fk, Ajayi OC, Sileshi G, Matakala P, Kwesiga FR, Ham C, Kadzere I, Mhango J, Mng'omba SA, Chilanga T, Mkonda A (2008a) Creating opportunities for domestication and commercializing Miombo Indigenous Fruit Trees in Southern Africa. In: Akinnifesi, FK, Leaky RRB, Ajayi OC, Silesh G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds.) Indigenous Fruit Trees in the Tropics Domestication, Utilization and Commercialization. Wallingford, UK, pp137-170

Akinnifesi FK, Sileshi G, Ajayi OC, Chirwa PW, Kwesiga FR, Harawa R (2008b) Contribution of Agroforestry Research and Development to Livelihood of Smallholder Farmers in Southern Africa:2.Fruit, Medicinal, Fuelwood and Fodder Tree Systems. Agricultural Journal 3(1): 76-88

Barwick M (2004) Tropical and Subtropical Trees: A worldwide Ecyclopaedic Guide, Thames and Hudson, London, UK

Bizoux J, Mahy G (2007) Within –Population Genetic Structure and Clonal Diversity of a Threatened Endemic Metallophyte, *Viola calaminaria* (Violaceae). American Journal of Botany 94 (5): 887-895

Buiteveld J, vendramin GG, Leonardi S, Kamer K, Geburek T (2007) Genetic diversity and differentian in European beech (Fagus sylvatica L.) stands varying in management history. Forest Ecology and Management 247:98-106

Carmora A, Casas A (2005) Management, phenotypic patterns and domestication of Polaskia chichipe (Cactaceae) in the Tehua Can Valley, Central Mexico. Journal of Arid Environments 60: 115-132

Chapman JL, Reiss MJ (2000) Ecology Principals and Applications (2nd Edition). Cambridge University Press, UK

Chirwa M, Chithira V, Kayambazinthu D, Dohse C (2006) Distribution and Population Structures of *Adansonia digitata* in Some Parts of Ntcheu, Dedza and Mangochi Districts, Malawi. FRIM Report No. 06002

Chung M, Nason J, Chung M, Kim K, Park C, Sun B, Pak J (2002) Landscape-Level Spatial Genetic Structure in *Quercus acutissima* (Fagaceae). American Journal of Botany 89 (8): 1229-1236

De Smedt S, Alaerts K, Kouyate AM, Van Damme P, Potters G, Samson R (2011) Phenotypic variation of baobab (*Adansonia digitata* L.) fruit traits in Mali. Agroforest Syst 82:87-97. DOI 10.1007/s10457-010-9357-0

Donahue JK, Upton JL (1996) Geographic variation in leaf, cone and seed morphology of *Pinus greggii* in native forests. Forest Ecology and Management 82: 145-147

Environmental affairs Department (2006) National Biodiversity Strategy and Action Plan. Ministry of Energy, Mines and Natural Resources. The Government Printer, Lilongwe, Malawi
FAO (1982) Fruit bearing Forest trees. FAO Forest Paper, Vol 34

Franks SJ, Richards CL, Gonzales J, Cousins JE, Hamrick (2004) Multi-scale genetic analysis of *Uniola paniculata* (Poaceae): A coastal species with a linear, fragmented distribution. American Journal of Botany 91 (9): 1345-1351

Franzel S, Akinnifesi FK, Ham C (2008) Setting Priorities among Indigenous Fruit Tree Species in Africa: Examples from Southern, Eastern and Western African Regions. In: Akinnifesi, FK, Leaky RRB, Ajayi OC, Silesh G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds.) Indigenous Fruit Trees in the Tropics Domestication, Utilization and Commercialization. Wallingford, UK, pp1-27

Gebauer J, El-Siddig K, Ebert G (2002) Baobab (*Adansonia digitata* L.): a Review on a Multipurpose Tree with Promising Future in the Sudan. Gartenbauwissenschaft, 67(4).S. 155-160

Green RA (1932) Composition of the pulp and seeds of *Adansonia digitata*. Botanical Gazette, vol.94(1): 215-220

Gruenwald J, Galizia MD (2005) Market Brief in the European Union for selected natural ingredients derived from native species. *Adansonia Digitata* L. Baobab. The United Nations Conference on Trade and Development (UNCTAD). Bio Trade Initiative/BioTrade Facilitation Programme (BTFP)

Hardcastle PD (1978) A preliminary silvicultural classification of Malawi. Forestry Research Institute of Malawi. Forestry Research Record No. 57

ICUC (2002) Fruits for the Future, Baobab. International Center for Underutilized crops. Factsheet No.4, March 2002, Southampton, UK

Kalinganire A, Weber JC, Uwamariya A and Kone B (2008) Improving Rural Livelihoods through Domestication of Indigenous Fruit Trees in the Parklands of the Sahel. In: Akinnifesi FK, Leaky RRB, Ajayi OC, Silesh G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds.) Indigenous Fruit Trees in the Tropics Domestication, Utilization and Commercialization. Wallingford, UK, pp186-200

Katsvanga CAT, Jim L, Gwenzi D, Muhoni L, Masuka P, Moyo M (2007) Characterisation of community identified *Uapaka kirkiana* phenotypes for domestication. Journal of Sustainable Development in Africa Volume 9 No.4:356-366

Mkonda A, Lungu S, Maghembe JA, Mafongoya PL (2003) Fruit-and seed-germination characteristics of *Strychnos cocculoides* an indigenous fruit tree from natural populations in Zambia. Agroforestry Systems 58: 25-31

Moiselet D (1998) The Baobab Tree. http://www.senegal-online.com/anglais/pares-faune-flore/baobab.htm. Site visited on 8/28/03

Msanga, H.P., 1998. Seed germination of indigenous trees in Tanzania: including notes on seed processing, storage and plant uses. Canadian Forest Service, Edmontort, Alberta

Munthali CRY (1999) Seed and seedling variation of *Pterocarpus angolensis* DC from selected natural populations of Malawi. Dissertation, University of Stellenbosch

Munyanziza E (1996) Miombo trees: Ecology strategies, silviculture and management. Ambio 25, 7:454-458

Mwase WF, Bjornstad A, Ntupanyama YM, Kwapata MB, Bokosi JM (2006) Phenotypic variation in fruit, seed and seedling traits of nine *Uapaka kirkiana* provenances found in Malawi. Southern African Forestry Journal, No.208, 15-21

Nour A, Magboul, BI, Kheir NH (1980) Chemical composition of baobab fruit (*Adansonia digitata* L.). Trop. Sci. 1980, 22(4): 383-388

Owen M (1999) Desk Study: Baobab fruit and oil. SAFIRE, June 1999

Ramadhani T, Schmidt E (2008) Marketing of Indigenous Fruits in Southern Africa. In: Akinnifesi, FK, Leaky RRB, Ajayi OC, Silesh G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds.) Indigenous Fruit Trees in the Tropics Domestication, Utilization and Commercialization. Wallingford, UK, pp224-236

Sacande M, Ronne C, Sanon M, Joker D (2006) *Adansonia digitata* L. SEED LEAFLET. No. 109 January 2006, Millennium Seed Bank Project, Kew, UK

Saka JDK, Kadzere I, Ndabikunze BK, Akinnifesi FK, Tiisekwa BPM (2008) Product Development: Nutritional Value, Processing and Utilization of Indigenous fruits from the Miombo Ecosystem. In: Akinnifesi, FK, Leaky RRB, Ajayi OC, Silesh G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds.) Indigenous Fruit Trees in the Tropics Domestication, Utilization and Commercialization. Wallingford, UK, pp288-309

Sidibe M, Williams JT (2002) Baobab, *Adansonia digitata*. International Centre for Underutilised crops, Southampton, UK

Teklehaimanot Z (2008) The Role of Indigenous Fruit Trees in Sustainable Dryland Agriculture in Eastern Africa. In: Akinnifesi, FK, Leaky RRB, Ajayi OC, Silesh G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds.) Indigenous Fruit Trees in the Tropics Domestication, Utilization and Commercialization. Wallingford, UK, pp204-223

Venter SM, Witkowski ETF (2011) Baobab (*Adansonia digitata* L.) fruit production in communal and conservation Land-use types in southern Africa. Forest Ecology and Management 261:630-639

Zobel B, Talbert J (1984) Applied Forest Tree Improvement. John Wily and Sons, Inc. New York

Wright JW (1976) Introduction to Forest Genetics. ACADEMIC PRESS, New York

4 Genetic variation among and within provenances of *Adansonia digitata* (Baobab) in seed germination and seedling growth from selected natural populations in Malawi

Abstract

Baobab (*Adansonia digitata*) trees have a wide variety of subsistence and economic values across sub-Saharan Africa. Knowledge of the genetic variation within and between the species is essential for management and designing domestication, improvement and conservation strategies. Fifty-nine half-sib families were evaluated in the nursery to determine the genetic variation and control within and between the populations from four silvicultural zones. Seed germination and seven seedling growth parameters were assessed. Total phenotypic variance, family variance, within family variance, individual narrow sense heritability, and additive genetic coefficient of variation (AGCV) were derived from observed and expected mean squares. There were highly significant differences in seed germination, plant height, root collar diameter, number of leaves, shoot height and diameter, tuber diameter and weight. Provenance variation in germination ranged from 46.7 ±3.3% to 68.7±3.3% while tree –to- tree variation ranged from 6.3±8.6% to 95.5±8.6%. Coefficient of variation in seedling growth variables ranged from 18.6% to 43.6%. Individual narrow sense heritabilities (h^2) ranged from 0.07 to 0.71. AGCV ranged from 3.21% to 14.67%. Morphological traits showed that mainland populations were genetically distant from the island one. High and moderate additive genetic control of traits and AGCV show the potential that Baobab can also respond well to tree improvement. High phenotypic variation found in the study offers an opportunity to effect selection of superior attributes at both provenance and individual tree-to-tree level.

Key words: Indigenous; Breeding; Selection; Diversity

4.1 Introduction

The miombo woodland contains over 200 tree species that bear edible fruits (Akinnifesi et al. 2008). More than 80% of the rural population in sub-Saharan Africa is poor and traditionally depend on forest for most of their livelihood such as use of fruits (Kalaba et al. 2010). Harvesting of the indigenous tree fruits from the wild can increase rural annual income by US$300 to US$2000 per household (Kalaba et al. 2010). Economic impact analysis of indigenous fruit trees has shown that fruit collection, consumption and sale reduce income poverty by 33% (Akinnifesi et al. 2008). The contribution of indigenous fruits of the miombo woodlands is especially important in the Southern African Development Community (SADC) mainly for the marginalized groups in the society, such as women, children and the landless (Akinnifesi et al. 2006; Shackleton et al. 2007). For instance, indigenous fruits contribute on average about 42% of the annual food basket and a majority of households rely on indigenous fruits during famine (Campbell et al. 1997). In a study conducted in Zimbabwe, Mithofer et al. (2006) reported that indigenous fruits reduced the probability of falling below the poverty threshold, thereby vulnerability to severe famine, during the critical food shortage period by 30%. In the case of *A. digitata,* farmers sell fruits to buy food. Shackleton and Shackleton (2004) have reported that non timber food products (NTFPs) assist households in times of adversity such as death or retrenchment of the head of the household or breadwinner, droughts, floods, frosts or diseases leading to crop failure or death of livestock. Furthermore, collection of indigenous fruits contributes between 5.5 and 6.5% of the total household income in the rural communities of Southern Africa (Akinnifesi et al. 2008; Kalaba et al. 2009).

About 85% of rural households in South Africa rely on edible fruits and wild spinaches (Shackleton and Shackleton 2004) and collect about 104 kg of edible fruits and 58 kg of wild spinaches. In addition, some indigenous fruit trees are important sources of medicine for the rural communities with about two-thirds of households utilizing indigenous fruit trees for medicinal purposes (Kalaba et al. 2010). In southern Africa, farmers traditionally integrate crops and fruit trees such as baobabs on their farm land either as wild, semi-domesticates or cultivated (Akinnifesi et al. 2008). A global study by Ruiz-Perez et al. (2004) indicated that harvesting of non-timber forest products from wild and semi-wild or lightly managed forests, is a viable subsistence strategy of households, providing extra income to household who depend on agriculture or off-farm income sources. *Adansonia digitata* (Baobab) tree is a characteristic species for drier areas, especially sites with annual rainfall between 500 and 800 m (Shivcharn and Gunnar 2004). The baobab is at its best on deep well-drained soil at altitudes of between 450 to 600 m above sea level with a rainfall between 300 and 500 mm per annum (Wickens 1982; Wilson 1988). However, it also occurs from sea-level to at least 1500 m and from areas receiving as little as 90 mm of rainfall to as much as 1400 mm annually, with extension to higher rainfall influenced by man (Wilson 1988). It extends from northern South Africa and Namibia to Ethiopia, Sudan and fringes of the Sahara (Gebauer et al. 2002; Sidibe and Williams 2002; Wickens 1982; Wilson 1988). Baobab has many socio-economic uses with nearly every part of the tree being used by human beings (Sidibe and Williams, 2002). According to ICUC (2002), baobab oil from seed is for cooking and is sold in local markets as well as international markets for use in cosmetics industries. Furthermore, its pulp is used for synthesis of beverages at local scale and processed industrially. Presently, pulp powder is an important export commodity to EU markets (Gruenwald and Galiza 2005). In Australia, a Baobab Fruit Company is conducting trials on the use of tubers as carrots with an initial cultivation project of 4000 plants (Gruenwald and Galiza 2005). The leaves are sold as well-sought important vegetables in parts of West Africa. Apart from manufacturing juice, baobab fruit has huge potential for making jam, oil and wine (Akinnifesi et al. 2008; Ham and Akinnifesi 2006).

Past research and extension efforts towards indigenous fruits were hampered due to biasness towards cultivation of exotic fruit tree species (Akinnifesi et al. 2008). However, for two decades, the World Agroforestry Centre (ICRAF) and its partner institutions have conducted research especially on domestication of indigenous fruit trees in southern Africa (Akinnifesi et al. 2008). For example, field trials on provenance and family variation have been done in southern Africa for *Uapaka kirkiana* and *Sclerocarya birrea* (Akinnifesi et al. 2008; Chirwa et al. 2007). In recent market studies (Akinnifesi et al. 2008), baobab has emerged as one of the priority species for domestication in southern Africa. Indeed, nutritional studies on *A. digitata* in Malawi have shown that baobab nutritional value is high for the supply of ascorbic acid, fat, carbohydrates, phosphorus, calcium, magnesium, iron, potassium and sodium all important for mankind (Saka et al. 1992; Saka and Msonthi 1994; Saka et al. 2008).

The supply of *A. digitata* in Malawi continues to rely on wild populations which are unfortunately dwindling due to various factors (Chirwa et al. 2006**)**. However, sustainable supply of baobab products can only be achieved through planting (Shivcharn and Gunnar 2004). In order to succeed in baobab domestication, an understanding of genetic variation is important (Zobel and Talbert, 1984).

Genetic diversity is the basis of all biodiversity and is a key requirement for the long term survival of a species especially in environments subject to climatic change or introduction of new pest, pathogens and competitors (Pakkad et al. 2008). Knowledge of the genetic variation within the species is essential to design a strategy to promote the use and conservation of indigenous fruit trees meant for on-farm cultivation (Haq et al. 2008). Genetic divergence information is also required in identifying parents for hybridization programmes intended to identify more heterotic recombinants (Sandhu et al. 2006). With large geographical distribution of baobab, morphotypes have developed (Sidibe and Williams 2002). Within the species, there is evidence showing the occurrence of a number of local forms differing in habit, size, quality of the fruit and leaf vitamin content (Assogbadjo et al. 2009). In addition, in Mali and Sudan variation in tree bark colour, pulp taste, tree height and diameter and leaf vitamin C content have been reported. Sidibe and Williams (2002) have proposed assessment of patterns of genetic diversity of baobab in relation to local distribution. Genetic variation among and within provenances of different species have been evaluated through seedlings in several studies (Mwase et al. 2007; Mwitwa et al. 2007, 2008; Ngulube et al. 1997). This study was undertaken to characterize, quantify the genetic variation and estimate the genetic control of *Adansonia digitata* seed germination and seedling growth characteristics in five natural populations existing in four diverse ecological conditions using a nursery study.

4.2 Methodology

4.2.1 Study area

A total of 59 open pollinated seedlots (half-sib families) from Karonga, Likoma Island, Salima, Mwanza and Chikwawa were selected on the basis of differences in silvicultural classification of Malawi (Hardcastle 1978) (Table 4.1, Fig.4.1). Likoma Island was included because of its isolation. According to Hardcastle (1978), silviculture zone L has mean annual rainfall (MAR) >1600 mm with predominantly weathered ferralitic soils. Silviculture zone Ba has MAR ranging between 710 to 850 mm and characterized by calcimorphic soils overlaying vertisols. Silviculture zone J has MAR ranging between 1200 to 1600 mm and characterized by ferrallitic soils whilst Silviculture zone A has MAR ranging between 710 to 840 mm with vertisols. Seed was collected from randomly selected parent trees located at distances of approximately 100 m apart.

4.2.2 Nursery evaluation

The nursery experiment was carried out at Mzuzu University green house from 1st October, 2008 consisting of 59 treatments made up of open pollinated seedlots (59 families) nested in five provenances. The experiment was laid out as a complete randomized block design with four replications consisting of twenty five black polythene tubes as a treatment unit. The rooting medium was dark-grey miombo soil mixed with sand in the ratio of 2:1 respectively. Prior to sowing, seed pre-treatment involved nicking by removing a small part of seed coat. Two seeds were sown per tube at a depth of about 4 cm. After 30 days, seedlings were thinned to one per tube. Water was applied when necessary to keep the medium moist.

Table 4. 1 Physical description of sites (populations/provenances) and number of trees (families) sampled

Site (Population/ Provenance)	Number of sampled families	Population code	Zone	Average annual Stress Period(weeks) *	Altitude (m)	Mean annual rainfall (mm)	Mean annual temperature (^{0}C)	Soil
Karonga	10	K	L	7" Field capacity: 15 12" Field capacity": 10	475-1000	>1600	23-25	Ferrisols dominant ; regosols
Salima	15	S	Ba	7" Field capacity:28 12" Field capacity: 26	200-1200	710-850	20-25	Alluvial calcimorphic soils above the vertisols
Mwanza	15	M	J	7" Field capacity:16 12" Field capacity: 11	900-1500	1200-1600	19-21	Sandy ferrallitic
Chikwawa	15	C	A	7" Field capacity:35 12" Field capacity: 35	<200	710-840	>25	Vertisols
Likoma Island	10	L	L	7" Field capacity: 15 12" Field capacity": 10	475-1000	>1600	23-25	Ferrisols, alluvial calcimorphic, regosols, lithosols

Source: Hardcastle, 1978.
* = Information on average annual stress calculations are reported by Hardcastle (1978)

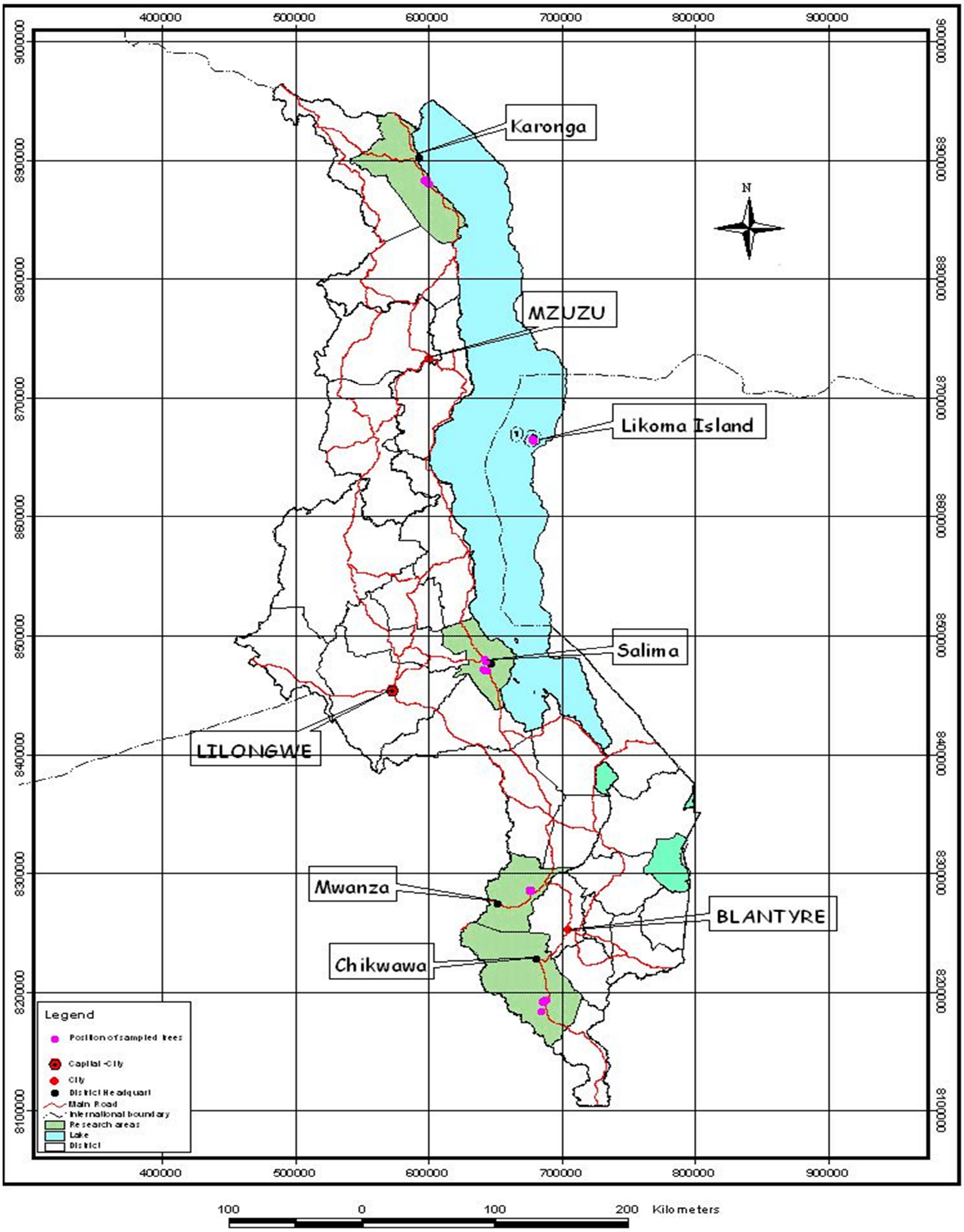

Fig. 4. 1 Map of Malawi showing 5 sampled populations of *Adansonia digitata*

4.2.3 Data collection

Seed was considered germinated when two true leaves appeared on the surface of the growing medium. Germinated seed were recorded daily for 30 days. At the age of three months all the seedlings were assessed for the number of leaves, root collar diameter, length and area of third leaf, total leaf area, and seedling height. At five months, only five seedlings were systematically selected from each treatment by sampling the first five seedlings in a row. To determine root parameters, seedlings were removed from the polythene tubes and soil washed carefully off the root system and left to dry before weighing. The root (tuber) and shoot were separated at the root collar. The seedlings were assessed for fresh tuber weight, tuber diameter, shoot diameter and height.

4.2.4 Data analysis

Data were analysed as randomized complete block using the nested effects model. Arcsine transformation was done for germination percentage (Fowler et al. 1998) prior to analysis.

Least Significant Difference (LSD) was used to identify significant differences between provenances and families. Estimates of narrow-sense heritability (h^2) were calculated from the variance components, obtained from the decomposition of the mean squares obtained from analysis of variance procedure. Narrow-sense heritability was calculated according to the formula given by Zobel and Talbert (1984) as follows:

$$h^2 = \frac{\sigma_A{}^2}{\sigma_p{}^2} = \frac{4\sigma_f{}^2}{(\sigma_w{}^2 + \sigma_f{}^2)} \qquad \text{(Equation 1)}$$

Where coefficient of relationship was taken as 0.25 for half-sib families, σ^2_f the variance between families, σ^2_w the variance within families and σ^2_p total phenotypic variation.

The additive genetic coefficient of variation (AGCV) values were calculated using the formula given by Cornelius (1993) as follows:

$$AGCV = 100(\sigma_A/\mu) \qquad \text{(Equation 2)}$$

Where σ_A is the additive genetic standard deviation and μ the phenotypic (and genotypic) mean.

4.3 Results

4.3.1 Provenance variation in germination percentage

In the nursery, seed germination commenced within nine days after sowing for all populations. At 14 days, almost all viable seed had germinated in all populations. Seed germination was significantly different ($p \leq 0.001$) among provenances, with Chikwawa having the highest germination (68.7 ± 3.3 %) and Karonga having the lowest germination (46.4 ± 3.3%) at 30 days after sowing (Fig. 4.2).

4.3.2 Family variation in germination percentage

At 30 days after sowing, germination percentage was significantly different ($p \leq 0.001$) among families within provenance (Fig.4.3). Across all the provenances, mean germination percentage ranged from 6.3 ± 8.6% in one tree from Mwanza (M8) to 95.6 % ± 8.6 % in another tree in the same population (M6) (Fig.4.3). Within Chikwawa C3 (85.4±8.6%) and C5 (85.6±8.6%) were superior to the rest in germination whilst the least germination was observed in C15 (36.5±8.6%). Similarly, M6, M13, M15 had superior germination percentage (ranging from 84.8±8.6 to 95.6±8.6%). M8 was the most inferior with germination of 6.3±8.6%, but not significantly different from M5 and M11 both at 16.4±8.6%. In Salima, S2, S6 and S3 had superior germination ranging from 85.4±8.6 to 91.7±8.6% whereas the least germination was found in S4 (11.4±8.6%) and S7 (18.9±8.6%). In Likoma, L3 (95.2±8.6%) had the most outstanding germination with L9 (32.1±8.6%) having the least. In Karonga K12 (84.2±8.6%) was the best performer whilst K14 (13.9±8.6) was the least. However, K7, K13, K2 and K11 had poor germination as well ranging from 28±8.6 to 45.3±8.6%

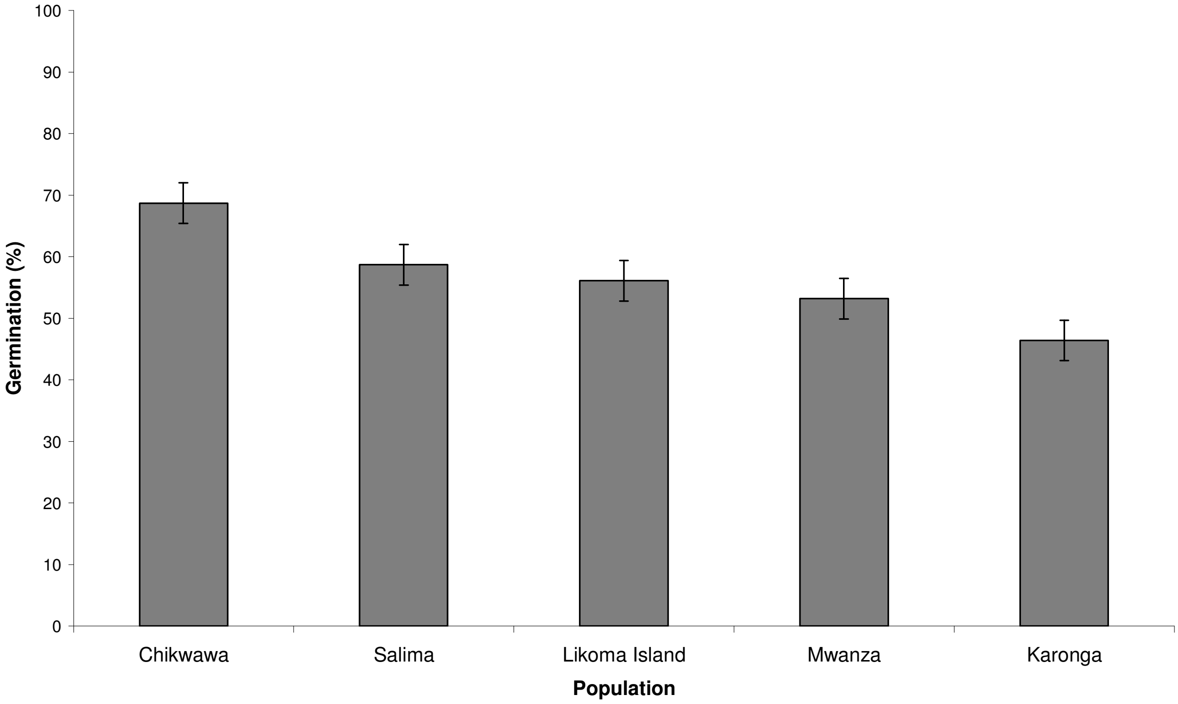

Fig. 4. 2 Variation in mean germination percentage among five populations of *Adansonia digitata*

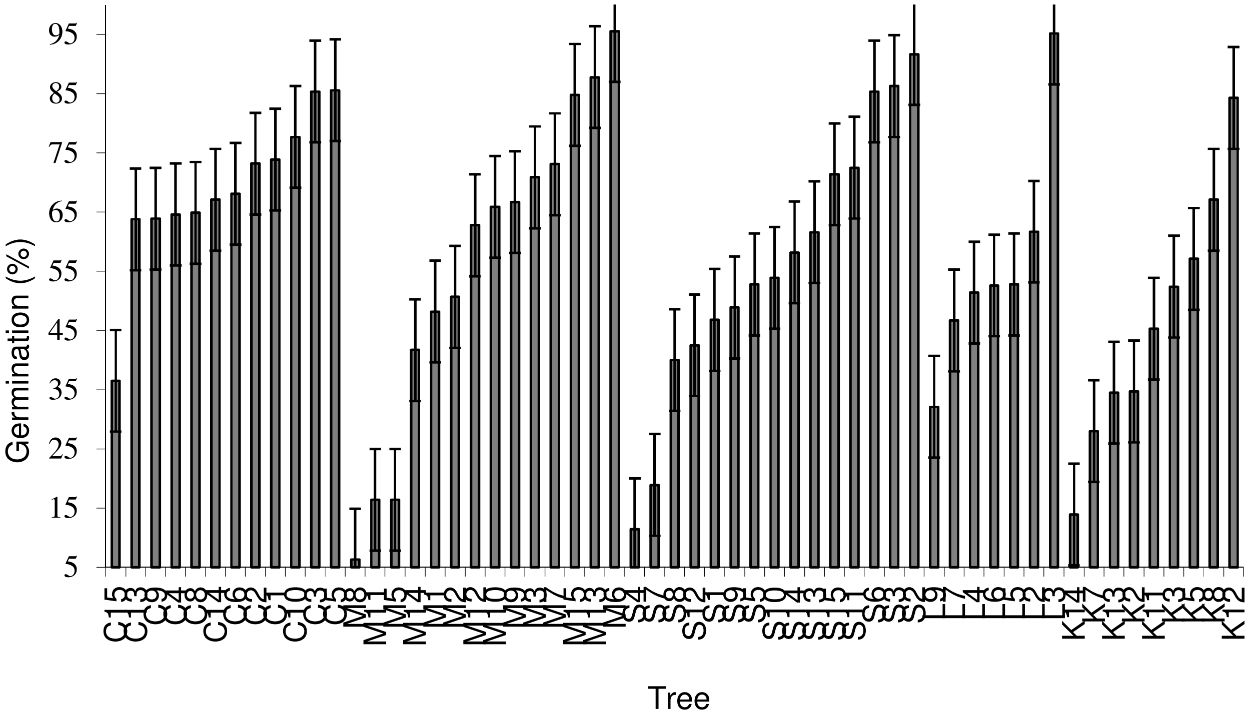

Fig. 4. 3 Variation in mean germination percentage between trees within family and among five provenances of *Adansonia digitata* at 30 days after sowing (C=Chikwawa, M=Mwanza, S=Salima, L=Likoma Island and K=Karonga; label of individual trees range from 1 to 15)

4.3.3 Provenance variation in seedling growth traits

After germination, most of the seedlings developed vigorously; although a few seedlings were nibbled by grass hoppers soon after germination. At three months after sowing, there were significant differences ($p \leq 0.001$) in plant height among provenances, with Karonga (16.7± 0.246 cm) having the tallest plants whilst the shortest were from Likoma Island (13.9 ± 0.256 cm), the rest of the populations were intermediate and were not significantly different (Table 4.2). Root collar diameter was significantly different ($p \leq 0.001$) among provenances, with Karonga (5.1±0.060 mm), Salima (5.1±0.048 mm), Chikwawa (5.1±0.045 mm) and Mwanza (5.1±0.046 mm) having the largest diameter whilst the smallest diameter were from Likoma Island (4.5±0.063 mm). The number of leaves on the whole seedling was significantly different ($p \leq 0.001$) among provenances with Karonga having the largest number of leaves (8.7±0.124) whilst Chikwawa (7.6±0.092), Mwanza (7.5±0.095) and Likoma Island (7.8±0.124) had the least. At five months after sowing, shoot height was significantly different ($p \leq 0.001$) among provenance, with Karonga (17.9±0.325 cm) and Salima (17.9±0.258 cm) having the tallest seedlings whilst the shortest were from Chikwawa (16.5±0.258 cm), Mwanza (15.8±0.263 cm) and Likoma Island (16.0±0.357 cm). Tuber weight was significantly different ($p \leq 0.001$) among provenances, with Chikwawa (21.8±0.574 g) having the heaviest whilst Likoma Island had the lightest (16.2±0.724 g). Tuber diameter was significantly different ($p \leq 0.001$) among provenances, with Chikwawa having the largest diameter (21.8±0.265 mm), whilst Karonga, Salima, Mwanza and Likoma were intermediate (ranging from 20.0±0.271 to 20.4±0.270 mm). Shoot diameter was significantly different ($p \leq 0.001$) among provenances with Karonga, Salima, Chikwawa and Mwanza having the largest diameter ranging from 5.3±0.113 to 5.6±0.111 mm whilst Likoma Island had the smallest (4.8±0.154 mm).

Table 4. 2 Mean plant height (cm), root collar diameter (rcd, mm) and number of leaves after three months; and shoot (cm) and tuber diameter (mm) and weight (g) after five months after sowing

Provenance	**Plant height** (cm)	Plant rcd (mm)	Number of Leaves	Shoot height (cm)	Tuber weight (g)	Tuber diameter (mm)	Shoot diameter (mm)
	3 months after sowing			5 months after sowing			
Karonga	16.7±0.24a	5.1±0.06a	8.7±0.12a	17.9±0.32a	20.7±0.72b	20.1±0.33b	5.4±0.14a
Salima	16.1±0.19a	5.1±0.04a	8.3±0.09b	17.9±0.25a	20.7±0.57b	20.3±0.26b	5.6±0.11a
Chikwawa	14.8±0.18b	5.1±0.04a	7.6±0.09c	16.5±0.25b	23.9±0.57a	21.8±0.26a	5.4±0.11a
Mwanza	14.5±0.18b	5.1±0.04a	7.5±0.09c	15.8±0.26b	21.5±0.58b	20.4±0.27b	5.3±0.11a
Likoma	13.9±0.25c	4.5±0.06b	7.8±0.12c	16.0±0.35b	16.2±0.79c	20.0±0.27b	4.8±0.15b
LSD	0.616	0.063	0.311	0.864	1.93	0.890	0.307
CV%	25	18.6	24.1	24.5	43.6	20.6	33.3

Means followed by the same letter are not significantly different

4.3.4 Family variation in seedling traits

Variation in seedling height within all provenances was significantly different ($P \leq 0.001$) among the families (Fig. 4.4). Across all provenances, seedling height ranged from 10.9 ± 0.52 cm in one tree in Mwanza (M8) to 19.4 ± 0.52 cm in another tree in Salima (S5). Superior growth in

Chikwawa was observed in C7, C16 and C2 ranging from 17±0.52 to 17.9±0.52 cm with C15 (11.9±0.52 cm) being the most inferior. The most outstanding growth in Mwanza was found in M15, M7, M3 and M12 ranging from 15.2±0.52 to 16.1±0.52 cm whilst M8 (10.9±0.52 cm) was the most inferior. In Salima the best performer was S5 (19.4±0.52 cm) while S15 (12.7±0.52) and S14 (13.3±0.52 cm) were the worst. In Likoma the most outstanding growth was found in L7 (15.6±0.52 cm) with L2 and L4 being the poor performers at 12.5±0.52 cm. In Karonga superior growth was observed in K3 (18.7±0.52 cm) and K5 (18.2±0.52 cm) whilst K14 (13.4±0.52 cm) was the most inferior.

Variation in tuber weight within all provenances was significantly different ($P \leq 0.001$) among the families (Fig. 4.5). Across all the provenances, tuber weight ranged from 14.4 ± 1.5 g in one tree in Likoma Island (L5) to 30.9 ± 1.5 g in another tree in Chikwawa (C7). Within Chikwawa, outstanding growth was observed in C7 (30.9±1.5g) and C8 (30.8±1.5g) whilst the inferior growth was found in C2, C3 and C16 ranging from 17.9±1.5 to 19.4±1.5g. Within Mwanza, superior growth was observed in M2, M5, M13, M1 and M11 ranging from 23.3±1.5 to 26.2±1.5g whereas the inferior growth was found in M6, M3, M10 and M14, ranging from 16.2±1.5 to 18.6±1.5g. In Salima, superior growth was found in S15 (25.9±1.5g) whereas S5, S10 and S2 were inferior ranging from 14.2±1.5 to 16.5±1.5g. Within Likoma, the best growth was observed in L6 (19.3±1.5g) whereas the least was in L5 (14.4±1.5g) and L9 (14.7±1.5). In Karonga the outstanding growth was found in K14 (21.4±1.5g) whilst the worst was in K11 (15.1±1.5g).

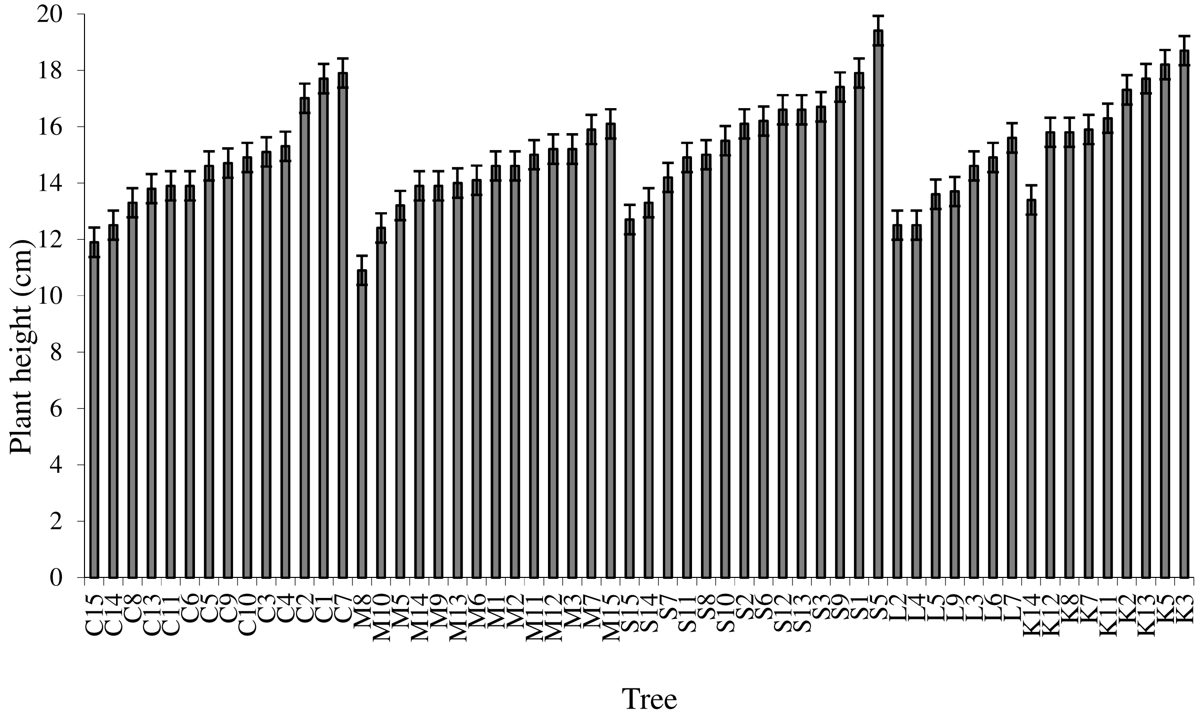

Fig. 4. 4 Family variation in plant height within five provenances in *Adansonia digitata* three months after sowing (C=Chikwawa, M=Mwanza, S=Salima, L=Likoma Island and K=Karonga; label of individual trees range from 1 to 15)

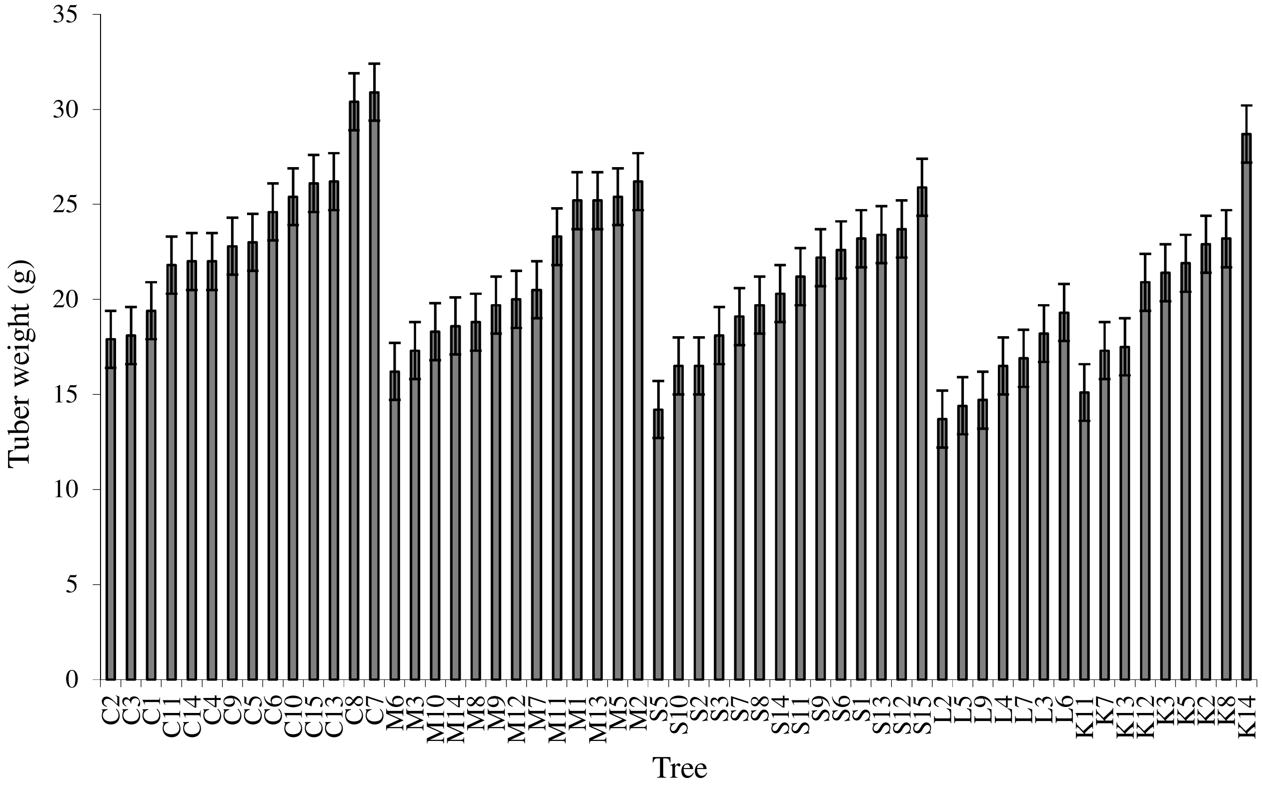

Fig. 4. 5 Family variation in tuber weight within five provenances in *Adansonia digitata* five months after sowing (C=Chikwawa, M=Mwanza, S=Salima, L=Likoma Island and K=Karonga; label of individual trees range from 1 to 15)

4.3.5 Phenotypic and genetic variation and control of seedling growth parameters

Results in Table 4.3 show genetic control and variance components of seedling traits. Strong additive gene effects was found for germination percentage (h^2= 0.53), plant height (h^2= 0.71) total leaf area (h^2= 0.65) and number of leaves (h^2= 0.66) 36 days after sowing but decreased as time elapsed. Moderate additive gene effects was found in height to first leaf (h^2=0.44), tuber weight (h^2= 0.34), third leaf area (h^2= 0.32), tuber diameter (h^2= 0.20) and root collar diameter (h^2= 0.15). Shoot diameter showed the lowest genetic control (h^2= 0.07). Within population variation ($\sigma^2_{w)}$ ranged from 87.2% to 96.4% of the total variation (σ^2_p) for all the traits. AGCV was moderate for germination percentage (11.2 %), tuber diameter (12.8 %), total leaf area (12 %), and third leaf area (14.7%) whilst that for the other traits could be considered low (< 10).

Table 4. 3 Values of total phenotypic variance (σ^2_p), family variance ($\sigma^2_{f(p)}$), within family variance (σ^2_w), individual-plant- narrow sense heritability (h^2) and additive genetic coefficient of variation (AVCV) for 13 variables in *Adansonia digitata* in Malawi

Variable	σ^2_p	σ^2_w	$\sigma^2_{f(p)}$	h^2	AGCV
Germination percentage	0.031	0.027	0.004	0.53	11.16
Five months after sowing					
Shoot diameter	3.150	3.097	0.054	0.07	4.36
Shoot height	17.409	15.735	1.674	0.38	7.71
Tuber weight	86.233	78.943	7.290	0.34	12.80
Tuber diameter	17.366	16.507	0.859	0.20	4.49
Number of leaves	3.718	3.480	0.238	0.26	6.16
Plant height	14.658	13.508	1.150	0.31	3.21
3 months after sowing					
*Third leaf length	0.555	0.5180	0.036	0.26	8.30
*Third leaf area	70.998	63.526	6.472	0.32	14.67
Total leaf area	968.209	811.374	156.835	0.65	12.04
Number of leaves	2.438	2.035	0.403	0.66	10.07
Plant height	700.870	576.579	124.295	0.71	9.00
Root collar diameter	0.877	0.845	0.032	0.15	3.77
Height to first leaf	70.203	62.470	7.733	0.44	9.51

*Third leaf length and area included due to its uniqueness (peculiar) in form

4.4 Discussion

4.4.1 Germination percentage

The prevailing differences in provenance and families in germination in the nursery investigation were essentially genetic in origin (Sniezko and Stewart 1989). Germination for all populations and families started 9 to 11 days after sowing and within 14 days most of the viable seed had germinated. In this case seed origin seems to have had no influence on the initiation of germination. Since days to germination were the same, this may imply that the population origin is the same and that these populations have so far not evolved differently for this trait (Andersen et al. 2008). Thus days taken to start germination have failed to characterize the population into different ecological (silvicultural) zones. It is anticipated that high-vigour seeds germinate faster than low-vigour seed under any condition (Schmidt 2000). In the current study, germination capacity was poor mostly for Karonga population which is mainly attributed to fruit handling. However, mean germination was generally high for the other populations and most of the families within populations (Figs. 4.2 & 4.3). The results indicate need for careful post handling of fruits to retain high seed physiological quality (viability). However, some families such as M8, M11, M5, S4 and S7 (Fig.4.3) had poor germination despite proper fruit and seed handling. It may be inferred that the origin of the problem was maternal. Poor germination may be a sign of inbreeding (Zobel and Tarbert 1984). According to Andersen et al. (2000) most of the variation in germination in *Abies guatemalensis* was due to proportion of seeds with viable embryos. Effectiveness of pollinators and breeding system has an impact on viability of embryos (Zobel and Tarbert 1984). *Adansonia digitata* L is outcrossing (Assogbadjo et al. 2006) but tree density in Malawi is dwindling such that in most populations there may be only two (2) adult trees per hectare (Chirwa et al. 2006) increasing chances of related mating. Variation studies in germination between and within populations in miombo species including baobabs are still scanty. Chithila et al. (2008) found variation in germination between four baobab provenances in Malawi ranging from 0 to 60% depending on method of pretreatment. Munthali (2007) reported commencement of germination in Malawian baobabs at 7 days after sowing and germination percentage ranging from 23 to 75%, depending on pretreatment method. ICUC

(2002) reported that one is unlikely to have 100% viability in baobab but that healthy seeds should give 70-85% germination. In this study, some families achieved up to 95% germination showing high physiological quality. Published work in other miombo species such as in *Uapaka kirkiana* (Mwase et al. 2007; Ngulube et al. 1996) have reported significant variation in seed germination between indigenous populations in Malawi. The variation has been attributed to genetic as well as environmental factors. The presence of significant population and family variation in germination percentage shows need for proper selection of seed source. The differences in germination in the present study likewise could be linked to family variation (genetic), fruit maturity at collection, fruit and seed handling and original ecological factors (Schmidt 2000). Sustainable supply of indigenous fruits can only be attained through planting (Akinnifesi et al. 2006). Highest quality transplants are required in domestication programmes; hence high germination capacity is essential (Ngulube et al. 1997).

4.4.2 Seedling growth

Understanding variation existing in the species is prerequisite before starting any tree breeding programme (Zobel and Tarbert, 1984). A number of population genetics studies through nursery studies have been done (Andersen et al. 2008; Marcar et al. 2002; Masamba et al. 1999; Mwase et al. 2007; Mwitwa et al. 2008; Munthali 1999; Ngulube 1996; Sniezko and Stewart 1989). Variation in seedling traits during the nursery phase is usually considered genetic in origin (Ngulube et al. 1997). The study showed that there were substantial variation in shoot and root traits between provenances and families in baobabs. The clusters of tuber weight, seedling diameter and plant height vividly showed that main land populations are similar genotypes while the one from the Likoma Island is different (Table 4.2). The clusters also appeared to show that seed sources from drier sites (with long duration of stress period) (Table 4.1) had generally vigorous growth, for instance Chikwawa was mostly superior in most growth traits while Likoma Island was mostly underperforming in majority of the traits. According to Ngulube et al. (1997), variation in plant biomass discloses overall differences in growth ability among populations. Generally, Chikwawa from the lowest altitude had the most outstanding growth in most traits. Similarly, Andersen et al. (2008) have reported superior growth in populations from low altitude areas compared to high altitude area. The results show that farmers who would like to establish nurseries for production of baobab root tubers (carrots) should choose the Chikwawa population as a first choice followed by the other mainland populations and avoid the Likoma Island population due to its inferiority. Marcar et al. (2002) have shown that better performing *Eucalyptus grandis* provenances in glasshouse trials also performed equally well in the field trials. It may also follow therefore that if the biomass at seedling stage is maintained up to later stages, as expressed through the differences in biomass and its partitioning in this study, then this might already be an indication of populations that will have superior growth later (Ngulube et al. 1997). The baobab populations may also be having different net assimilation capacity leading to variation in seedling parameters. The differences found could be attributed to strong natural selection and ecological barriers to gene flow (Sorensen et al. 1990). However, provenance and family trials are still required to assess juvenile and mature correlations and genotype x environmental interaction (Chirwa et al. 2007; Zobel and Tarbert 1984). Notwithstanding, present results might project variation in fruit traits in future as well. These nursery results depict possibility of identifying population variation easily and cheaply useful for countries that can not afford molecular studies. The current study compares well with studies done on baobab in Benin (Assogbadjo et al. 2006) where morphometric analysis showed significant differences within and among populations across the climatic zones largely influenced by abiotic factors (temperature and rainfall). Variation in seedling growth traits have been associated with distance between locations, elevation and latitude as well which are linked with adaptative variation of species (Sorensen et al. 1990). Subsequently, clusters found in present results in different traits might be an indication that populations in baobabs exists as ecotypes partitioned by both climatic and geomorphological factors. This study has been done at national level but *A. digiata* occurs at extremely wide geographical scale. Hence, it will be important to study the pattern of genetic variation at regional scale (southern Africa) to design domestication and tree improvement activities. High genetic variation found between and within populations in all traits as shown by high coefficient of variation means that for tree improvement to succeed, selection should be at both population and family level (within population).

4.4.3 Heritability

Narrow-sense heritabilities provide a basis for predicting the genetic gain that could be achieved from selection on a particular trait (Marcar et al. 2002). The estimates in Table 4.3 have shown that there were considerable differences in genetic control of traits. For instance, germination percentage, total leaf area, number of leaves, plant height and height to first leaf were more strongly genetically influenced. Only shoot diameter and root collar diameter showed weak additive genetic effects (0.07 to 0.15) but the rest of the traits have moderate genetic control. Low heritability values mean that phenotype is a weak predictor of genotype (Marcar et al. 2002). There were fluctuations in value of additive genetic effect as seedlings were approaching dormant stage in growth. Apical dormancy in seedlings of *Pterocarpus angolensis* has been reported (Mwitwa et al. 2007) and has been attributed to seasonal changes in climate that has effects on the physiological response of plants. High heritability values indicate that much of the variation for a given trait observed in a population had a genetic origin (Zobel and Talbert, 1984). Strong and intermediate genetic control is favourable for selection in breeding programmes as it shows breeding programmes need not use large numbers of families to attain high genetic gains. The variance components have shown that much of the variation in most traits was explained by the within- family variation meaning that selection intensity will be high at this level. The results are supported by Zobel and Talbert (1984) who have indicated that more than 90% of the variation in forest trees resides within families. AGCV values have indicated existence of moderate genetic diversity in germination percentage, tuber weight, total leaf area, third leaf area, and number of leaves. The high values could mean preponderance of heterozygosity in the population which is favourable for evolution since it favours high genetic diversity (Zobel and Talbert, 1984). There is generally scanty information on genetic information in most tropical tree species. Mwitwa et al. (2007), reported narrow sense heritability for seventeen shoot and root variables ranging from low to moderate (0.09 to 0.35); and also in shoot die back in *Pterocarpus angolensis* of 0.07 in the first growing season and 0.42 in the second season (Mwitwa et al. 2008). These values are similar to those from this study; only that some of the traits in this study have high heritability. Furthermore, heritability has been shown to change with season or time (Mwitwa et al. 2007); which has also been depicted in this study. This may imply that selection should be done at the time plants are vigorously growing.

These results also compare well with reported work by Cornelius (1993) who analysed trends of heritabilities and AGCV for several tree species as follows: for heritability, mean ranged from 0.21 to 0.50 and median 0.19 to 0.48 while for AGCV the mean ranged from 5.34 to 23.10 and median 5.10 to 20.30. Sirisena and Senanayake (2000) reported similar values of heritability in banana clones ranging from 4 to 46% and genetic coefficient of variation ranging from 5 to12%. Hence, it can be implied that genetic control of traits in *Adansonia digitata* may be similar to other tree species. High and moderate heritabilities may be due to wide genetic variability of the germplasm belonging to different genomic groups (Sirisena and senanayake 2000). AGCV of greater than 10% indicate some promise for genetic improvement (Sirisena and Senanayake 2000). The high estimates of narrow-sense heritabilities coupled with considerable morphometric variation observed may indicate that superior gains would be attained in baobab from selection and breeding.

4.5 Conclusion

The initiation of seed germination has shown that baobab populations are genetically close. Even though, germination percentage showed significant differences between and within the populations, the trait can not be easily used to characterize the populations according to silvicultural zones. However, while seedling variations cannot be easily used to specifically delineate populations into definite ecotypes, the general trend emerging was that mainland trees were genetically distant to Likoma Island population and that the severity of stress period of the locations seemed to have further genetically diverged populations into ecotypes even on the mainland. These results are also important to tree breeders as they may detect variation at an early stage and guide in breeding; and the use of phenotypic variation may also provide a faster method of detecting variation between and within the populations particularly useful for countries that can not afford use of biotechnological methods in showing population diversity. The high genetic variation and genetic control of traits are encouraging results showing great potential of improving baobab through selection and breeding. The low germination capacity in some families despite good fruit and seed handling may be linked to inbreeding as a result of very low *Adansonia* tree densities in Malawi thereby promoting mating of close relatives. This is an area that should be critically researched since inbreeding has numerous harmful effects on fruit trees.

4.6 Acknowledgements

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4.7 Reference

Akinnifesi FK, Sileshi G, Ajayi OC, Chirwa PW Kwisiga FR, Harawa R (2008) Contributions of Agroforestry Research and Development to Livelihood of Smallholder Farmers in Southern Africa: 2. Fruit, Medicicinal, Fuelwood and Fodder Trees Systems. Agricultural Journal 3(1): 76-88

Akinnifesi FK, Kwesiga F, Mhango J, Chilanga T, Mkonda A Kadu CAC, Kadzere I, Mithofer D, Saka JDK, Silesh G, Ramadhani T, Dhliwayo P (2006) Towards the Development of Miombo Fruit Trees as Commercial Tree Crops in Southern Africa. Forests, Trees and livelihoods, Vol. 16:103-121

Andersen JP, Cordova JPP, Nielsen UB, Kollmann J (2008) Provenance variation in germination and seedling growth of *Abies guatamalensis* Rehder. Forest ecology and Management 255: 1831-1840

Assogbadjo AE, Kyndt T, Chadare FJ, Sinsin B, Gheysen G, Eyog-Matig O, Van Damme P (2009) Genetic fingerprinting using AFLP cannot distinguish traditionally classified baobab morphotypes. Agroforest Syst 75: 157-165

Assogbadjo A.E, Kyndt T, Sinsin B, Gheysen G, Van Damme P (2006) Patterns of Genetic and Morphometric Diversity in Baobab (*Adansonia digitata*) Populations Across Different Climatic Zones of Benin (West Africa). Annals of Botany 97: 819-830

Birot Y, Christophe C (1993) Genetic structure and expected genetic gains from multitrait selection in wild populations of Douglas fir and Sitka spruce. Silvae Genetica,32, 5-6

Campbell, B., Luckert, M., Scoones, I., 1997. Local Level Valuation of Savanna Resources:
A Case Study from Zimbabwe. Economic Botany 51:59-77

Chirwa M, Chithira V, Kayambazinthu D, Dohse C (2006) Distribution and Population Structures of *Adansonia digitata* in Some Parts of Ntcheu, Dedza and Mangochi Districts, Malawi. FRIM, Zomba, Malawi

Chirwa PW, Bwanali RJ, Meke, Sagona W, Munthali CRY, Mwabumba L (2007) Growth and phenology of a three-to four-year-old *Sclerocarya birrea* international provenance trial in Malawi. Southern hemisphere Forestry Journal, 69(1):49-54

Chithira V, Tembo C, Namoto M (2008) The germination of baobab (*Adansonia digitata* L.) seeds 90 days after storage. FRIM Report No. 08001. ODC: 181.525

Cornelius J (1993) Heritabilities and additive genetic coefficients of variation in forest trees. Canadian.Journal Forestry Research. Vol.24: 372-379

Fowler J, Cohen P, Jarvis P (1998) Practical statistics for field biology. John Wiley and Sons, New York

Gebauer J, El-Siddig K, Ebert G (2002) Baobab (*Adansonia digitata* L.): a Review on a Multipurpose Tree with Promising Future in the Sudan. Gartenbauwissenschaft, 67(64).S. 155-160

Gruenwald J, Galiza M (2005) Market brief in the European Union for selected natural ingredients derived from native species: *Adansonia digitata* L. Baobab.UNCTAD/BioTrade Facilitation Programme. Nertherlands and Switzerland.

Haq C, Bowe C, Dunsiger (2008) Challenges to Stimulating the Adoption and Impact of Indigenous Fruit Trees in Tropical Agriculture. In: AKinnifesi FK, Leak RRB, Ajayi OC, Silesh G, Tchoundjeu Z, Matakala P, Kwesiga FR. (Eds). Indigenous Fruit Trees in the tropics Domestication Utilisation, and Commercialization. Columns Design Ltd, Reading, UK

Ham C, Akinnifesi FK (2006) Zimbabwe, Zambia, Malawi and Tanzania: priority fruit species and products for tree domestication and commercialization. IK Notes No.94

Hardcastle PD (1978) A preliminary silvicultural classification of Malawi. Forestry Research Institute of Malawi. Forestry Research Record No. 57

ICUC (2002) Fruits for the Future, Baobab. International Center for Underutilized crops. Factsheet No.4, Southampton, UK

Kalaba FK, Chirwa PW, Syampungani S, Ajayi OC (2010) Contribution of agroforestry to biodiversity and livelihoods improvement in rural communities of Southern African regions. In: Teja Tscharntke, Christoph Leuschner, Edzo Veldkamp, Heiko Faust, Edi Guhardja, Arifuddin Bidin (Eds.).Tropical rainforests and agroforests under global change.Springer Berlin.

Kyndt T, Assogbadjo AE, Hardy OJ, Kakai RG, Sinsin B, Van Damme P, Gheysen G (2009) Spatial Genetic Structuring of Baobab (*Adansonia digitata*, Malvaceae) in the traditional Agroforestry systems of West Africa. American Journal of Botany 96(5): 950-957

Marcar NE, Crawford DF, Saunders A, Matheson AC, Arnold RA (2002) Genetic variation among and within provenances and families of *Eucalyptus grandis* W. Hill and E. globules Labill. Subsp. Globules seedlings in response to salinity and waterlogging. Forest Ecology and Management 162: 231-249

Marcar NE, Crawford DF, Saunders A, Matheson AC, Arnold RA (2002) Genetic variation among within provenances and families of *Eucalyptus grandis* in response to salinity and waterlogging. Forest Ecology and Management 162: 231-249

Masamba CRL (1999) Factors influencing accelerated seedling tree growth in Malawian provenances of *Faildherbia albida* Del. Dissertation , Wye College, University of London

Mithöfer D, Waibel H, Akinnifesi FK (2006). The role of food from natural resources in reducing vulnerability to poverty: A case study from Zimbabwe. Working Paper 2006 No. 1, 13 pp. Development and Agricultural Economics. Faculty of Economics and Management, University of Hannover, Germany

Munthali AM (2007) Seed preatment and germination of *Adansonia digitata* L. Dissertation, Mzuzu University

Munthali CRY (1999) Seed and seedling variation of *Pterocarpus angolensis* DC from selected natural populations of Malawi. Dissertation, University of Stellenbosch

Mwase WF, Bjornstad A, Ntupanyama YM, Kwapata MB, Bokosi JM (2006) Phenotypic variation in fruit, seed and seedling traits of nine *Uapaka kirkiana* provenances found in Malawi. Southern African Forestry Journal, No.208, 15-21

Mwitwa JP, Munthali CRY, van Wyk G (2007) Half-sib family variation in shoot and root traits of seedlings of *Pterocarpus angolensis* (Family: Fabaceae; syn. Papilionaceae). Southern Hemisphere Forestry Journal, 69(2): 91-94

Mwitwa JP, Munthali CRY, van Wyk G (2008) Heritability of shoot die-back and root biomass in sixteen *Pterocarpus angolensis* (Fabaceae) half-sib families from Malawi, Namibia and Zambia. Southern Hemisphere Forestry Journal, 70(3): 221-226

Ngulube MR, Hall JB, Maghembe JA (1997) Fruit, seed, and seedling variation in *Uapaka kirkiana* from natural populations in Malawi. Forest Ecology and Management 98: 209-219

Pakkad G, Ueno S, Yoshimaru H (2008) Genetic diversity and differentiation of *Quercus semiserrata* Roxb. in northern Thailand revealed by nuclear and chloroplast microsatellite markers. Forest Ecology and Management 255: 1067-1077

Palmer E (1997) A field Guide to the TREES of Southern Africa. William Collins Sons and Co Ltd. London

Saka JDK, Kadzere I, Ndabikunze BK, Akinnifesi FK, Tiisekwa BPM (2008) Product Development: Nutritional Value, Processing and Utilization of Indigenous Fruits from the Miombo Ecosystem. In: AKinnifesi FK, Leak RRB, Ajayi OC, Silesh G, Tchoundjeu Z, Matakala P, Kwesiga FR. (Eds). Indigenous Fruit Trees in the tropics Domestication Utilisation, and Commercialization. Columns Design Ltd, Reading, UK

Saka JDK, Msonthi JD (1994) Nutritional value of edible fruits of indigenous wild trees in Malawi. Forest Ecology and Management, 64, 245-248

Saka JDK, Msonthi J.D, Sambo EY (1992) Dry matter, acidity and ascorbic acid contents of edible wild fruits growing in Malawi. Trop. Sci, 32, 217-221

Sandhu JS, Singh P, Singh A (2006) Genetic divergence in chickpea in different environments. Tropical Science 46(1), 23-30

Schmidt L (2000) Guide to handling of Tropical and Subtropical Forest Seed. Danida Forest Seed Centre. Krogerupvej 21 DK-3050 Humlebaek, Denmark

Sidibe M, Williams JT (2002) Baobab. *Adansonia digitata* L. International Centre for Underutilised Crops, Southhampton, UK

Sirisena JA, Senayayake SGJN (2000) Estimation of variability parameters within 'Mysore' banana clones and their implication for crop improvement. Scientia horticulturae, 84:49-66

Shackleton C, Shackleton S (2004) The importance of non-timber forest products in rural livelihood security and as safety nets: a review of evidence from South Africa. South African Journal of Sciences 100:658-664

Shackleton S, Shanley P, Ndoye O (2007) Invisible but viable: recognizing local markets for non-timer forest products. International Forestry Review Vol. 9(3):697-712

Shivcharn SD, Gunnar G (2004) Local management practices influence the viability of the baobab (*Adansonia digitata* Linn.) in different land use types, Cinzana, Mali. Agriculture, Ecosystems and Environment 101: 85-103

Sniezko RA, Stewart HTL (1989) Range-Wide Provenance Variation in Growth and Nutrition of *Acacia albida* seedlings propagated in Zimbabwe. Forest Ecology and Management 27: 179-197

Sorensen FC, Robert KC, Jerry FF (1990) Geographic variation in growth and phenology of seedlings of *Abies procera*/*A. mannifica* complex. Forest Ecology and Management 36: 205-232

Zobel B, and Talbert J (1984) Applied Forest Tree Improvement. John Wiley&Sons, New York

5 Genetic differentiation and diversity of *Adansonia digitata* (Baobab) in Malawi using microsatellite markers

Abstract

Baobab (*Adansonia digitata*) belonging to Bombacaceae family, is one of the most widely used important indigenous` tree species in sub-Saharan Africa, valued in the cosmetic industry for its seed oil, and powdery fruit pulp for juice making. Because of its high potential for domestication in southern Africa understanding its genetic diversity and population structuring is warranted. The study investigated the level of genetic diversity and differentiation of five populations of *Adansonia digitata*. sampled from four diverse silvicultural zones in Malawi. Variation at nine microsatellite loci were examined in 150 individual trees. Low mean genetic diversity was expressed through genetic diversity indices: Nei's genetic diversity (h, 0.18±0.03), Shannon Information Index (I, 0.21±0.07), observed number of alleles (na, 1.47±0.10), effective number of alleles (ne, 1.23±0.04) and percentage polymorphic loci (pp, 48%). The low genetic variation found is attributed to the population growing in marginal areas of genetic centre of diversity of the species as well as climatic and anthropogenic factors. Moderate genetic differentiation were observed among populations (Gst = 0.13) probably due to large number of common alleles and long-distance gene dispersal resulting in a homogenisation effect. Clustering of individual trees by genetic similarity coefficients indicated that mainland trees were genetically closer than the trees on Likoma Island. The results suggest that seed distribution and tree improvement should recognise the presence of ecotypes and conservation measures should protect all the populations due to existence of private alleles which are of adaptive importance.

Key word: race; polymorphism; provenance selection; outcrossing

5.1 Introduction

Adansonia digitata (Baobab) has a huge geographical range spanning the drier parts of west Africa, east and south Sudan, drier parts in Angola across to Mozambique and up to northern South Africa (Guy 1971). The extensive distribution map of baobab is documented in Wickens (1982) and Sidibe and Williams (2002). Such a distribution should result in formation of distinct geographical races (Zobel and Talbert 1984) that are adapted to various ecological conditions. Hence, the need to explore patterns of genetic diversity in relation to baobab distribution (Sidibe and Williams 2002).

Molecular studies have been done in order to assess genetic diversity in baobabs (Assogbadjo et al. 2009; Kyndt et al. 2009; Pocktsy et al. 2009; Larsen et al. 2009). Assogbadjo et al. (2009) showed that there was genetic structuring and low to high genetic diversity between baobab populations in different climatic regions of Benin (West Africa). Kyndt et al. (2009) found high levels of genetic structuring present in baobabs at regional scale (Benin, Ghana, Burkina Faso and Senegal) and within-population level which was unexpected considering its dispersal by bats and human exchange of seed. However, Assogbadjo et al. (2009) using AFLP markers could not distinguish traditionally classified baobab morphotypes. Pocktsy et al. (2009) established that the tetraploid *A. digitata*, or its diploid progenitor originated in West Africa and migrated subsequently throughout the continent, and beyond, through natural and human–mediated terrestrial and overseas dispersal. Larsen et al. (2009) developed and tested eighteen microsatellite primers (SSR-primers) for tetraploid *Adansonia digitata* and its relatives showing different alleles per locus and different allele sizes. Most published results on baobab are predominantly from West Africa. There is, however, scanty published information on molecular studies for baobabs existing in southern Africa. Inspite of the paucity of genetic diversity information on baobab, domestication of some priority indigenous fruit species has been advanced in southern Africa (Akinnifesi et al. 2008). According to Larsen et al. (2009), it is pertinent to carry out gene flow studies in baobabs to provide insight into dispersal processes that shape the genetic structure. In addition they indicated that estimates of seed dispersal and differentiation between populations is vital for monitoring impacts from human influence and for forecasting consequences of climate change. Over time, baobab demography has been influenced substantially by anthropogenic factors (land -use pattern, trampling by domesticated livestock), climate (prolonged drought), elephant damage (Edkins et al. 2007; Wilson 1988), fire, clearing during cultivation, browsing (Chirwa et al. 2006) which have had adverse impact on genetic diversity. It is known that positive correlation exists among the levels of genetic diversity and fitness in plants (A´vila-di´az and Oyama 2007). For baobab domestication to succeed, it requires understanding of the genetic diversity since it is the fabric of evolution, the base material on which adaptation depends with high levels of genetic diversity considered as acceptable for conferring the ability to respond to threats such as diseases, parasites, predators and environmental change (Amos and Harwood 1998). The current study was undertaken to assess genetic diversity and differentiation in subpopulations of baobab sampled in different silvicultural zones in Malawi. The aim was to examine whether the delineation of silvicultural zones have subsequently structured the genetic composition of the trees substantially and also to estimate the gene flow among the populations using microsatellite markers.

5.2 Methodology

5.2.1 Sample collection and DNA extraction

The study populations were selected based on the silvicultural zones (A, Ba, L and J) delineated by Hardcastle (1978) (Table 5.1, Fig.5.1) based on climate and geomorphology. In addition, Likoma Island population, apart from occurring in silviculture zone L, was included due to its geographical isolation. According to Hardcastle (1978), silviculture zone L has mean annual rainfall (MAR) >1600 mm with predominantly weathered ferralitic soils. Silviculture zone Ba has MAR ranging between 710 to 850 mm and characterized by calcimorphic soils overlaying vertisols. Silviculture zone J has MAR ranging between 1200 to 1600 mm and characterized by ferrallitic soils whilst Silviculture zone A has MAR ranging between 710 to 840 mm with vertisols.

A total of 150 individuals, representing five wild populations (karonga, Likoma Island, Salima, Mwanza and Chikwawa) were sampled (Fig.5.1). Thirty randomly selected trees at a minimum distance of 100 m represented a population. Four young leaflets were collected, dried in the field and preserved in silica gel in sealed 300 ml plastic bottles. Total genomic DNA was isolated from leaf tissue following CTAB (Cetyltrimethyammonium Bromide) method (Gawal and Jarrent 1991).

Table 5. 1 Physical description of populations (site/provenance)

Populatio-n	Silviculture Zone	Average annual Stress Period (weeks)	Altitude (m.a.s.l)	Mean nnual rainfall (mm)	Mean annual temperature (^{0}C)	Soil
Karonga (pop1)	L	7" Field capacity: 15*, 12"Field capacity": 10	475-1000	>1600	23-25	Ferrisols dominant regosols
Likoma Island (pop2)	L	7" Field capacity: 15 12" Field capacity": 10	475-1000	>1600	23-25	Ferrisols, alluvial calcimorphic, regosols, lithosols
Chikwawa (pop3)	A	7" Field capacity:35 12" Field capacity: 35	<200	710-840	>25	Vertisols
Salima (pop4)	Ba	7"Field capacity:28 12" Field capacity: 26	200-1200	710-850	20-25	Alluvial calcimorphic soils above the vertisols
Mwanza (pop5)	J	7" Field capacity:16 12" Field capacity: 11	900-1500	1200-1600	19-21	Sandy ferrallitic

Source: Hardcastle (1978)

* = Information on average annual stress calculations are reported by Hardicastle (1978)

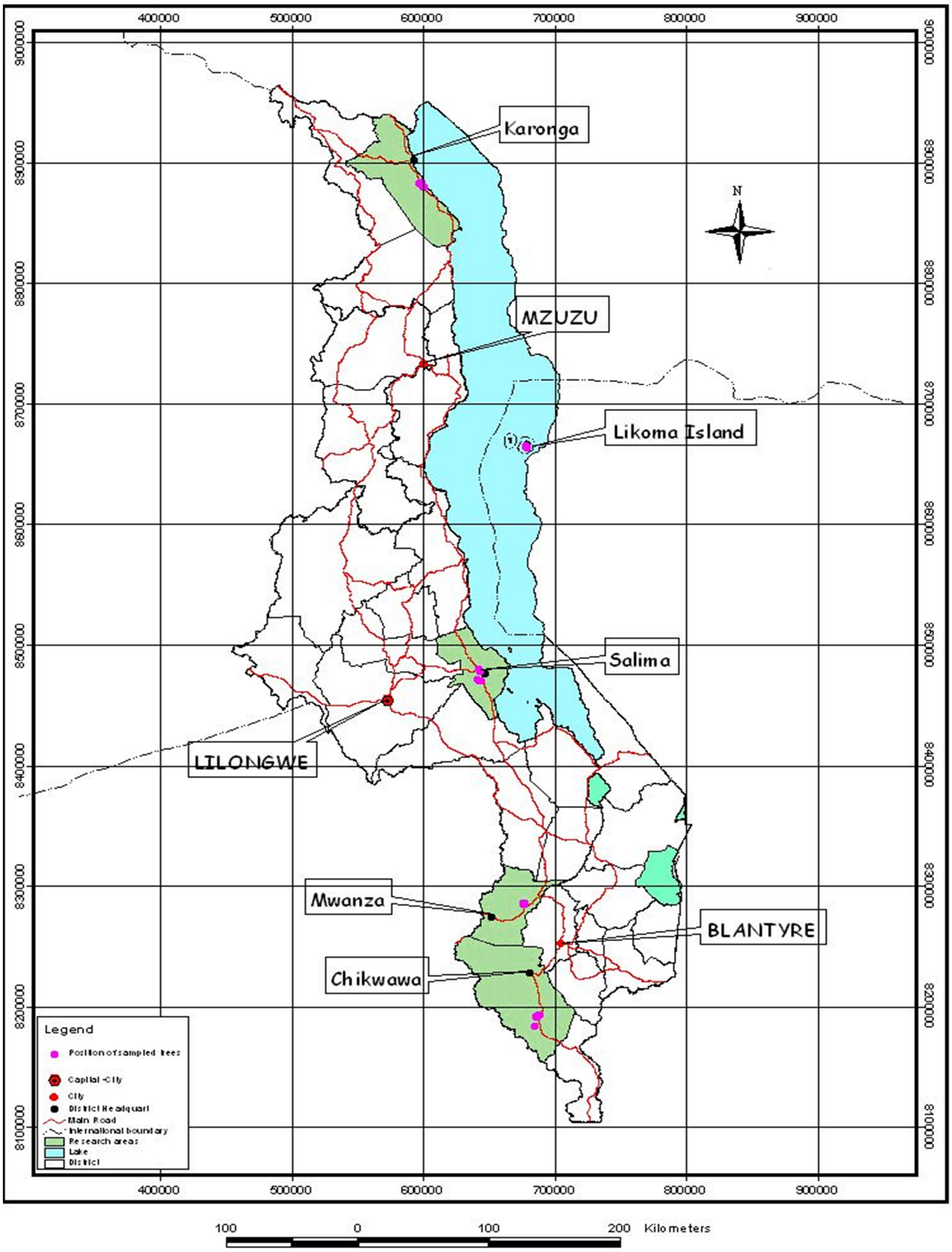


Fig. 5. 1 Map of Malawi showing location of Chikwawa, Mwanza, Salima, Likoma Island and Karonga sampled Adansonia digitata L populations

5.2.2 Microsatellite analysis

Primer sequences specific for nine microsatellite loci described by Larsen et al. (2009) were used in this study (Table 5.2). The Polymerase Chain Reaction (PCR) conditions were optimised for the nine microsatellite to produce scorable amplification products. PCR cocktail included the following reaction reagents: 5.7 µl PCR grade water (double distilled water), 1 µl of 10mM DNTP mix, 1.25 µl of 10 x PCR buffer, 1.6 µl of 25 mM Magnesium Chloride ($MgCI_2$), 0.75 µl of both forward and reverse *A. digitata* microsatellite primers, 0.06 µl of 5 u/µl Taq DNA polymerase in storage buffer and 2 µl of 25ng/ µl template DNA. Each reaction tube had final volume of 12.5 µl PCR master mix. PCR programme comprised ten cycles of amplification initialised with a denaturation step at 94 ^{o}C for 30 sec, annealing step at primer specific temperature for 15 sec (Table 2) and extension step at 72 ^{o}C for 30 sec. Another 30 cycles of amplification followed consisting of denaturing step at 89 ^{o}C for 30 sec, annealing step at primer specific temperature for 15 sec, extension step at 72 ^{o}C for 30 sec and final extension at 65 ^{o}C for 20 m. The soaking temperature was 4^{o}C. The PCR products were separated on 6% polyacrylamide gels stained with silver nitrate as described in Promega Silver Sequence™ DNA Sequencing System Technical Manual.

According to Giang et al. (2003), microsatellites as a co-dominant genetic marker enables detection of both homozygotes and heterozygotes that improves the insufficiency of AFLP and could be used for examining mating system. Five or six microsatellite loci are adequate to do genetic characterization of tree populations (Glaubitz and Moran 2000). Estimating the exact number of copies of individual alleles is difficult among polyploidy species. Therefore data is often analysed as a binary data matrix and SSR markers are treated as dominant markers (Changadeya 2009). Hence, the presence or absence of each PCR amplification product was scored as "1" or "0", respectively and data matrix was generated. Using the data matrix, POPGENE Version 1.31 freeware (Yeh et al. 1999) was used to calculate measures of genetic variation within and between baobab populations. The following variables were calculated: observed number of alleles (na), effective number of alleles (ne) (Kimura and Crow 1964), number of polymorphic loci (p), percentage of polymorphic loci (pp), Nei's genetic diversity (h) (Nei 1973) and Shannon's information index (I) (Lewontin 1974). Significance of the various genetic indices was determined by 95% Confidence Interval. The presence of zero in the interval meant no significant difference. Genetic differentiation (G_{st}), which measures among-population component of genetic variation was calculated to determine the proportion of total variation that was due to differences between population allele frequencies. Total heterozygosity (H_t), gene diversity of individuals relative to their population (H_s) and gene flow (N_m) were also determined. Dendrogram based on Nei's 1972 using the Unweighted Pair-group Method based on Arithmetic averages (UPGMA) modified from NEIGHBOR procedure of PHYLIP Version 3.5 was constructed. Further, Nei's unbiased measure of pairwise genetic identity and genetic distance (Nei 1978) were calculated. Individual tree similarity was analysed using NTSYSpc version 2.11c (Rolhf 2001). Pairwise similarity matrices were used to construct dendrograms from the Sequential Agglomerative Hierarchical and Nested (SAHN) clustering method using the Unweighted Pair-group Method with Arithmetical averages (UPGMA) (Sneath and Sokal 1973).

Table 5. 2 Microsatellites used in this study with loci name, forward and reverse sequences and annealing temperature

Locus name	Primer sequences 5'-3'		Annealing* temperature (°C)
	F	R	
Ad01	CATTGCCAGGAATGCTTTTGC	GGATTGCCAGGTCTACTAC	55
Ad03	GGATCAAATTATGGTTAAGGC	CCAATTTTGAGCCAATTCTCA	50
Ad04	GTTGCTTGTGTGCTTACCC	CATCCCTCTCCCCATTCC	55
Ad05	CTCAACAAGGTTCGGATGTCGTATG	GTCTGCCGGGTGTTTTGCATG	58
Ad07	TAGAAAATTAGCAGATAAGTGC	GATTTCGGTGATATGTTGTAG	41
Ad08	TCTAAAGCCTGTAAGGAAAAATGGG	TTCTCCGTTCACTCTGTACTTCC	54.5
Ad09	TACCACTTCTCCAGATGCTAC	ACTGGCTAGAGATGCGTTG	53.7
Ad14	CTTGATTGGAATACGGGAAATGGAG	CCAAACCAATTGGACTTTGACCTTC	56
Ad18	ACCGCTTCCGTTCTCATTCC	ACCACCACTACACCGTCATTG	56.5

*Annealing temperature after optimisation
Source of primers: Larsen et al (2009)

5.3 Results

5.3.1 Genetic diversity values

The molecular diversities of individual populations calculated from microsatellite bands are presented in Table 5.3. Genetic diversity as measured by; observed mean number of alleles (na), expected mean number of alleles (ne), Nei's (1973) gene diversity (h), Shannon's Information Index (I) among populations was not significantly different in all the diversity indices. The average number of alleles (na) ranged from 1.39±0.10 (in Salima) to 1.59±0.10 (in Karonga) with an average of 1.49±0.10. The effective number of alleles (ne) ranged from 1.20±0.06 (in Salima) to 1.29±0.7 (in Karonga) with an average of 1.23±0.04. The Nei's (1973) gene diversity (h) ranged from 0.12±0.03 (in Salima) to 0.18±0.04 (in Karonga) with an average of 0.18±0.13. The Shannon's Information index (Lewontin 1972) (I) ranged from 0.18 ± 0. 04 (in Salima) to 0.27± 0.05 (in Karonga) with an average of 0.21± 0.07. The number of polymorphic loci (p) ranged from 25 (in Salima) to 38 (in Karonga) with an average of 31 and the percentage polymorphic loci (pp) ranged from 39% (in Salima) to 59% (in Karonga) with an average of 48%.

Table 5. 3 Mean genetic diversity values of five *Adansonia digitata* populations based on nine microsatellite loci

Population	N	Na	Ne	H	I	p	pp
Chikwawa	26	1.50±0.10	1.22± 0.07	0.13± 0.04	0.21±0.05	32	50
Karonga	25	1.59±0.10	1.29±0.07	0.18±0.04	0.27±0.05	38	59
Likoma	26	1.53±0.10	1.23±0.06	0.14±0.04	0.22±0.05	33	52
Mwanza	27	1.42±0.10	1.21±0.06	0.12±0.03	0.19±0.05	27	42
Salima	26	1.39±0.10	1.20±0.06	0.12±0.03	0.18±0.05	25	39
Mean	26	1.49±0.10	1.23±0.04	0.18± 0.03	0.21±0.07	31	48

Mean values are followed by standard error n = number of samples, na = observed number of alleles, ne = Effective number of alleles (Kimura and Crow, (1964), h = Nei's (1973) gene diversity, I = Shannon's Information index (Lewontin 1972). P = number of polymorphic loci, pp = percentage of polymorphic loci

5.3.2 Population differentiation and gene flow

Among population differentiation (G_{st}) and gene flow (N_m), values are presented in Table 5.4. Population differentiation fluctuated depending on the combination of populations. Taking all the populations together showed the highest differentiation of 13%. Subgrouping the populations further showed that including Salima in the northern region populations of Karonga and Likoma Island increased the population differentiation by 6% (from G_{st} = 0.08 to 0.13). This is an indication of isolation by distance. The lowest differentiation of 4% was between Chikwawa and Mwanza both occurring in the Southern region. Gene flow (N_m) ranged from 3.4 to 13.0. The highest number of migrants per generation (13.0) was observed between Chikwawa and Mwanza. The overall migration rate amongst all the populations was amongst the lowest observed (3.47). Including Salima in Northern region reduced migration rate from 5.7 to 3.4 indicating geneflow influenced by isolation by distance.

Table 5. 4 Mean genetic diversity, differentiation and gene flow of all five and subdivided populations

Population grouping	n	H_t	H_s	G_{st}	N_m
All five populations	130	0.158± 0.03	0.138± 0.02	0.13	3.5
Karonga & Likoma	51	0.173± 0.03	0.159± 0.03	0.08	5.7
Karonga, Likoma & Salima	78	0.166 ± 0.03	0.145± 0.02	0.13	3.4

Mean values are followed by standard error; n= number of individuals; Gst = Genetic differentiation; Nm = gene migration; Ht = gene diversity over all groups; Hs= gene diversity of individual relative to their population.

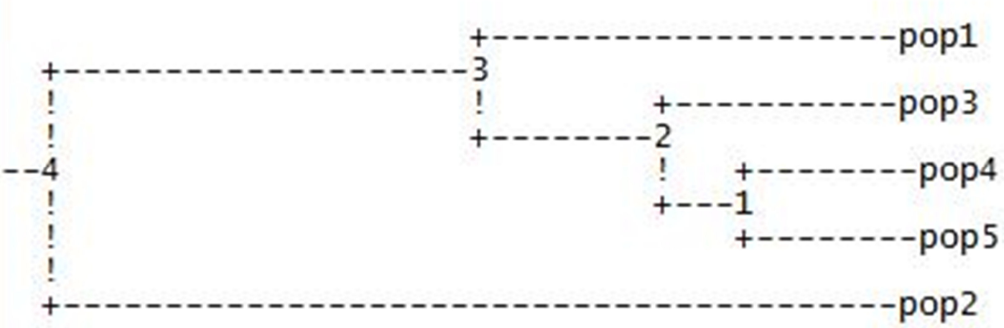
The genetic identity and genetic distance between pairs of populations are shown in Table 5.5. The genetic distance was smallest (0.0099) between Salima and Mwanza populations where as the largest distance (0.0561) was between Likoma Island and Chikwawa populations. Likewise, the highest genetic identity (0.9902) was between Mwanza and Salima populations whilst the lowest genetic identity (0.9455) was between Chikwawa and Likoma Island populations.

Table 5. 5 Nei's genetic identity (above diagonal) and genetic distance (below diagonal)

Population	Karonga	Likoma	Chikwawa	Salima	Mwanza
Karonga	-	0.9669	0.9785	0.9739	0.9775
Likoma	0.0337	-	0.9455	0.9482	0.9592
Chikwawa	0.0217	0.0561	-	0.9838	0.9887
Salima	0.0264	0.0532	0.0164	-	0.9902
Mwanza	0.0227	0.0417	0.0114	0.0099	-

5.3.3 Population structuring

The UPGMA tree constructed based on the genetic distance (Nei, 1972) is given in Fig. 5.2. The dendrogram divided the populations into two major clusters namely, Likoma Island cluster (pop2) and Karonga (pop1), Mwanza (pop5), Salima (pop4) and Chikwawa (pop5) cluster.



```
                       +-------------------pop1
  +--------------------3
  !                    !        +----------pop3
  !                    +--------2
--4                             !   +-------pop4
  !                             +---1
  !                                 +-------pop5
  !
  +----------------------------------------pop2
```

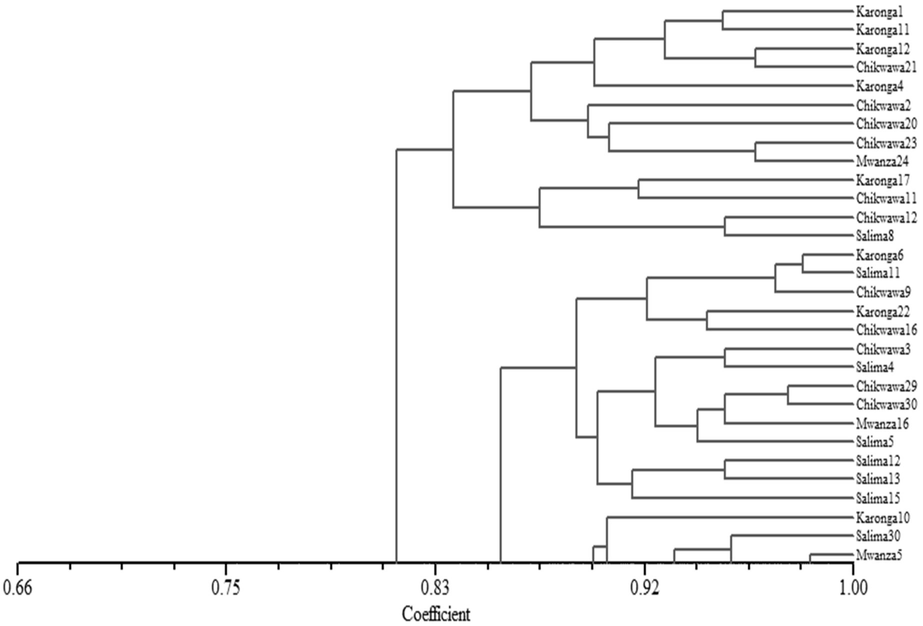
Length between: 4&3=1.12707, 3&2 = 0.48761, 2&1=0.20026, 3 & pop 1=1.18091, 2 & pop3= 0.6933, 1 & pop4 = 0. 49304, 1& pop5 = 0.49304, and 4 & pop2 = 2.30798.

Fig. 5. 2 Dendrogram for five populations (Karonga (1), Likoma (2), Chikwawa (3), Salima (4) and Mwanza (5) based on Nei's (1972) Genetic distance method= UPGA—Modified from NEIGHBOR procedure of PHYLIP version 3.5

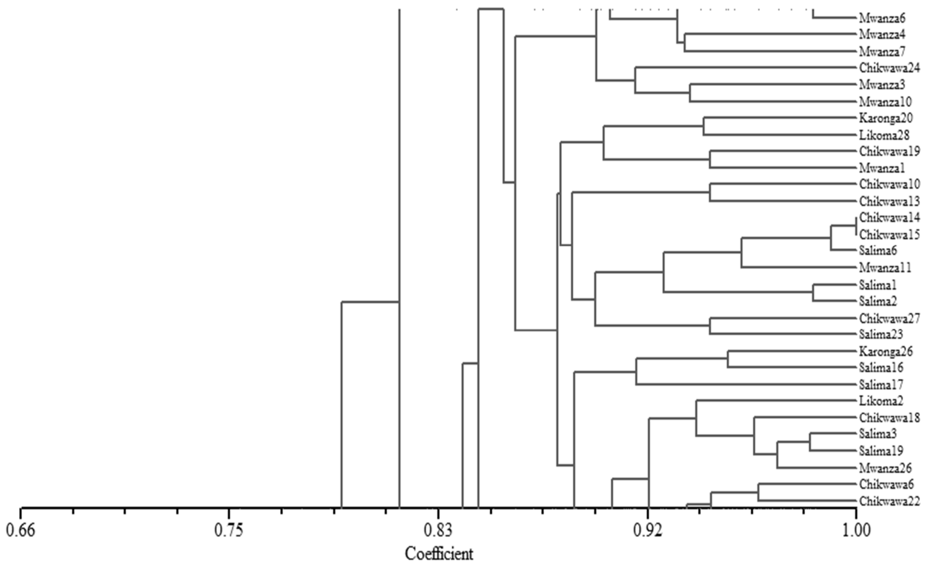
5.3.4 Genetic relationship in individual trees

The dendrogram showing similarity among the individual trees from the five populations is shown in Fig. 5.3. The genetic similarity coefficients ranged from 0.66 (for Likoma tree 6) to 1.00 (for Likoma 13 and 14 trees and Chikwawa 14 and 15 trees). The clustering was not clearly based on silvicultural zonation. Broadly, the genetic similarity coefficients predominantly grouped trees from the mainland trees separate from the Likoma Island trees.

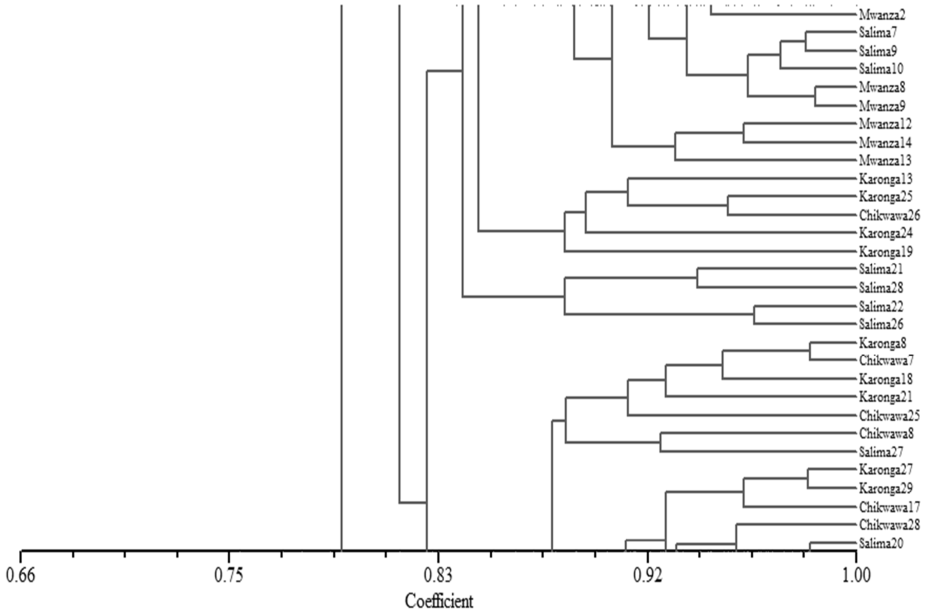
(a)


Karonga1
Karonga11
Karonga12
Chikwawa21
Karonga4
Chikwawa2
Chikwawa20
Chikwawa23
Mwanza24
Karonga17
Chikwawa11
Chikwawa12
Salima8
Karonga6
Salima11
Chikwawa9
Karonga22
Chikwawa16
Chikwawa3
Salima4
Chikwawa29
Chikwawa30
Mwanza16
Salima5
Salima12
Salima13
Salima15
Karonga10
Salima30
Mwanza5
0.66
0.75
0.83
0.92
1.00
Coefficient

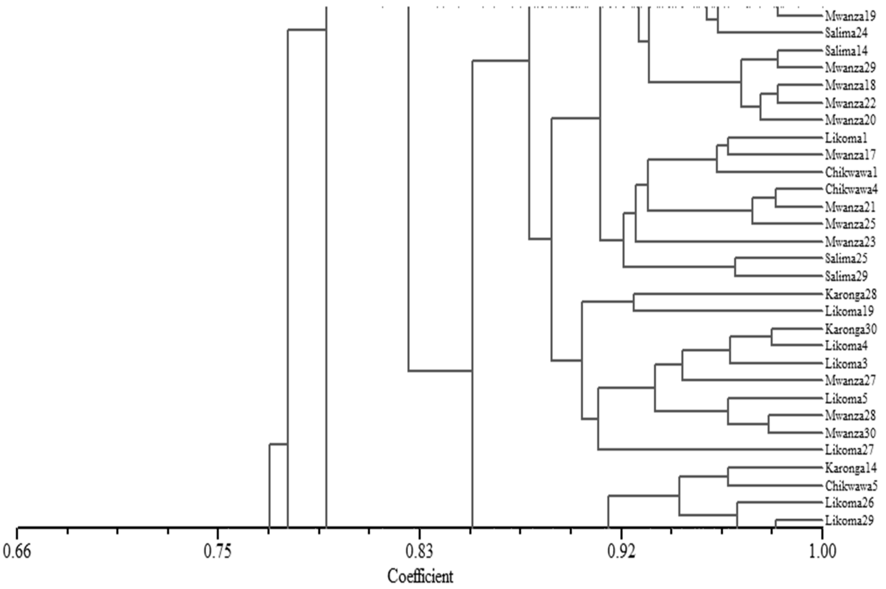
(b)


Mwanza6
Mwanza4
Mwanza7
Chikwawa24
Mwanza3
Mwanza10
Karonga20
Likoma28
Chikwawa19
Mwanza1
Chikwawa10
Chikwawa13
Chikwawa14
Chikwawa15
Salima6
Mwanza11
Salima1
Salima2
Chikwawa27
Salima23
Karonga26
Salima16
Salima17
Likoma2
Chikwawa18
Salima3
Salima19
Mwanza26
Chikwawa6
Chikwawa22
0.66
0.75
0.83
0.92
1.00
Coefficient

(c)


Mwanza2
Salima7
Salima9
Salima10
Mwanza8
Mwanza9
Mwanza12
Mwanza14
Mwanza13
Karonga13
Karonga25
Chikwawa26
Karonga24
Karonga19
Salima21
Salima28
Salima22
Salima26
Karonga8
Chikwawa7
Karonga18
Karonga21
Chikwawa25
Chikwawa8
Salima27
Karonga27
Karonga29
Chikwawa17
Chikwawa28
Salima20
0.66
0.75
0.83
0.92
1.00
Coefficient

(d)



(e)

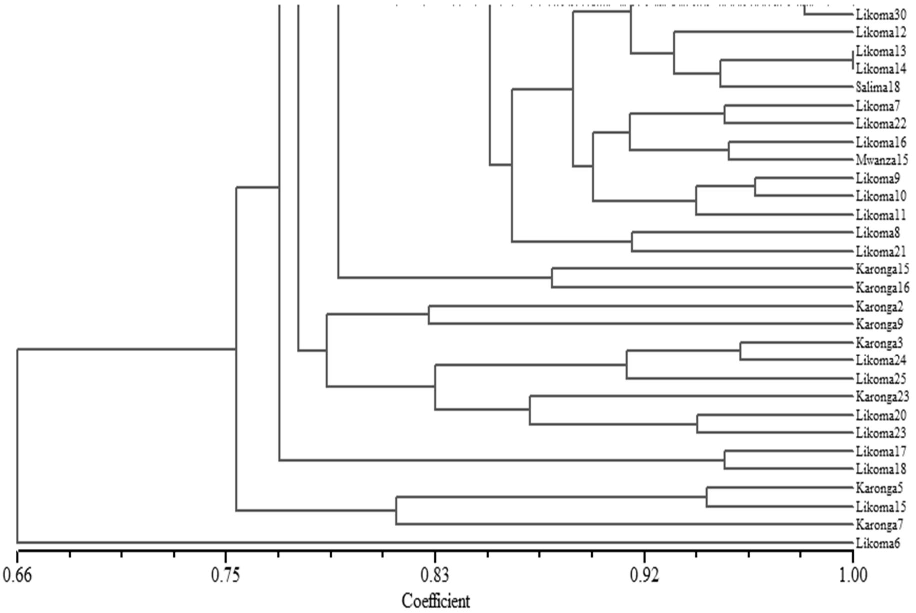


Fig. 5. 3 Genetic relationships among 150 trees in five populations (Karonga, Chikwawa, Mwanza, Salima and Likoma Island) analysed with microsatellites

5.4 Discussion

5.4.1 Genetic variability

Assessment of genetic variation is important for executing plant domestication, conservation and breeding programmes (Sreekumar and Renuka 2006). The common genetic diversity parameters in assessing genetic richness in baobabs has been percentage of polymorphic loci (pp) and Nei's genetic diversity index (h) (Assogbadjo et al. 2006; Assogbadjo et al. 2009; Kyndt et al. 2009). The proportion of polymorphic loci amplified in this study ranged from 39% to 59% with an average of 48% whereas the Nei's genetic diversity (h) ranged from 0.12±0.03 to 0.18±0.04 with an average of 0.18±0.03. In contrast to these results, relatively higher values of percentage polymorphic loci (pp) and Nei's genetic diversity (h) have been reported in baobabs in West Africa. For instance, Assogbadjo et al. (2006) reported (pp) varying from 91.2 to 94.9% and (h) ranging 0.28-0.37; Assogbadjo et al. (2009) reported (pp) ranging 94.1% to 100%, (h) ranging 0.29 to 0.37 whereas Kyndt et al. (2009) reported (pp) ranging 41.7% to 96.1%, (h) ranging 0.22-0.35. The high levels of polymorphism suggest high levels of genetic variation in a species (Sreekunar and Renuka 2009). Changadeya (2009) reported that percentage polymorphic loci (pp) values greater than 50% depict high genetic variation. The average percentage polymorphic loci (pp) and Nei's genetic diversity (h) values in the present study suggest that the variation in baobab is generally low in Malawi. The high genetic diversity (pp = 94.1 to 100%, h = 0.22 to 0.37) found in West Africa could be explained by the fact that it is the centre of diversity for *Adansonia digitata* (Pocktsy et al. 2009), whilst the Malawian populations could be considered as growing in the fringes (periphery) of the centre of diversity of the species. According to Pocktsy et al. (2009), the tetraploid *A. digitata* or its diploid progenitor originated in West Africa and migrated subsequently throughout the tropical parts of the continent through natural and human –mediated terrestrial and overseas dispersal. Baobabs in Malawi could also be considered to occur in the marginal areas (Wickens 1982) colonised after the fluctuation in equatorial rainfall forest during the Miocene-Pleistocene (Pocktsy et al. 2009). The extension into higher rainfall areas of its natural distribution could have been extended by human activities (Wickens 1982) introducing a founder effect on genetic diversity. In addition, according to Yea

(2000), the variations reported above could be a result of differences in frequencies of alleles at a locus caused by random mating, random genetic drift, mutation, natural selection and migration.

The results on genetic variation within the population (Table 5.3) though not significantly different showed consistently that Karonga had the highest diversity in all genetic indices followed by Likoma Island and Chikwawa populations whilst Salima followed closely by Mwanza had the least diversity. The difference in genetic diversity could be attributed to several factors; the major factor within populations being the geographic distribution mode of gene dispersal (Sreekunar and Renuka 2006). Kyndt et al. (2009) reported that long-lived tree species such as baobab show high levels of within–population genetic diversity and low among population genetic diversity due to their high levels of pollen-flow and multigeneration populations. Interestingly, all the populations in this study have shown relatively low genetic variation at both within and among populations. One may suspect that natural causes (genetic drift, inbreeding, and strong selection) have led to genetic depletion in the absence of gene flow from immigrating seeds or pollen (Pluess and Stöcklin, 2004). The factor of population size is critical in the maintenance of genetic variation (Pluess and Stöcklin, 2004). For instance, genetic drift is insignificant in large populations but severe in small populations. It is known that chance fluctuation in allele frequencies could be traced to founder effects (one time drift) (A´vila-di´az and Oyama 2007; Yea, 2000) that can have evolutionary consequences. For instance, genetic drift occurs when some factor subdivides populations into fragments resulting in each fragment evolving independently. Thus the frequency of the alleles in each fragment would depend on the sample of the original population within which it started. Historically, Guy (1971) reported that drought severely reduced baobab demography and Edkins et al. (2007) also reported that in places with high elephant populations, large baobab trees are damaged severely during drought. Anthropogenic factors, elephant damage, mortality from black rust, trampling and livestock browsing, and prolonged drought have also shaped genetic diversity of baobabs in its geographical area (Guy (1971; Wilson 1988). This may also apply to Malawi (Chirwa et al. 2006); and thus genetic drift may have occurred at different levels due to varying reduction in boabab populations. Differences in time of tree flowering within and between populations happening in Malawi may reduce the effective number of trees in baobab populations differently; setting in varying degrees of random drift (Dawson et al. 2009). Genetic depletion of baobab might have been caused by man also through selective utilisation of trees (Schaberg et al. 2008) especially by directional selection by only retaining on farm superior genotypes that provided useful products. This could be an area for further study especially to establish how human demographics may have impacted on baobab diversity in different populations. Salas-leiva et al. (2009) reported that disturbance of the eco-system may have an effect on pollinators which influence tree genetic structure. So far in the present study sites, it is unknown how much pollinators have been affected by habitat destruction and their subsequent contribution to genetic structuring. Salas-leiva et al. (2009) have also reported that genetic structuring is affected by local geomorphology. The sites in this study have diverse physical conditions (Table 5.1) for soil, mean annual temperature and rainfall, which might have caused changes in genetic diversity due to differences in microevironmental conditions. Tree age is another major factor on intra-population genetic variation (Pluess and Stöcklin, 2004). In this study, the sampled populations were composed of both adult and young trees but the ratios varied from site to site. In order to understand the impact of human selection, it would be proper to investigate variation between old and young trees which will also depict existence of new introductions (Maghuly et al. 2006). A´vila-di´az and Oyama (2007) stated that composition of founding populations affect intra-specific genetic diversity. For example, if the founder population was a handful of individuals, then it will be genetically narrow whereas populations with large diverse founding population result in high levels of genetic diversity. The pedigree of this study population is unknown: thus one may just speculate that genetic structure of founding population for different sites has an effect on the obtained levels of genetic indices. Mwase et al. (2006) indicated that distribution range and population size have strong relationship within population genetic variation in tropical tree species with restricted populations displaying significantly less variation than those with wide distribution. In this study, distribution range and population size seem not to correspond to genetic diversity levels. For instance, baobab densities and distribution are higher in Salima and Mwanza and yet these populations have shown low genetic variation. On the contrary, populations in Karonga, Likoma Island and Chikwawa having restricted patchy distribution with relatively low tree densities had higher genetic diversity. It is scientifically established that trees species must have large pool of genetic diversity if they are to survive environmental pressures (Yea 2000). The low polymorphism such as the one found in Salima and Mwanza might be a sign of genetic erosion and populations could be considered endangered. The question remains what is narrowing the genetic variation?

When populations were pooled, genetic diversity showed that mean heterozygosity (H_t and H_s) (Table 5.4) for Karonga and Likoma Island populations was higher than the other combinations. However, the genetic diversity decreased when Salima from central region was combined with Karonga and Likoma Island populations The results further showed least heterozygosity in Chikwawa and Mwanza as a group both occurring in southern Malawi. From the findings, it may be inferred that the hotspot for genetic variation in Malawi reside in the populations in the north in silviculture zone L. High levels of variability may imply either high rates of gain or low rates of loss: where as low levels of diversity may imply either low rates of gain or rapid loss (Amos and Harwood, 1998). Frank et al. (2004) reported that genetic variation in populations may result from limitations to gene flow, genetic drift, spatial variability in selection, and a combination of these factors. For conservation purpose all the populations should be protected since rare alleles exist in them.

5.4.2 Population structuring and genetic differentiation

The UPGMA tree constructed based on the genetic distance (Nei 1972) grouped populations into two major clusters with with an Island population clustered separately from the mainland populations. On the mainland there were also minor clusters with Karonga and Chikwawa belonging to separate groups whilst Mwanza and Salima were in the same group. The clustering did not correspond to silvicultural zonation (Table 5.1) of Malawi (Hardcastle 1978). The results however seem to follow the isolation –by-distance model (Kyndt et al. 2009). The clustering may be considered as ecotypes or races of baobabs existing in Malawi. Ecotypes are generally taken as populations adaptable to specific habitats which may have an implication in domestication, seed distribution, conservation and tree breeding strategies. The Gst values showed that the sampled populations were differentiated by 13% (which comes from alleles private to each population) meaning that 87% were common among the populations. High similarity of alleles may deduce the sharing of common ancestral alleles and the absence of alleles generated subsequent to evolution (Esselman et al. 2000).

The genetic similarity coefficients (Fig.5.3) in results showed that trees from the five populations were 66% to 100 % genetically similar. Generally, the results depicted that mainland trees are genetically closer than the Island trees. The contributing factor could be difference in age and origin among populations in different silvicultural zones (Assogbadjo et al. 2006). Mixture of individuals from different populations in the same cluster may mean that those individuals came from the same origin (refugia) (A´vila-di´az and Oyama 2007) or as a result of multiple introductions from several refugia. The other possible explanation is that even though populations occur in different silvicultural zones they have not undergone significant differentiation (Maghuly et al. 2006). Human influence of gene flow in baobab has been reported as very prominent in Benin (Assogbadgo et al. 2006). With long history of slave trade in Malawi, Arabs might have moved baobab seed from one locality to another (Wickens 1982). The results mirror those reported by Assogbadjo et al. (2006) and Pocktsy et al. (2009). Assogbadjo et al. (2006) found that some genotypes of *A. digiata* populations growing in different climatic zones of Benin belonged to more than one gene pool. Pocktsy et al. (2009) also found that some A. *dig*itata haplotypes were found belonging to more than one region which suggested natural colonization and/or human introductions to the areas involved.

According to Yeh (2000), the G-statistics (G_{st}) values ranging between 0.050 to 0.150 indicates moderate genetic differentiation; 0.151 to 0.250 is representative of large gene differentiation; and above 0.250 represent very large gene differentiation. In this study, genetic differentiation of 13% amongst the five populations and three populations of Karonga, Likoma Island and Salima of 6% could be considered moderate where as 4% population differentiation for Chikwawa and Mwanza was low. When compared to baobab results in West Africa: Kyndt et. al. (2009) found differentiation among populations ranging 2% to 28% whilst Assogbadjo et al. (2006) found 17.63%. The genetic differentiation found in baobab in West Africa was largely attributed to isolation –by- distance pattern rather than representing actual discrete entities. Furthermore, the organisation of genetic diversity came about essentially from spatially restricted geneflow with some influence of human seed exchange (Kyndt et al. 2009). Relatively high levels of genetic structuring found in baobab at both large and within-population level was unexpected in regard to its dispersal by bats and the influence of human exchange of seeds (Kyndt et. al. 2009). Kelly et al. (2004) reported that extensive gene flow leads to low population differentiation. This could be the case with baobab populations in Malawi considering high gene flow revealed Nm>1. However, Assogbadjo et al. (2006) reported that genetic structuring in baobabs showed evidence of physical isolation of the populations when sampled in different climatic zones revealing substantial amount of genetic structuring between populations despite the species having long gene flow. Changadeya (2009) interpreted the low population differentiation in bananas as being a result of the existence of many common alleles shared among populations in contrast to relatively few private alleles behind the differentiation. Sharing of 87% common alleles among populations should be cause of low differentiation observed. In addition, gene flow events in tetraploid species like baobab involve the movement of twice the number of genes transported than in a diploid species leading to less differentiation among the populations (Nassar et al. 2003). Mating behaviour has major influence on genetic structuring in trees (Chung et al. 2004; Kyndt et al. 2009). Baobabs are generally outcrossing (Assogbadjo et al. 2006; Kyndt et al. 2009) with fruit bats paradoxically considered the sole natural pollinators (Assogbadjo et al. 2006; Kyndt et al. 2009; Wickens, 1982). However, baobab is reported to have a mixture of bat, insect, animal and wind mating systems (Sidibe and Williams 2002; Wickens 1982). People have also enhanced gene flow between baobab populations since it is partly domesticated (Kyndt et al. 2009). Geburek (2000) has reported that gene flow reduces genetic differentiation among populations and may retard evolution. The high values of gene migration found in this study (Nm = 3.4- 13) indicate extensive gene flow which should enhance homogeneity of allele frequencies between close wild *A. digitata* populations (Zolgharnein et al. 2010). One advantage of extensive gene flow (Nm >1) in a species is that it counteracts genetic drift (Hamrick and Nason 2000). This partly reduces the worry of continued fragmentation and reduced baobab tree density. Varying bat foraging ranges have been reported up to a maximum of 15 km (Lack 1978; Kynd et al. 2009, Nassar et al. 2003). With this behaviour of bat movement, substantial gene dispersal should result into erasing spatial genetic structuring within a population. Thus, 13% genetic structuring in Malawi should suggest inter-population differentiation caused by other factors in addition to bat influence. It is known that G_{st} values are dependent on life history traits, colonisation events and the extent of the area under study (Pluess and Stöcklin 2004). Overall, the results in the study seem to support differentiation due to geographical isolation with one exception of that between Salima and Mwanza (Fig.5.2).

Furthermore, the genetic distance and identity values (Table 5.2) also support isolation by distance model. For example, Likoma and Chikwawa are the most genetically distant at a distance of about 750 km and have highest values for genetic distance (0.0561) and lowest genetic identity (0.9455). The G_{st} value of 0.13 (Table 5.3) further indicated that maximum genetic differentiation is reached by moving from Northern populations (Karonga and Likoma) to Salima a distance of about 600 km. This is supported by Harmrick and Nason (2000) who reported that species with discontinuous ranges generally have more heterogeneity among their populations than those with more continuous distributions. In the study, there are mountain barriers between Karonga and Salima whilst Lake Malawi separates Likoma Island from the rest of mainland populations. Genetic identity and genetic distance values (Table 5.3) further identify Salima and Mwanza as homogenous populations even though they are separated at a distance of a bout 300 km. The UPGMA dendrogram (Fig.5.3) further confirm that the two populations are genetically similar. Inter-population homogeneity (between Mwanza and Salima) could be interpreted as that the two were a relic of a previously more continuous distribution of *A. digitata* as well as high rates of past gene flow by long-distance pollen flow (Chung et al. 2002), human and bat seed dispersal (Kyndt et al. 2009; Assogbadjo et al. 2006) animal and water dispersal (Wickens 1982; Sidibe and Williams 2002). With moderate differentiation found in the results among five populations, this may indicate that populations may not be considered as a single interbreeding unit (Snustad and Simmons 2010). Rare alleles present in the populations means that these populations with time may have evolved independently and thus domestication, conservation and tree breeding strategies should recognise it.

5.5 Conclusion

At national level the results have shown that Karonga, Likoma Island and Chikwawa are the genetic diversity hotspots in Malawi. Conservation should therefore prioritise these populations. Compared to populations in West Africa, all Malawian populations have shown indications of genetic erosion. For management purposes, it is important that the genetic diversity in all populations do not decrease further. Impoverished level of genetic diversity in Salima and Mwanza should be a matter of great concern since baobab is an important Agroforestry species in the areas. Low genetic diversity is not healthy for outbreeding species facing environmental and climatic change. In populations with low polymorphism, policies should be put inplace to curtail cutting of baobab trees. Further more communities should be encouraged to reverse the genetic erosion through artificial regeneration using seedlings. Seed distributors (Forestry Research Institute of Malawi) for forestry operations should ensure during seed collection within the selected stand to spread the collection over the largest possible number of dispersed trees; collecting from atleast 15 trees at a minimum distance of 100 metres (Schmidt 2000). In addition equal number of fruits should be collected from each mother tree in an accession. It is hypothesized that human factors have a major role on the genetic erosion being experienced in baobab populations. This is an area for further study especially to establish how human demographics may have impacted on baobab diversity. Examining genetic diversity in adult and young trees would reveal impact of anthropogenic factors. On the other hand, the impact of geomorphology should be investigated on genetic structure of baobabs. The impact of deforestation on baobab pollinators and subsequent effect on tree genetic diversity should also be investigated since mating system is critical in tree genetic structure. The results have shown moderate genetic differentiation among the populations. Moderate genetic structuring among populations, implies that many alleles are common among the populations with few rare alleles present due to environmental adaptation. Thus the populations should not be taken as panmictic (single interbreeding unit). Presently, it is unknown how the genotypes would respond once seed is distributed to a unique habitat. Provenance and family trials are required to define the extent of distribution of the germplasm. In addition, on long term basis, breeding seed orchards should be established to promote genetic conservation and broadening the genetic resource through mixing of distant genes. Infusion of foreign genetic materials should also be included in the breeding orchards to increase the genetic base. The long distance gene flow found in this study may imply that spatial genetic differentiation might be low within the population. Therefore, it will be rational to collect seed for domestication and tree breeding from as many mother trees as possible to achieve broad genetic base. The practice in seed collection in wild plants of taking trees spaced at 100 m as genetically unrelated (Schmidt 2000) needs review with the extensive gene flow happening in baobab. The UPGMA tree constructed based on the genetic distance (Nei 1972) grouped populations into two major clusters which may be inferred as ecotypes of baobab. The present grouping contrasts the Hardcastle's (1978) silvicultural zonation implying the criterion for tree seed distribution for forestry operations should be reviewed in Malawi. The clustering of individual trees through genetic similarity coefficients has generally shown that mainland trees are genetically closer than trees from the Island. Specific clustering has shown that genotypes might belong to several gene pools due to either natural distribution, anthropogenic influence, water and animal dispersal. Present results are important in tree domestication, management and improvement strategies at national level. However, baobab is widely distributed in southern Africa; it is therefore recommended to assess the genetic diversity at regional scale. The information gathered will guide in domestication, conservation, breeding strategies and general management (seed collection, seed distribution and afforestation activities) at national and regional level.

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5.7 Reference

A´vila-di´az I, Oyama K (2007) Conservation Genetics of an endemic and endangered epiphytic *Laelia speciosa* (Orchidaceae). American Journal of Botany 94 (2): 184-193

Akinnifesi FK, Silesh G, Ajayi OC, Chirwa PW, Kwesiga FR, Harawa R (2008) Contribution of Agroforestry Research and development to Livelihood of Smallholder Farmers in Southern Africa: 2 Fruit, Medicinal, Fuelwood and Fodder Tree Systems. Agricultural Journal 3 (1): 76-88

Amos W, Harwood J (1998) Factors affecting levels of genetic diversity in natural populations. Philosophical. Transactions: Biological Sciences. Vol., 353 No. 1366:177-186

Assogbadjo AE, Kyndt T, Chadare FJ, Sinsin B, Gheysen G, Eyog-Matig O, Van Damme P (2009) Genetic fingerprinting using AFLP cannot distinguish traditionally classified baobab morphotypes. Agroforestry Systems 75: 157-165

Assogbadjo AE, Kyndt T, Sinsin B, Gheysen G, Van Damme P (2006) Patterns of Genetic and Morphometric Diversity in Baobab (*Adansonia digitata*) Populations Across Different Climatic Zones of Benin (West Africa). Annals of Botany 97: 819-830

Changadeya W (2009) Genetic Morphological and Socio-Economic Characteristion of Musa (Banana) Cultivars Growing in Malawi. Dissertation. University of Malawi

Chirwa M, Chithila V, Kayambazinthu D, Dohse C (2006) Distribution and Population Structures of *Adansonia digitata* in Malawi. FRIM, Zomba, Malawi

Chung MI, Nason J, Chung MG, Kim K, Park C, Sun B, Pak J (2002) Landscape-level spatial genetic structure in *Quercus acutissima* (Fagaceae). American Journal of Botany 89(8): 1229-1236

Chung MY, Nason JD, Chung MG (2004) Spatial genetic structure in populations of the terrestrial orchid *Cephalanthera longibracteata* (Orchidaceae). American Journal of Botany 91(1): 52-57

Dawson IK, Lengkeek A, Weber JC, Jamnaclass R (2009) Managing genetic variation in tropical trees: Linking knowledge with action in agroforestry ecosystems for improved conservation and enhanced livelihoods. Biodiversity Conservation 18: 969-986

Edkins MT, Kruger LM, Harris K, Midgley JJ (2007) Baobabs and elephants in Kruger National Park: nowhere to hide. Blackwell Publishing Ltd. African. Journal of. Ecology. Doi:10.1111/j.1365-2028.2007.00798.x

Esselman E J, Crawford DJ, Soren B, Stuessy TF, Anderson GJ, Silva MO (2000) RAPD Marker Diversity within and divergence among species of *Dendroseris* (Asteraceae: Lactuceae). American Journal of Botany 87(4): 591-596

Franks SJ, Richards CL, Gonzales E, Cousins JE, Harmlic JL (2004) Multi-scale Genetic analysis of *Uniola paniculata* (Poaceae) A coasta species with linear fragmented distribution. American Journal of Botany, 91 (9): 1345-1351

Gawal NG, Jarret RL (1991) Cytoplasmic genetic diversity in banana and plaintain. Euphytica, 52:19-23

Geburek T (2000) Effects of Environmental pollution on the genetics of Forest trees. In: Young A, Boshier D, Boyle T (eds.), Forest Conservation Genetics. Principles and Practice. CSIRO Publishing, United Kingdom

Giang LH, Hong PN, Tuan MS, Harada K (2003) Genetic variation of *Avicennia marina* (Forsk) Vierh. (Avicenniaceae) in Vietnam revealed by microsatellite and AFLP markers. Genes Genet. Syst. 78: 399-407

Glaubitz JF, and Moran GF (2000) Genetic Tools: The Use of Biochemical and Molecular Markers. In: Young A, Boshier, D and Boyle T (eds.), Forest Conservation Genetics. Principles and Practice. CSIRO Publishing, United Kingdom

Guy GL (1971) The Baobabs: Adansonia Spp. (Bombacaceae). Journal of the Botanical Society of Southern. Africa 57: 31-37

Hamrick JL Nason JD (2000) Gene flow in Forest Trees. In: Young A , Boshier D, Boyle T (eds.). Forest Conservation Genetics. Principles and Practice. Principles and Practice. CSIRO Publishing, United Kingdom

Hardcastle PD (1978) A preliminary silvicultural classification of Malawi. Forestry Research Institute of Malawi. Forestry Research Record No. 57

Kelly BA, Hardy OJ, Bouvet J (2004) Temporal and Spatial genetic structure in *Vitellaria paradoxa* (shea tree) in an agroforestry system in southern Mali. Molecular Ecology 13:1231-1240

Kyndt T, Assogbadjo AE, Hardy, OJ, Kakaïa RG, Sinsin, B, Van Damme P, Gheysen G (2009) Spatial genetic structuring of baobab (*Adansonia digitata*, Malvaceae) in the traditional Agroforestry systems of West Africa. American Journal of Botany 96(5):950-957

Lack A (1978) The Ecology of the Flowers of the Savanna Tree *Maranthes Polyandra* and their visitors, with Particular Reference to Bats. The Journal of Ecology, Vol. 66, No.1:287-295

Larsen AS, Vaillant A, Verhaegen D, Kjaer ED (2009) Eighteen SSR-primers for tetraploid *Adansonia digitata* and its relatives. Conservation Genet Resour. DOI 10.1007/s12686-009-9075-y

Maghuly F, Pinsker W, Praznik W, Fluch S (2006) Genetic diversity in managed subpopulations of Norway spruce [*Picea abies* (L) Karst]. Forest Ecology and Management 222: 266-271

Mwase WF, Bjornstad A, Stedje B, Bokosi JM, Kwapata MB (2006) Genetic diversity of *Uapaka kirkiana* Muel. Arg. Populations as revealed by amplified fragment length polymorphism (AFLPs). African Journal of Biotechnology Vol. 5 (13): 1205-1213

Nassar JM, Hamrick JL, Fleming TH (2003) Population Genetic Structure of Venezuelan Chripterophilous Columnar Cacti (Cactaceae). American Journal of Botany 90 (11): 1628-1637

Pluess AR and Stöcklin J (2004) Population genetic diversity of the clonal plant *Geum reptans* (Rosaceae) in the Swiss Alps. American Journal of Botany 91(12): 2013-2021

Pocktsy JL, Lumaret R, Mayne D, Mohamed VA, Abutaba YIM, Sagna M, Raoseta S, Danthu P (2009) Chloroplast DNA phylogeography suggest a West African centre of origin for the baobab, *Adansonia digitata* L (Bombacoideae, Malvaceae). Molecular Ecology 18: 1707-1715

Salas-leiva DE, Mayor-Duran VM, Toro-Perea N (2009) Genetic diversity of the Black mangrove (*Avicennia germinans* L.) in Colombia. Aquatic Botany 91: 187-193

Scharberg PG, Dehayes, D.H, Hawley GJ, Nijensohn SE (2008) Anthropogenic alterations of genetic diversity within tree populations: Implications for forest ecosystem resilience. Forest Ecology and Management 256: 855-862

Sidibe M, and Williams JT (2002) Baobab. *Adansonia digitata*. International Centre for Underutilised Crops, Southampton, UK

Snustad DP, Simmons MJ (2010) Principles of Genetics (Fifth Edition). International student Version. John Wiley & Sons, Inc. Australia

Sreekumar VB, Renuka C (2006) Assessment of genetic diversity in *Calamus thwaitesii* BECC. (Arecaceae) using RAPD markers. Biochemical Systematics and Ecology 34, 397-405

Wickens GE (1982) The Baobab: Africa's Upside-down Tree. Kew Bulletin, Vol. 37, No. 2: 173-209

Wilson RT (1988) Vital Statistics of the baobab (*Adansonia digitata*). African. Journal of. Ecology. Volume 26: 197-206

Yeh FC (2000) Population genetics. In: Young A, Boshier D and Boyle T (eds.), Forest Conservation Genetics. Principles and Practice. CSIRO Publishing, United Kingdom

Zobel B, Talbert J (1984) Applied Forest Tree Improvement. John Wily and Sons, Inc. New York

Zolgharnein H, Kamyab M, Keyvanshokooh S, Ghasemi A, Nabavi SMB (2010) Genetic Diversity of *Avicennia marina* (Forsk.) Vierh. Populations in the Persian Gulf by Microsatellite Markers. Journal of Fisheries and aquatic Science 5 (3): 223-229

6 Reproductive biology of *Adansonia digitata* (Bombacaceae) in Malawi, southern Africa

Abstract

Reproductive process (mating system, pollinating agents), fruit and seed physiology of *A. digitata* was investigated in natural populations in Malawi. Autogamy (spontaneous self-pollination) lead to fruit development which aborted within the first month showing strong self-incompatibility. Wind pollination resulted into successful fruit development but growth was eventually inferior compared to naturally pollinated fruits probably inferring ineffective pollination (pollen limitation). Buds took about 10.1±0.45 to 13±0.6 days to reach anthesis and a further 6.9±0.42 to 11.2±0.43 days to initiate fruiting. Fruit set among trees ranged from 34 to 69%. The fastest fruit growth took place between December and January coinciding with frequent and abundant rainfall. Seed took > 5 months to reach physiological maturity at 42% moisture content but required pre-treatment to enhance germination thereafter. The dominant flower visitors were insects belonging to the orders Hymenoptera and Coleoptera; but their role as pollinators was unlikely. High fruit abortion and pest predation further warrants intensive study that will help to understand the biology and ecology of both the *Adansonia digitata* and the associated pests and diseases.

Keywords: Mating system; Insecta; Phenology; Autogamy; Baobab

6.1 Introduction

Flowering plants account for about 220,000 species (Bawa 1995), and their success attributed to their reproductive biology particularly their pollination and seed dispersal by animals. Animal-pollinated plant families are mostly tropical in distribution and woody whilst wind-pollinated families are cosmopolitan or temperate in distribution (Bawa 1995). Eckert et al. (2009) have reported that mating system influences the effective population size and opportunities for gene flow and consequently the genetic diversity and differentiation of populations and the rate of creating evolution. Parra-Tabla and Vargas (2004) have also presented flowering phenology as one of the most important characteristics in the life history of plants since it greatly determines fitness through sexual reproduction. Several factors have been mentioned that affect pollination dynamics in plants. Plants have evolved in such a way that they synthesise molecules as colour signals in flowers and pollen grain attractive to specific pollinator organisms (Miller et al. 2009). Lobo et al. (2005) have attributed bat-pollination success to factors such as changes to habitats, breeding system of the plant: whether predominantly outcrossing or not, intensity of dryness or availability of water, other chiropterophilic flowers flowering at the specific time, diversity of fruit species available in an area, latitudinal gradient of an area and attractiveness of the nectar as a resource. The factors highlighted may have an influence on baobab pollination depending on site conditions. One good example in plants where environmental factors have caused a difference in mating system is in *Ceiba pentandra* (Bombacaceae) which has bat pollination (*Glossophaga soricina* and *Phyllostomus discolor*) in Guanacaste a relatively dry site but no bat pollination in Osa a wet site in Costa Rica (Lobo et al. 2005). The study showed that it is possible for a tree species to have different mode of mating system depending on geographical conditions within a relatively close distance of < 600 km. This restrains generalization of mating system of a species in its entire geographical distribution.

The history of pollination mechanism in *A. digitata* is outlined by Sidibe and Williams (2002). Bat pollination of Baobab was proposed as far back as 1935; and later on in 1945, fruit bat *Eidolon helvum* was recorded pollinating baobabs in West Africa. Subsequently, *E. helvum*, *E. pomorphorus* gambebiensis and *Rousettus aegyptiacus* were recorded in East Africa. However, suggestions of wind pollination also emerged although it is said to have been disproved as a mating system for the species (Wickens 1982). Other pollinators of baobab were proposed such as bush babies (*Otolemur crussicaudatus* and *Galago senegalensis).* Furthermore, Sidibe and Williams (2002) citing Baum (1965) have indicated that other *Adansonia* species have multiple breeding systems such as pollination by fruit bats and lemurs in *Brevitubae* and long-tongued hawkmoths in *Longitubae.* Undisclosed number of animals other than the major pollinators is known to visit the baobab flowers to exploit nectar and/or pollen (Sidibe and Williams 2002). With the contrasting information on the pollination, there is need to study specific pollinators for the species in different localities. In addition, some of the fruit bats mentioned are habitat specific, mostly thriving in Madagascar. Several species of fruit bats are known in Malawi such as *Epomophorus labiatus, E. wahlbergi, Epomops dobsonii, Rousettus aegyptiacus, R. lanosus, Eidolon helvum, Plerotes anchietae* and *Lissonycteris angolensis* (Radimsumbera and Benda, 2007). The bats live in different habitats and their role as plant pollinators is not yet well defined. Recent studies are also revealing new information on baobab mating systems. Andrioafidison et al. (2006) reported that *A. grandidier* apart from bat pollination is also pollinated by lemurs. They further argued that even though Baun (1995) reported exclusive bat pollination in *A. suarezensis,* they did not rule out lemurs as pollinators. Likewise, Yumoto (2000) when verifying earlier reports that *Durio* species were exclusively bat pollinated found contrasting results; *D. grandiflorus* and *D. oblongus* were pollinated by spider-hunters (Nectariniidae) whilst *D. kutejensis* was also pollinated by giant honey bees and birds. The contradicting reports point to the dangers of extrapolating mating system of a species from one locality to another. On the other hand, different reports on the same species may indicate change in mating system over time. A good example is a species *Ceiba pentandra* a pan tropical tree that has chiropterophilic flowers but the breeding system varies throughout its range. For instance, in south Asia and Africa the species is completely self-compatible whereas in Brazil it is self-incompatible and mixed breeding system in Panama (Lobo et al. 2005). Bawa (1977) also reported that *Maranthes polyandra* had two pollinators, bats and genets. The breeding system in the *C. pentandra* provides opportunity to understand breeding system plasticity in relation to environmental variables such as pollinator diversity and abundance (Lobo et al. 2005).

Information on breeding system of *A. digitata* populations in specific geographical zones is lacking even though it is one of the important species identified for domestication in southern Africa (Akinnifesi et al. 2008). It is however known that fruit production and progeny vigour is affected by mating mechanism (Lobo et al. 2005). Plant reproductive biology can profoundly influence population dynamics (Ngulube 1996). The present study was undertaken to investigate the reproductive biology of baobab especially the mating system and fruit and seed physiology.

6.2 Methodology

6.2.1 Study site

The study site was at Chiwondo in Karonga District (Fig.6.1). The site has mean annual maximum temperature ranging from 28°C to 30°C with mean annual minimum temperature of 22 °C to 25 °C and mean annual rainfall ranging from 500 mm to 1200 mm. The climate is Tropical and has a wet and dry season. The wet season starts in November and ends in April or May (Fig.6.2), while dry season occurs from May to October with occasional rains during the month of June in some parts of the District. The lake shore plain zone where baobab thrives covers an area from the lake at an altitude of between 500 m to 600 m above sea level. Baobab mostly exists in depositional sites comprising of alluvial soils, often calcimorphic (Hardcastle 1978). The lakeshore plain is largely cultivated with isolated trees and small patches or relict woodland of baobab and palm (*Hyphanene ventricosa)* being dominant vegetation (Hardcastle 1978).

6.2.2 Experimental procedure and data collection

6.2.2.1 Mating system

From November, 2007 to March 2008, observation on flowering was done at Chiwondo following the methodology by Ngulube et al. (1998). Twelve trees (Fig. 6.2) were randomly selected at a minimum distance of 50 m to 100 m. The following treatments were investigated: (i) Selfing and apomixis (BS1) - three green buds per tree irrespective of size were covered by cotton cloth to deter any pollination agents (insects, birds, bats and wind); (ii) Wind pollination

(BS2)-three green buds were covered with green mosquito net to inhibit access by animals; (iii) Natural pollination (BS3) – a label was tied to three buds per tree to act as a control. Flowering process and fruit development was observed and recorded initially every 3 days up to fruiting stage (17th December, 2009) using a scale of 0 to 8 as described in flower phenology section below and subsequently once every month fruit length (cm) was assessed.

6.2.2.2 Flower phenology

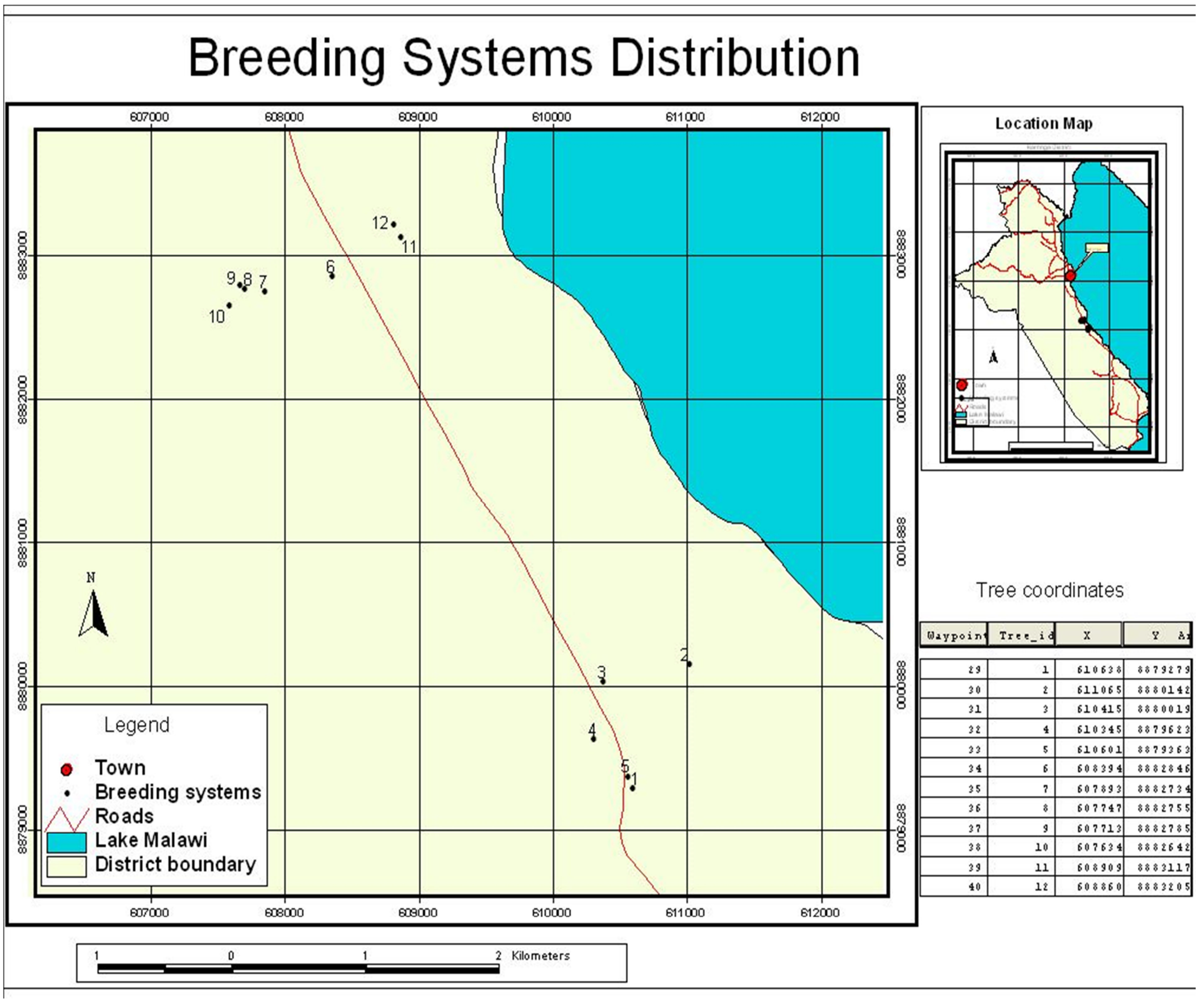
The flowering phenology study was done from November to December, 2007 at the peak of flowering period following the methodology by Ngulube et al. (1998) and Lack, (1978). In four randomly selected trees, one hundred green buds were tagged, observed and scored using a scale of 0 to 8 as follows: (0)Small buds, inflorescence sepals closed (bud stage closed); (1) Large buds, inflorescence sepals cracked (opening); (2) Sepals and petals fully open; (3) Inflorescence petals, anthers, stigma, turned brown; (4) Inflorescence petals, anthers, stigma wilting; (5) Inflorescence petals, anthers, stigma fallen off; (6) Bud fallen before sepals open; (7) Fruit forming and (8) Fruit aborted. Assessment was done every 3 days up to 17th December (a period of 32 days) when all the tagged buds had developed into fruits.

6.2.2.3 Fruit physiology

The fruit physiology study was a continuation of the flowering phenology only that three trees with adequate number of fruits were sampled. Assessment of fruit physiology started January 2007 to March 2008 spanning 103 days. Five randomly tagged fruits from each of three trees were assessed for fresh fruit weight (g) size [length and width (cm)] at the end of each month. The condition / maturity of the seed was assessed for the status of the embryo (observation on growth of the embryo) and moisture content percentage was measured. Five replications with five seeds per replicate were dried at 90 °C for 17 hrs. Physiological maturity of the embryo (seed) was tested through germination capacity. Germination test was done at 55% moisture content. To further test whether pretreatment was required at an early stage of development of the seed, nine mature fruits collected (from the ground after self falling) from one tree (Tree 2, Fig. 2) were tested for germination capacity at 42% seed moisture content. In order to test presence of dormancy, one batch of seed was nicked and the other was not. Completely randomised block design was used with 20 black polythene tubes filled with miombo soil as a treatment unit. Germination was monitored every day for 30 days. Germination was scored only when the seedling developed two leaves.

6.2.2.4 Flower visitors

The study was done in two days on 26 and 27th November, 2007. Observation and collection of insects using sweep nets was done from 08:00 to 19:00 at one tree similar to the methodology by Ngulube et al. (1998). Few flowers were also dissected to check for presence of insects. Insects collected were preserved for identification. Identification of insects was done at the Insect Museum in Biology Department, Chancellor College, University of Malawi.



Tree coordinates

Waypoin	Tree_id	X	Y	A
29	1	610638	8879279	
30	2	611065	8880142	
31	3	610415	8880019	
32	4	610345	8879623	
33	5	610601	8879363	
34	6	608394	8882846	
35	7	607893	8882734	
36	8	607747	8882755	
37	9	607713	8882785	
38	10	607634	8882642	
39	11	608909	8883117	
40	12	608860	8883205	

Fig. 6. 1. Map showing location of sampled trees in Karonga

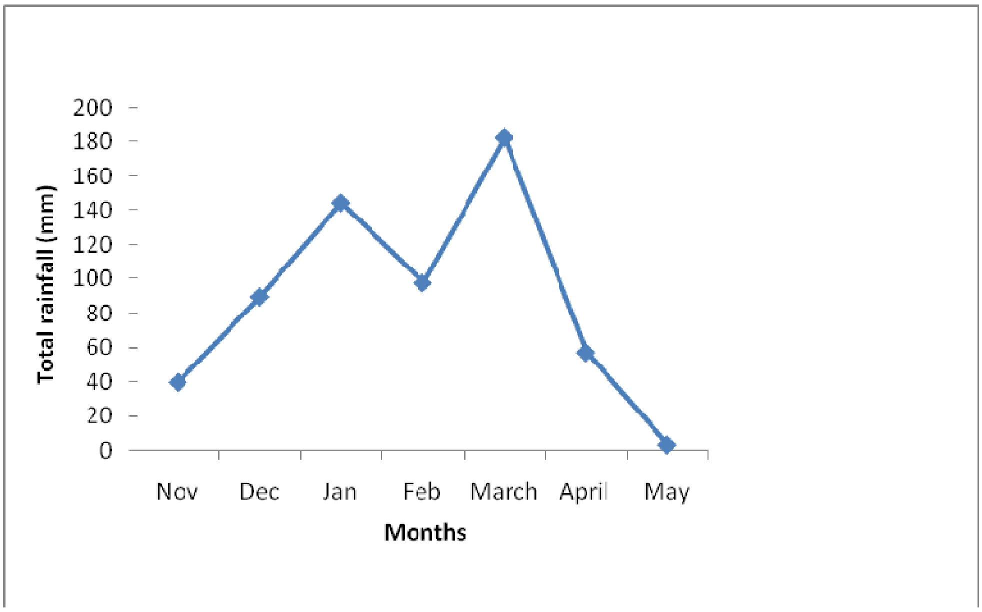


Fig. 6. 2. Total Rainfall for 2007-2008 season in Karonga district, Malawi

6.2.2.5 Data analysis

All the data were subjected to descriptive analysis using Genstat statistical package. The Standard errors were used to detect differences between treatment means.

6.3 Results

6.3.1 Mating system

Table 6.1 shows the results of mating system. In all the treatments, 100 % of the buds flowered. Fruit development ranged from 12 in BS2 to 22 in BS3. All the developed fruits in BS1 aborted. The largest number of fruits (8) developed into maturity in natural pollination (BS3). There was significant difference ($p \leq 0.05$) in fruit length between naturally pollinated flowers (BS3) and flowers bagged using mosquito net (BS2) with the largest fruit length (9.88±1.31 cm) in naturally pollinated treatment (BS3).

Table 6. 1 Number of inflorescence that flowered, developed fruits, aborted and mean length of fruit of the tree treatments (BS1, BS2, BS3)

Treatment	Number of buds flowered (Score 2 to5)	Number of formed fruits (score 7)	Number of fruits aborted (score 8)	Number of fruits	Fruit length (cm)
Covered with cotton (BS1)	33	13	13	0	
Covered with mosquito net (BS2)	33	12	7	5	4.6±1.61
Natural pollination (BS3)	33	22	14	8	9.8±1.31

Means followed by standard error

6.3.2 Flowering Process

There were significant differences ($p \leq 0.05$) in number of days buds took to flower among the trees (Table 6.2) with tree 4 having the shortest period (10.1±0.45) whilst tree 5 had the longest period (13.4±0.6). There were also significant difference ($p \leq 0.05$) in number of days taken from anthesis to fruit formation among the trees with tree 5 having the shortest period (6.9 ± 0.42) and tree 4 the longest period (11.2 ± 0.43). The percentage of fruit development ranged from 34% in tree 2 to 69% in tree 4.

Table 6. 2 Mean number of days for a bud to open and a fruit to develop after flowering and number (%) of fruits that developed with natural pollination

Tree	Days to anthesis	Days to fruit formation	% fruits developed
1	12.0 ±0.61*	8.2± 0.56	48
2	11.6 ± 0.63	10.3± 0.43	34
3	11.9 ±0.56	9.8 ± 0.63	50
4	10.1±0.45	11.2 ± 0.43	69
5	13.4±0.6	6.9 ± 0.42	51

* Mean followed by standard error

6.3.3 Fruit physiology

6.3.3.1 Seed development

At 46 days of fruit development, seed was at milk stage with 73±0.79% moisture content and the embryo was undifferentiated such that it was a mass of fluid. At 75 days of fruit growth, seed moisture content had decreased to 60±1.26%. At this stage, the seed components were clearly differentiated into seed testa, cotyledons and embryo (light green in colour). The seed was still physiologically not mature since germination failed. At 106 days of fruit growth, seed moisture content had decreased further to 55±1.09% and attained 2.5% germination. At 135 days of fruit growth, seed moisture content had reached 42±1.34% and germination percentage attained 22% without pretreatment and 43% with nicking.

6.3.3.2 Fruit weight

There were significant differences ($p \leq 0.05$) in fruit weight among the naturally pollinated trees. The fastest fruit weight gain was within 75 days of fruit development. Most vigorous growth was in Tree 5 whilst the lowest was in tree 4 (Fig. 6.3). After 75 days, fruit weight started decreasing a possible sign of maturity. Tree 5 was fast maturing genotype such that within 103 days of fruit growth, children had already harvested and consumed the fruits: hence no data could be collected in March.

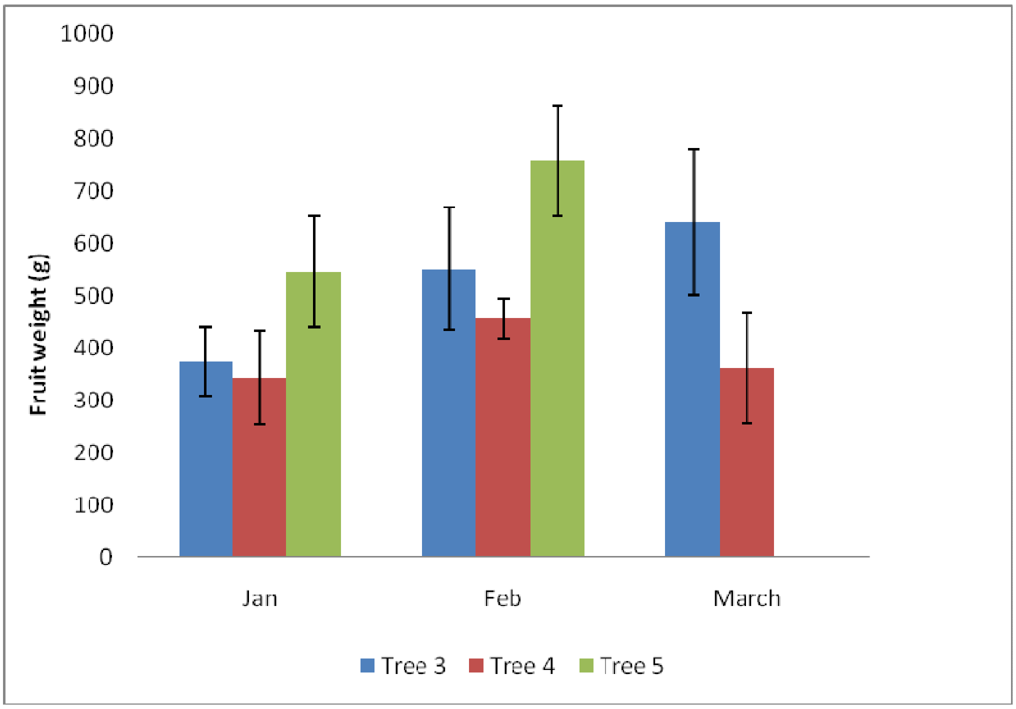


Fig. 6. 3. Mean fruit weight (g) of the three naturally pollinated trees at 97 days of fruit development

6.3.3.3 Fruit length and width

There were significant difference ($p \leq 0.05$) in fruit length among the trees with tree 3 and tree 4 having the largest fruits and tree 4 the smallest. The fastest growth in fruit length was within 46 days from fruit initiation (Fig. 6.4). After 46 days, fruit length increment was minimal in tree 3 and 5 whilst it had reached maximum length in tree 4. There was no increase in fruit length after 75 days of fruit growth. Fruit width was not significantly different among the trees. The fastest growth was attained within 46 days of fruit development (Fig 6.5) and thereafter there was minimal increment.

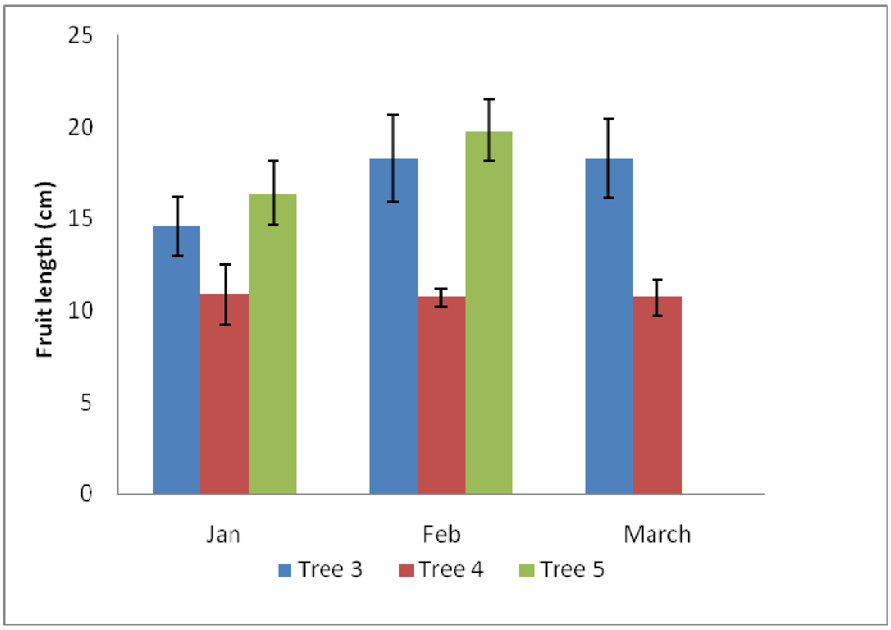


Fig. 6. 4 Mean fruit length (cm) for the three naturally pollinated trees at 97 days of fruit development

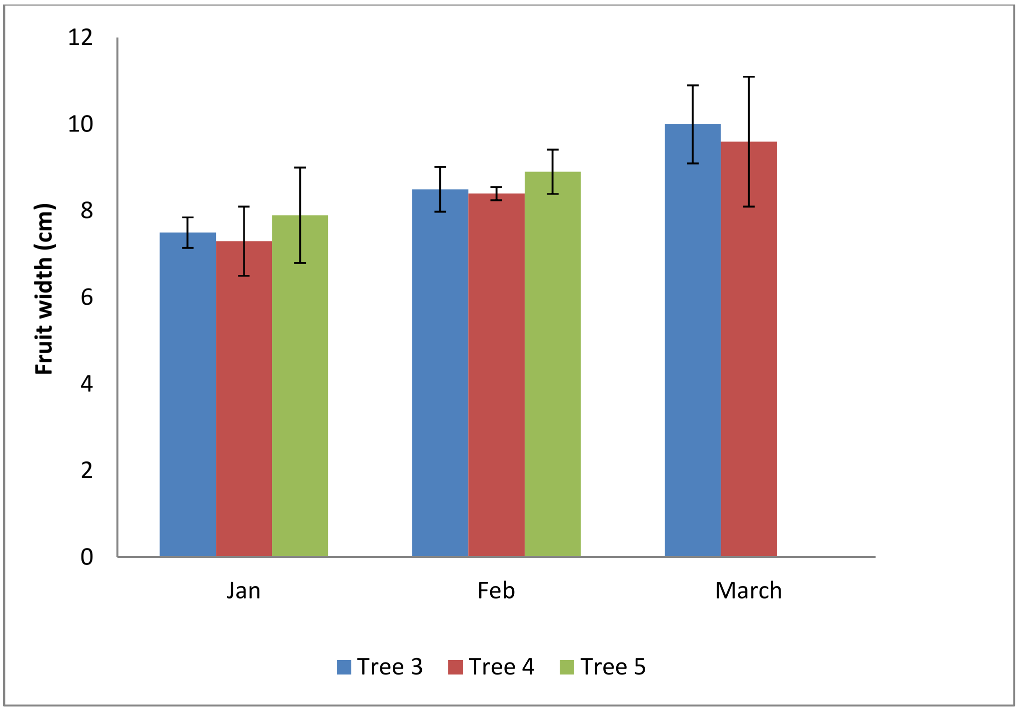


Fig. 6. 5 Mean fruit width (cm) for the three naturally pollinated trees at 97 days of fruit development

6.3.3.4 Flower visitors

Six orders of class insecta were recorded visiting flowers in two days (Table 6.3). The most abundant order were Hymenoptera (124) of the family Chrysidae followed by Coleoptera (7) of the family Curculionidae.

Table 6. 3 Taxonomy of insects that visited baobab flowers

Class	Order	Family	Number of insects
Insecta	Lepidoptera-(butterfly)	Geometridae	1
Insecta	Diptera-(green fly)	Syrphidae	1
Insecta	Coleoptera-(beetles)	Curculionidae	7
Insecta	Hymenoptera	Chrysdidae	124
Insecta	Hymenoptera	Formicidae	2
Insecta	Hemiptera-(bugs)	*	-
Insecta	Diptera-(short bodied)	*	3
Insecta	Hymenoptera-(wasps)	*	2

*Not identified

6.4 Discussion

6.4.1 Mating system

Development of fruits covered with cotton (BS1) suggest occurrence of autogamy or that spontaneous self-pollination may have taken place. The fact that all fruits that developed in BS1 aborted within the first month of development may indicate that pollination was limited or there may have been existence of late self-incompatibility. Similarly, Coupland et al. (2009) reported a higher rate of maternal abortion in bagged treatments of *Avicennia marina* than in unbagged control flowers was attributed to inbreeding depression. Furthermore, the authors presented insect attack on developing fruits and maternal regulation as causes of fruit abortion. This could mean that in *A. digitata,* self-incompatibility is delayed in the species. In addition, 100 % abortion may mean that apomixis (agamospermy) does not exist in the species. Autogamy has been associated with high fruit set in some tree species while in others it results in low fruit set (Coupland et al. 2006). There was a lot of fruit shedding observed under the baobab trees which could be a result of selfing. Successful fruit development took place in treatments covered with mosquito net (BS2) even though growth (mean length) was inferior compared to the naturally pollinated fruits (BS3). The outcome shows that baobab pollination is not exclusively bat-pollinated. Wind pollination is suspected in this study. This finding concurs with that of Sidibe and Williams (2002) who also suggested wind pollination in baobab. Similarly, Wickens (1982) suggested that since *A. digitata* pollen is lightly held by the anthers at the same time the stigma is very receptive, wind pollination should be a possibility. In addition, the author indicated that carrion smell which is attractive to bats, also attracts *Chrysomyia marginalis* (*blue beetle*) and other three nocturnal moths, the American Bollworm (*Heliothis armigera*), Red bollworm (*Diparopsis castanea*) and spring bollworm (*Earias biplaga*) which could not have effected pollination due to obstruction. Earlier reports support present findings because apart from bats (*Galago crassicaudatus*) and insects, baobabs have been said to be also wind pollinated (Gebauer et al. 2002; Sakande et al. 2006). However, the reduction in fruit size in wind pollinated baobab may imply inefficient pollination. Vigorous growth in naturally pollinated flowers indicates the importance of effective pollination in baobab. The result may explain the variation of fruits within the tree. For instance, those fruits well pollinated may attain optimal growth (adequate pulp with many seeds). The results may also shed light on variation of seed from one fruit to another within the tree such that poorly pollinated fruit may have less number of seed. What causes some fruits to be better pollinated than others in the same tree is yet to be investigated. Yumoto (2000) while studying effectiveness of pollinators found that during the day time, nectar volume showed large variations between flowers which suggested that the consumption of nectar by animal visitors varied among flowers which consequently affected pollination effectiveness thereby contributing to variation in fruit set and size. Thus pollinator influence in fruit set may also be functional in baobab. Ineffective pollination in seed orchards in forestry has been addressed by supplemental pollination (Zobel and Tarbert, 1984). The high abortion rate in fruits in all treatments is an issue of concern demanding further research to discover the root cause. According to Eckert et al. (2009), mating success or outcross rate in hermaphroditic species depends on factors that affect outcross pollination such as the density and dispersion of conspecific individuals and the abundance and foraging behaviour of pollinators. This may be applicable to baobab since it is also hermaphroditic (Assogbadjo et al. 2006). It has also been reported that maternal regulation of fruit set and inbreeding depression cause early fruit mortality (Coupland et al. 2006). High fruit abortion in large fruits occurs because it is metabolically expensive to produce adequate substrates for numerous developing fruits (Coupland et al. 2006). Maternal regulation is highly suspected in baobab due to the sheer number of flowers and buds produced, but few fruits growing to maturity. Venter et al. (2011) reported 41% of adult baobab trees producing consistently <5 fruits per year. In the study area, a lot of deforestation has taken place such that most of associated tree species of *Adansonia* are cleared and it is common to see isolated baobab trees. Human intervention therefore could be linked to change of the environment of the tree and the foraging behaviour of pollinators. It has been reported that anthropogenic habitat fragmentation decreases opportunities for outcross pollination (Eckert et al. 2009). Garcia-Papi and Garcia-Martinez, (1984) have also reported that increase in fruit set depends on use of appropriate pollinators which is associated with greater early growth rate of the ovaries preceded by a significant increase in size of the fertilized ovules. In the current study, ineffective pollination in obstructed treatments could have resulted in suboptimal growth of fruits. This supports the hypothesis that the development of the fruit is controlled by the number of seeds in a way that following poor pollination few ovules got pollinated which resulted in minimal fruit growth (Garcia-Papi and Garcia-Martinez 1984).

Fig. 6. 6 Photo of a fungal (sooty disease) infected fruit

6.4.2 Fruit physiology

Variation in days taken for buds to reach anthesis and to fruit initiation following anthesis was not substantial (Table 6.2). These traits could be highly genetically controlled. The findings are backed by Kimmins (1987) who reported that traits that are critical for survival in plants are not influenced much by the environmental conditions, implying that they are highly genetically controlled. While Sacande et al. (2006) observed that each flower wilts and falls within 24 hours of opening, the population in this study showed a lot of variation whereby some flowers fell off few hours after anthesis while some flowers withered gradually retaining freshness for more than 24 hours; and some floral parts withered and persisted to the developing fruits for a long period. However, similar observation as in this study were observed by Wickens (1982) and Sakande et al. (2006) that buds begin to open from late afternoon soon after sunset and are fully open by the following morning; and that the calyx and corolla curl back to expose the stamens and stigma (see Fig.6.7). Conversely, where as Wickens (1982) observed that in the course of progressive wilting, next morning the calyx and corolla straighten and envelop the stamens, these parts mostly remained fully open in the present study. In addition, in the late afternoon not all the corolla became detached and slided down the staminal tube and hanged limply over the stamens. Fruit set in naturally pollinated trees varied from moderate (34%) to high 69%. Fruit load may be influenced by the micro-environmental conditions such as nutrient level and pollinator effectiveness. James et al. (1998) argued that in the absence of sufficient pollination in outcrossing species, a shift of mating system to selfing may occur causing reduced seed set. Such an increase in selfing may culminate in inbreeding depression in the long run. It may also happen in biparental inbreeding if pollen dispersal distances are hindered by fragmentation and near neighbours are genetically close. Eventually, genetic diversity within fragments may be lost by genetic drift and fragmented populations may fail to procreate. Baobab density in Karonga from observation in the study area is about 3-4 trees per ha. Some of the trees are considered male and their role in pollination is not yet clear. Depending on the nature of pollinators, the breeding system may be affected either causing low or high abortion rates as a result of shifts in pollination mechanisms. Several factors have also been suggested for flower or fruit abortion in tree crops such as weather conditions, predation and the amounts of resources the maternal parent can provide for growth and development (Ngulube et al. 1998). For instance, most of the flowers or fruit abscission could be biotic as a result of limitation in nutrients and water (*e.g.* excessive rainfall); and abiotic (*e.g.* predation) conditions, genetic or developmental abnormalities (Ngulube et al. 1998). In the current study, high abscission of fruits is suspected due to heavy fruit infestation of pests and diseases. For instance, on 17th December, 2007 beetles were recorded eating fruit components such as buds, sepals and anthers leading to severe damages and unidentified pest caused severe resin oozing out of perforations in fruits which in most cases prematurely detached from the tree. On the same date, it was also observed that fruits in tree 6 had heavily fruited but at the same time there was also heaviest fruit abscission due to sooty bark disease (compared to other trees around it). Most of the fruits were dying right from the peduncle (see Fig 6.6); the cause of which could be due to fungal attack following heavy rainfall. High insect infestation of the fruit in this species is of concern since it is currently important in nutrition and trade. In addition, strong winds from Lake Malawi may have also caused abscission since baobab trees are the only remnants in parklands- receiving heavy wind blows. Further research is required to investigate the causes of fruit abortion and the relationship between bud, flower and fruit productivity.

Baobab seem to have evolved in such a way that fruit development is extremely fast from December to January in about 46 days (Fig.6.4 & 6.5). Thereafter, growth increment is reduced up to 103 days (March) in some trees. However, in other trees, within 75 days (by February) fruits start losing weight which is a process of maturing. These results indicate two types of genotypes in the population: early maturing trees and late maturing ones. According to Akinnifesi et al. (2008), there is variation in fruit maturity in indigenous fruit trees such as *Uapaka kirkiana*. This variation gives an opportunity to develop cultivars that will mature at different times; thereby extending fruiting season and contributing to rural livelihood**.** Both fruit length and width in Baobab increased very fast between December and January. However, by February, length had reached the maximum point whilst width continued extending. This shows that with continuous deposition of substrates in the fruits, width increases but at a minimal scale. Seed was physiologically mature at 42% moisture content at four months after fruit initiation. The other phenomenon that has been discovered is that the seed developed physical dormancy as soon as it reached physiological maturity. Sidibe and Williams (2002) have also reported dormancy in seed of baobab. Thus it is important to carry out pretreatment before sowing the seed to enhance germination. Growth of baobab fruits through evolution may be linked to rainfall regime as depicted in Fig.6.2. Rainfall in Karonga started in November the time flowering is at the peak, by 17th of December, 2007 nearly all tagged flowers in the study had developed fruits. This is the time the area receives heavy frequent rainfall up to January. This is the time the tree takes advantage of moisture by manufacturing and depositing most substrates into fruit development reaching maximum size by end of February. After January, rainfall decreases and at the same time increment in fruit development decreases; while in other trees, fruits in February start losing weight a sign of the maturing process. By the time rains come to end in May, fruits are physiologically mature to start a new generation; the whole cycle taking about 6 months. Similarly, literature has also reported that fruiting in baobab is in tandem with rainfall and that fruit growth takes 5 to 6 months (Janick and Paul 2008; Wickens 1982).

6.4.3 Flower visitors

Flying insects were very active from 08:00 to 10:00 am then from 15:00 to 18:00 pm; a function of tolerable temperatures in the very hot areas. The highest flower visitors in the study were orders Hymenoptera and Coleoptera of the families Chrysdidae and Curculionide, respectively (Table 6.3). Most of the flying insects were caught hovering around and foraging on the anthers probably picking pollen. No insect was observed perching or blushing against the stigma which is well separated from the stamens (see Fig.6.7). Abundant beetles and ants were also observed lodging between sepals and petals as well as inside the hole of the style where the stigma protrudes. The role of these beetles and ants might be pollination or predation. It is doubtful that flying insects visiting the flower were the pollinators since they did not visit the stigma even though the stigma was sticky and produced aromatic scent. It may be speculated that these insects could have been pollen hunters and parasites. Many fruits were also observed with oozing lesions as sign of infestation at times causing massive premature fruit shedding. Some of the insects that hatched out of the collected fruits for morphometric studies belonged to the Phylum anthropoda, class insecta, order of Coleoptera with families Meloidae and Setonidae. This shows that some flower visitors come to lay their eggs in the flower. Wickens (1982) also reported that some Hymenopterous insects entered the baobab buds through the minute opening of the sepals in the late afternoon soon before the flowers are fully open to collect pollen from the ripe anthers and thus may assist in the pollination. The present study also found a multitude of ants and beetles lodging inside the style of the fully opened flowers probably effecting pollination. However, detailed studies are required to understand contribution of insect species inhabiting in open flowers. Wickens (1982) also proposed the need for studying faunal association with the baobab which is even presently lacking. Raebild et al. (2011) have presented need for research in pollination biology (mating system) in indigenous fruits selected for domestication. Generally, entomological information on miombo species is still scanty. However, according to Sileshi et al. (2008), a variety of mites and insects such as beetles, flies and caterpillars are destructive to blossoming miombo fruit tree species. Specifically, *Carpophilus hemipterus* has been singled out attacking *Adansonia digitata*. The same authors have indicated that although fruit-damaging insects are important pests of horticultural crops as well as miombo fruits in southern Africa, little work has been done with regard to biology and ecology. This calls for the need for intensifying entomological studies in baobab fruit tree species especially at present when domestication of indigenous fruit trees is promoted in the region.

Fig. 6. 7 Fully opened flower during the day showing distinct separation of stigma and anthers

6.5 Conclusions

The hypothesis that *A. digitata* is exclusively bat-pollinated has been disproved. However our findings depict that wind pollination is not a very effective means of pollination either. The results are generally showing that any obstruction or disturbance in pollination in the species will culminate in reduction of fruit growth and productivity. This is important since domestication creates new ecological set up and the change in environment may affect productivity of introduced trees. It may be important to understand the biology, horticulture or silviculture of the tree before large scale planting is done. While there is effective pollination leading to high fruit productivity still occurring in the area, human environmental disturbance in baobab growing areas is quite high. High fruit abortion in some trees may directly be associated with pollination effectiveness. The species has shown that self-pollination is possible even though self-incompatibility sets in timely causing death of the developing selfed fruits. The phenomenon may also explain the heavy abortions of fruits in some trees. However it has been reported that maternal control of fruit development is high in big fruits. Thus it is difficult to generalize the cause of abortion of fruits in baobab. A logical follow up to these results is to undertake controlled pollination to determine the relationship of pollination effectiveness and fruit development. Generally, the findings have shown that fruit development of baobab in the area shows strong relation with rainfall regime lasting 5 to 6 months. However, high insect and disease incidence observed was attributed to upsetting of ecological balance. Further studies on reproductive biology of *A. digitata* should consider both diurnal and nocturnal floral visitors that may contribute to baobab pollination.

6.6 Acknowledgements

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6.7 References

Akinnifesi FK, Sileshi G, Ajayi OC, Chirwa PW, Kwesiga FR, Harawa R (2008) Contributions of Agroforestry Research and development to Livelihood of Smallholder Farmers in Southern Africa:2. Fruit, Medicinal, Fuelwood and Fodder Tree Systems. Agricultural Journal 3 (1): 76-88

Andriafidison D, Andrianaivoarivelo RA, Ramilijaona OR, Razanahoera MR, Mackinnon J, Jenkins RKB, Racey PA (2006) Nectarivory by Endemic Malagasy Fruit Bats During the Dry Season. BIOTROPICA 38(1): 85-90

Assogbadjo AE, Kyndt T, Sinsin B, Gheysen G, Van Damme P (2006) Patterns of Genetic and Morphometric Diversity in Baobab (Adansonia digitata) Populations Across Different Climatic Zones of Benin (West Africa). Annals of Botany 97: 819-830

Bawa KS (1995) Pollination, seed dispersal and diversification of angiosperms. TREE vol. 10, no. 8, Cambridge, UK

Coupland GT, Paling EI, McGuinness KA (2006) Floral abortion and pollination in four species of tropical mangroves from northern Australia. Aquatic Botany 84: 151-157

Eckert CG, Kalisz S, Geber MA, Sargent R, Elle E, Cheptou P, Porcher E, Ree RH, Vallejo-Marin M, Winn AA (2009) Plant mating systems in a changing world. Trends in Ecology and Evolution Vol.xxx No.x. TREE-1151, doi:10.1016/j.tree.2009.06.013

Garcia-Papi MA, Garcia-Martinez JL (1984) Fruit set and Development in seeded and seedless Clementine mandarin. Scientia Horticulturae, 22: 113-119

Gebauer J, El-Siddig K, Ebert G (2002) Baobab (*Adansonia digitata* L.): a review on a multipurpose tree with promising future in the Sudan. Gartenbauwissenschaft, 67 (4).S. 155-160

Gruenwald J, Galiza I (2005) *Adansonia digitata* L. Baobab. Market Brief in the European Union for selected natural ingredients derived from native species. United Nations Conference on Trade and development (UNCTAD)

Hardcastle PD (1978) A preliminary silvicultural classification of Malawi. Forestry Research Institute of Malawi. Forestry Research Record No. 57

Jame T, Vege S, Aldrick P (1998) Mating System of Three Tropical Dry Forest Tree Species. BIOTROPICA 30(4): 587-594

Janick J, Paull RE (2008) Bombacaceae. *Adansonia digitata* baobab. The Encyclopedia of Fruit and Nuts. Cambridge University Press, UK

Kimmins J.P, (1987) Forest Ecology. A foundation for Sustainable Management (2ed). MacMillian Publishing Company. United States of America

Lack A (1977) Genets feeding on nectar from *Maranthes polyandra* in Northern Ghana. East. Africa. Wildlife. Journal, Volume 15:233-234

Lack A (1978) The Ecology of the flowers of the Savanna Trees Maranthesis polyandra and their Visitors, with Particular Reference to Bats. The Journal of Ecology, Volume 66. No.1: 287-295

Lobo JA, Quesada M, Stoner KE (2005) Effects of pollination by bats on the mating system of *Ceiba pentandra* (Bombacaceae) populations in two tropical zones in Costa Rica. American Journal of Botany 92(2): 370-376

Miller R, Owens SJ, Rørslette B (2009) Plants and colour: flowers and pollination. Optical Laser Technology, doi:10.1016/j.optlastec.2008.12.018

Ngulube MR (1996) Ecology and Management of *Uapaka kirkiana* in southern Africa. Dissertation. University of Wales, UK

Ngulube MR, Hall JB, Maghembe JA (1998) Reproductive Ecology of *Uapaka kirkiana* (Euphorbiaceae) in Malawi, Southern Africa. Journal of Tropical Ecology, Volume 14, No.6: 743-760

Parra-Tabla V, Vargas CF (2004) Phenology and Phenotypic Natural Selection on the Flowering Time of Deceit-pollinated Tropical Orchid, *Myrmecophila christinae.* Annals of Botany 94: 243-250

Radimsumbera J, Benda P (2007) Bocage's Fruit Bat (*Lissonycteris angolensis)*, A New Species for Malawi. Nyala 24: 61-63

Raebild A, Larsen AS, Jensen JS, Ouedraogo M, De Groote S, Van Damme P, Bayala J, Diallo BO, Kalinganire HAS, Kjaer ED (2011) Advances in domestication of indigenous fruit trees in the West African Sahel. New Forests 41: 297-315

Sacande M, Ronne C, Sanon M, Joker D (2006) *Adansonia digitata* L. SEED LEAFLET. NO. 109 January 2006

Sidibe M, Williams JT (2002) Baobab. *Adansonia digitata*. International Centre for Underutilized Crops, Southampton, UK

Sileshi G, Barklund P, Meke G, bandeira RR, Chilima C, Masuka AJ, Day RK, Akinnifesi FK (2008) Pest Management in Miombo Fruit Trees. In: Akinnifesi, FK, Leaky RRB, Ajayi OC, Silesh G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds.) Indigenous Fruit Trees in the Tropics Domestication, Utilization and Commercialization. Wallingford, UK, pp369-391

Venter SM, Witkowski ETF (2011) Baobab (*Adansonia digitata* L.) fruit production in communal and conservation land-use types in Southern Africa. Forest Ecology and Management 261: 630-639

Wickens GE (1982) The Baobab-Africa's upside-down tree. Kew Bulletin, Vol. 37, No.2: 173-209

Yumoto T (2000) Pollination of three Durio species (Bombacaceae) in a Tropical Rainforest in Sarawak, Malaysia. American Journal of Botany 87(8): 1181-1188

Zobel B, Talbert J (1984) Applied Forestry Tree Improvement. John Wiley & Sons, New York. U.S.A

7 *Adansonia digitata* (Baobab) as a sustainable healthy natural resource: Linking reproductive biology, genetic variation and socio-economic use

Abstract

The molecular results have shown that within population genetic diversity of *Adansonia digitata* in Malawi is low attributable to being in the peripheral region of the centre of genetic diversity. In addition, population genetic differentiation is moderate due to a large pool of alleles being common across the populations. The results have also revealed long gene flow which has a homogenisation effect. Furthermore, it has been revealed that there is substantial morphometric variation in fruit traits and seedling characteristics showing the potential of being used for selection at both provenance and tree to tree level. In addition, high heritability and additive genetic coefficient of variation imply that there is a potential of achieving high genetic gains through selection and tree breeding. Both molecular study and seed weight have characterised populations into two major races: the Mainland and Likoma Island ecotypes highlighting the possibility of using seed weight in characterising populations; a cost effective way in resource restricted environments. The study has revealed that the species is self-incompatible with no apomixes. Thus, wind pollination resulted into fruit formation even though growth was retarded emphasizing the need for effective pollination that is possibly enhanced in a mixed pollination system. This proves that the species has mixed pollination system and not exclusively bat pollinated as highlighted in some studies. The study also gives an insight in the potential and viability of the baobab industry on the *proviso* that there are enabling government policies that facilitate improved tenure, thereby promoting domestication and commercialization of the Baobab fruit resource.

7.1 Introduction

Baobab fruits play an important socio-economic role in the livelihood of many communities in its area of geographical distribution (Franzel et al. 2008; Sidibe and Williams 2002; Teklehaimahot et al. 2008; Wickens 1982); and are preferred by farmers and traded in southern African region (Akinnifesi et al. 2008). The species has many traditional and economic uses (Akinnifesi et al. 2008; Saka et al. 2008; Sidibe and Williams 2002). Environmentally, soil temperature is substantially reduced under the crowns of the Baobab and also reduces the velocity of wind in the parklands retaining greater soil moisture under tree crowns than in the open (Teklehaimahot et al. 2008). Kalinganire et al. (2008) have presented several uses of baobab such as for food (milk substitute and cream from fruit pulp; condiment for sauces from leaves), medicine (fruit pulp and seed), cordage (from bark), glue (from gum), pottery preparation (wood for firing pots) and enhancing soil fertility in West African Sahel. According to Sidibe and Williams (2002), young leaves are consumed as spinach and cooking sauces; fruit pulp is used in cool and hot drinks; seed kernels are eaten raw; cooked or ground and used in cooking; and the bark has multiple medicinal uses. Presently, seed are used for manufacturing of cooking oil and cosmetic industries (ICUC 2002; Gruenwald and Galiza 2005; Venter and Witkowski 2011). Young plants can be used as asparagus and substitutes of carrots (Gruenwald and Galiza 2005). Baobab pulp has also recently been approved for sale in EU and USA entering formal international markets: further increasing its value to rural communities in the form of cash generation (De Smedt et al. 2011; Gruenwald and Galiza 2005). There is a general increase in export of fruit pulp from Southern Africa and Senagal to international markets (Raebild et al. 2011).

Research work on baobab has picked up due to realisation of its importance especially in the West African Sahel region (Assogbadjo et al. 2006,2009; De Smedt et al. 2011; Dhillion and Gustad 2004; Gustad et al. 2004; Kyndt et al. 2009; Pocktsy et al. 2009; Maranz et al. 2007; Raebild et al. 2011). Raebild et al. (2011) have reported remarkable differences in survival in provenance trials as a result of genetic, environmental or random variation. They are further characterising provenances and families with respect to adaptation as well as fruit and leaf characteristics (including vitamin, medicinal properties and eco-physiological issues relating to drought stress and nematode infestation). De Smedt et al. (2011) have reported significant differences among provenances in phenotypic variation in baobab fruit traits. Maranz et al. (2007) explored how to harness superior nutritional qualities of exotic baobabs through cloning. Significant population differentiation and high genetic diversity have been revealed in baobab through molecular studies (Assogbadjo et al. 2006, 2009; Kyndt et al. 2009). Taxonomical study using molecular methods show that the centre of genetic diversity for *A. digitata* is West Africa (Pocktsy et al. 2009). Ethnobotanical and marketing studies have been reported in West Africa (Gustad 2004; Gustad et al. 2004) revealing important contribution to livelihood of rural communities. Recent studies have been done on baobab in southern Africa in population structure (demography) and fruit productivity (Venter and Witkowski 2011). These authors have also reported consistently low fruit production of less than 5 fruits for 41% of adult trees in a population: a great concern for a potential future horticultural crop. Saka et al. (2008) reported significant differences in nutritional values of baobab provenances in Malawi, which may be applicable to other traits as well. Most of the studies highlighted were concentrated in West Africa and yet the species span a large geographical distribution (Sidibe and Williams 2002; Wickens 1982). Hence, there is need for research to cover the entire geographical area (including Southern Africa). Sidibe and Williams (2002) proposed in depth scientifically-based research to investigate patterns of genetic diversity in relation to distribution. Similarly, Jama et al. (2008) reported need for studies in variability of baobabs in East and Central Africa. Furthermore, there are gaps in genetic control for many traits; genetic and morphometric characterisation of populations; mating systems and patterns (studies on pollinators; test of incompatibility system; degree of self pollination in populations); effective population sizes; and presence of inbreeding depression) (Raebild et al. 2011). Despite reviews on marketing studies in fruit trees in southern Africa (Ramadhani and Schmidt 2008), there is still a lot to be learnt through studying traditional knowledge use of the species and marketing at national level. The current study therefore focused on socio-economic use and marketing, reproductive biology and morphometric and genetic diversity of baobab in Malawi to contribute information that would ensure healthy population for wild and planted populations and a sustainable supply of baobab products for the upliftment of living standards of communities relying on non-timber forest products.

Specifically, the present study attempted to develop an understanding of (i) mating system and the physiology of fruit and seed development; (ii) morphometric characterisation (fruit and seed traits) of five populations growing in four different silvicultural zones classified based on climatic and geological factors; (iii) genetic characterisation of the five populations and determination of genetic control (heritability) of seedling growth characteristics using a nursery study; (iv) genetic diversity of the five wild populations and determination of gene flow using microsatellite markers; and (v) investigation into traditional knowledge in utilisation of baobab tree products and marketing. The ultimate goal was to generate information that could feed into domestication and commercialisation of baobab in southern Africa so that rural people could maximise benefits from the species following planting and proper tree management of wild populations. In the diagrammatic concept (Fig.7.1), the baobab resource is the current wild population which forms the basis of the whole supply of baobab products. The resource is directly influenced by the interaction of processes such as the morphometric (phenotypic), seedling, and molecular variation and the mating system (reproductive biology). In addition, exogenous factors (utilisation, policy, management) have influence on the socio-economic use of the baobab. For instance, with high morphometric variation there is wide choice for developing ideotypes for specific market products even with change in market demands. Degree of seedling variation too will determine progress in improving the resource through selection of superior genotypes that can enhance productivity and supply of products. On the other hand, poor selection strategies may lead to reduced genetic diversity that can drive the whole system to a halt, once inbreeding depression sets in the future base populations. The knowledge of the molecular diversity is fundamental as it provides information on the genetic diversity within and between the populations and gene flow. The information is important in designing tree breeding strategies. Molecular information will also guide on the distribution of germplasm since genotype–site marching is very important for optimum productivity of the forest trees. Furthermore, extent of gene flow will determine clonal forestry, design of seed orchards and general management of both planted and wild populations so that genetic drift does not reduce genetic diversity that can result into many lethal consequences such as reduced fruit set, reduced germination and plant vigour. Mating system is pivotal for all the processes that will see baobab populations maintaining healthy status and sustainable supply of the products. Baobab being an outcrossing species, related mating will result into poor fruit productivity, seed viability and erosion of the genetic variation due to genetic drift. Effective pollination is also pertinent so that fruit development reach optimum capacity and high productivity. Thus any disturbance on the mating system will have a direct effect on the whole trade and social dependence on the species. Finally, the exogenous factors will also influence the viability, and genetic diversity and structuring of the population; while conducive government policy will boost trade and build the population base through either domestication and improved land and tree tenure. How the beneficiaries (rural collectors) tend/nurture the trees will determine the future availability of baobab resources. The processes are highly interlinked such that it was prudent in this study to concurrently research on them as presented in the five following specific studies.

7.2 Genetic differentiation and diversity of Adansonia digitata (Baobab) in Malawi using microsatellite markers

This study has generally revealed that there is low genetic diversity within the provenances across the country. The attributing factor could be that the populations occur in the peripheral to the centre of genetic diversity which is in West Africa (Pocktsy et al. 2009). The same was found in *Avicennia marina* in Vietamese populations occurring in marginal sites of centre of genetic diversity (Giang et al. 2003). At national level, genetic hotspots exist in Karonga, Chikwawa and Likoma Island requiring conservation status. Furthermore, serious genetic erosion (lack of microsatellite diversity) has been revealed for Mwanza and Salima populations: presently attributed to anthropogenic causes such as agricultural expansion leading to directional selection by retaining only preferred genotypes in the parklands. Secondly, natural causes such as drought occurrences and elephant damage (Wilson 1988) may also have reduced the population. These natural causes as well as human impacts reduced population size which enhanced the loss of genetic variation (Giang et al. 2003; Scharberg et al. 2008) creating recent bottlenecks; perhaps in association with founder events (Chung et al. 2004). Low allelic richness might suggest loss of rare alleles during selection (Jones et al. 2006; Schaberg et al. 2008). It is known that

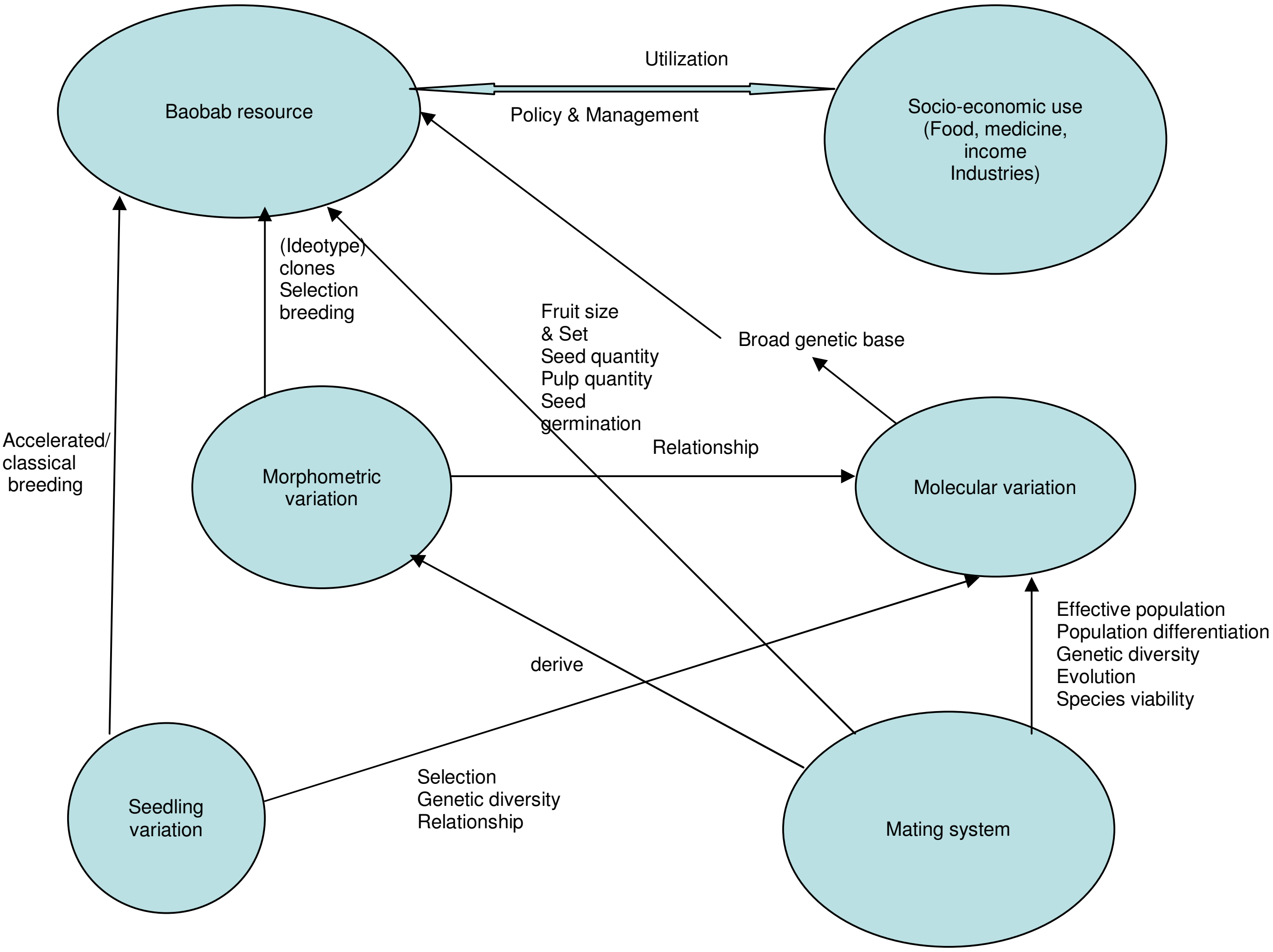

Fig. 7. 1 Linkage of socio-economic, reproductive biology, morphometric and genetic variation of *Adansonia digitata* in Malawi

forest management regime determine the number of mother trees that contribute to the next generation and thereby influencing the genetic diversity (Kramer et al. 2008). Removal of inferior phenotypes results in the loss of rare alleles, which could reduce the potential of populations to withstand environmental change (Pautasso 2009; Dawson et al. 2009). Outbreeding species are expected to harbour higher genetic diversity (Verma and Rana 2011). Low genetic diversity is lethal for fruit trees since reduction of heterozygosity as manifested in polymorphism percentage loci culminate into reduction of fruit set and growth, reduce seed germination (seed without embryo), and reduction in heterosis (Dawson et al. 2009; Zobel and Tarbert 1984). It is reported that the high levels of polymorphism are an indication of high levels of outcrossing among individuals and between populations (Sreekumar and Renuka 2006). Thus it may imply that outcrossing is still high in Karonga, Likoma Island and Chikwawa whilst it is decreasing in Salima and Mwanza. Low polymorphism in Salima and Mwanza is evidence of erosion of genetic diversity as well. Narrow genetic diversity means that the populations can not respond to environmental changes (Dawson et al. 2009; Heywood and Watson 1995; Kaushik et al. 2007; Kitzmiller 1990; Pautasso 2009) such as climatic change and advent of new pest and diseases. Furthermore, the study has revealed that there is generally moderate genetic differentiation among the five populations despite occurring in different silvicultural zones. The Gst value of 0.13 means that the populations have 87% of the alleles common whilst only 13% are private alleles probably due to environmental adaptation. The results manifest that the populations share and harbour a lot of the same copies of ancestral alleles (Esselman et al. 2000). In addition, it shows that most of the diversity resides within the populations (Nassar et al. 2003). With high common alleles, domestication, tree breeding, tree seed distribution should ensure that the genetic base is kept broad; otherwise inbreeding depression will set in rapidly. High gene flow (Nm>1) revealed is an encouraging result which has a homoginisation effect that reduce effect of bottlenecks (genetic drifts). Thus even though there is an increase in tree isolation due to deforestation (land defragmentation), chances that inbreeding depression will set in is reduced, thereby sustaining availability of baobab fruits and heterosis in plants. The results in the dendrogramme have shown two major races of populations of baobabs; the Likoma Island and the Mainland populations. Neutral markers such as microsatellites do not show adaptive traits (Assogbadjo et al. 2006): as such it will be beneficial to carry out field trials (provenance) to evaluate growth traits once seed is distributed from the Island to Mainland and *vice versa*. On the other hand, the 13 %, private genes reflect population substructure even on the mainland (Laushman 1993). This substructuring may infer special geographical adaptation influencing vital physiological processes at present not yet comprehended. The results have further shown that Mwanza and Salima might be panmictic (no mating restrictions) whilst the other populations are not. For tree breeding, it will be important to understand the flowering phenology before doing classical breeding such as establishment of seed orchards as it is known that flowering is influenced by onset of rainfall (Sidibe and Williams 2002) which varies across the populations in Malawi. In addition, gene flow between highly structured populations can produce outbreeding depression (Chung et al. 2004; Laushman 1993). Furthermore, the genetic identity and distance values have shown that most genetically distant populations were Likoma Island and Chikwawa showing that population differentiation is as a result of isolation by distance (Assogbadjo et al. 2009; Kyndt et al. 2009; Pautasso 2009). In the present study, the gene flow barriers could be Lake Malawi and mountain ranges on the mainland. Genetic similarity coefficients further separated the Likoma Island individuals from the mainland individuals. However, the aggregation of the mainland trees did not correspond with silvicultural zonation at all. The individuals seem to come from more than one origin and some genotypes belong to more than one gene pool. This could be due to influence of man through seed dispersal (Kyndt et al. 2009; Wickens 1982). The results also show need to re-look at silvicultural zonation of Malawi presently used as a tool for guiding tree seed distribution in the country.

7.3 Phenotypic variation in fruit and seed morphology of Adansonia digitata (Baobab) in five selected wild populations in Malawi

Knowledge in phenotypic variation is fundamental in developing efficient forest conservation, domestication and tree breeding strategies (Sreekumar and Renuka 2006). The results have revealed significant variation of high magnitude in fruit traits among and within the populations. The high polymorphism found means that at national level there is adequate breeding materials on which selection and breeding can be based on. Thus, there is no need for an infusion to improve traits such as fruit pulp and seed quantity characteristics of economic significance. In addition, the existing variation can support clonal forestry to reduce precocity in baobab from more than 10 years to 4 years (Kalinganire et al. 2008) and enhance productivity at the same time. These results are different from molecular study that has shown low genetic diversity existing in populations in Malawi. Similarly, studies on molecular genetic variation could not find a genetic explanation for the production of different size of fruits (length) (Assogbadgo et al. 2009; Venter and Witkoski 2011). Probably the actual frequencies of specific forms (shape) of fruits in different localities may reflect genetic diversity in relation to molecular diversity. However, fruit traits in this study can not be used to delineate populations in specific ecotypes due to overlaps in multiple comparison tests. The results showed that the larger the fruit the greater the number and quantity of seed whilst the relationship of fruit size and pulp seemed weak because in most cases fruit size did not translate into significantly higher level of pulp content. Hence, in tree breeding, indirect selection of candidate trees for pulp production should be avoided. On the other hand, fruit weight could be used for indirect selection for seed characteristics. This study did not look at taste and nutritional attributes which are also important. Saka et al. (2008) has reported significant variation among provenances in nutritional attributes of baobab in Malawi. Significant variation in fruit traits suggest existence of genetic variation that can be selected for during fruit domestication and tree improvement strategies (Mwase et al. 2006). High tree to tree variation suggests considerable opportunities for selection within each provenance with wide application in domestication and tree improvement. Selection at provenance and tree to tree level is advantageous as it ensures broad genetic base (De Smedt et al. 2011). Seed weight was higher in a fruit than pulp weight. This shows the potential of industries based on seed such as cooking oil, cosmetics and beverages. The trait had high variation as well indicating potential of developing ideotypes, assuming variation is strongly genetically controlled. Individual seed weight indicated pronounced evidence of divergence of baobab populations into two major races (ecotypes); the Likoma Island and Mainland races similar to molecular results. This implies that this morphological trait can be utilised to characterise tree populations in place of using expensive and time consuming molecular studies. Mwase et al. (2006) also reported divergence of populations based on seed weight in *Uapaka kirkiana*. In support of the results, Assogbadjo et al. (2009) have also presented a link between morphological diversity of baobab and both abiotic, environmental factors and genetic basis of tree height and number of main branches.

7.4 Genetic variation among and within provenances of Adansonia digitata (Baobab) in seed germination and seedling growth from selected natural populations in Malawi

Starting time for seed germination showed that the populations were genetically similar. This supports the molecular study which showed that populations in Malawi have 87% common ancestral alleles. Furthermore, high germination reached (such as 95%) shows effective pollination. Generally, all the populations had families with high germination inferring that all populations can be used for the supply of high physiologically quality seed. However, some families despite good fruit handling had poor germination. Thus it may be interpreted that it was due to inbreeding depression following related mating due to human selection. Poor pollination can not be linked to ineffective pollination because of the long gene flow (Nm>1) and high germination in some families show presence of effective pollination agents. Seedling growth traits revealed significant differences between and within provenances. The variation detected in the nursery is genetic in origin (Sniezko and Stewart 1989). If variation in seedling traits reflects the magnitude of variation for mature individuals, then there is ample opportunity for tree improvement through selection and breeding. However, these results should be supported with field trials to characterise provenances and families in relation to adaptation. In addition, there is need to assess genetic control of fruit traits since there is no information linking seedling variation and mature traits (Raebild et al. 2011). The high genetic control of most traits (h^2 ranging from 0.15 to 0.71) is an indication that some traits are strongly genetically controlled. This shows the potential of improving the species through selection of plus trees and breeding. Furthermore, additive genetic coefficient of variation (AGCV) of greater than 10 also showed potential of achieving high genetic gains through selection and breeding in baobab species. The advantage is that the selection could be done at both provenance and tree to tree level ensuring broad genetic base; thus reducing inbreeding depression in future base populations. The genetic characterisation in the majority of seedling traits clustered Likoma Island different from Mainland populations; similar to molecular results and individual seed weight results. This shows the potential of using seedling traits in identifying superior genotypes; with more application in tree breeding than the expensive molecular procedure. Apart from determining superior genotypes, nursery results generate heritability values important in designing tree breeding strategies. The results show that tree breeding programmes may use both individual seed weight and seedling parameters to predict population genetic structure.

7.5 Reproductive biology of Adansonia digitata. (Bombacaceae) in Malawi, southern Africa

The results have revealed self-incompatibility in baobab. This indicates the importance of promoting outcrossing in the species. Unfortunately, the population of baobabs are on the decrease in Malawi (Chirwa et al. 2006); and there is no deliberate planting and protection of wildlings to increase genetic variation. Even then, molecular study has shown long gene flow, related mating will result in inbreeding depression. The large coefficient of variation found in Table 3.2 apart from environmental effect could be as a result of ineffective pollination in this species as shown in Table 6.1 hindered pollination resulted into poor fruit development. Further more the large variation in seed germination in Fig 4.2 and 4.3 could be due to pollination effectiveness. Schmidt (2000) has attributed high frequency of inbred seed to low viability a factor which could be applicable to poor performance of families in Fig. 4.3. However, Fig 4.4 (plant height), Fig. 4.5 (tuber weight) and Fig.4. 3 (germination percentage) showed weak relationship. For instance, once seed germinated, seedling vigour seemed to have been a function of either genotype or amount of food reserves. For example S4, S7, M8, M11 and M5 had relatively outstanding growth despite having poor germination percentage. Results in Table 6.3 depicts the importance of effective pollination in sustainable fruit production such as quantity (number of fruits per tree) and quality (fruit length, width, seed number and weight and pulp weight) in baobab. The frightening statics of about 41% of fruiting adult baobab trees being classified as “poor producers” in South Africa, consistently producing <5 fruits per year (Venter and Witkowski 2011), could also be applicable to Malawi. Poor fruit production could be linked to self-incompatibility following related mating when considering massive flowering that happen in the species. Self-incompatibilty could also be due to small effective population during flowering season, since large proportion of baobab trees are composed of “male trees” that flower at the same time as female trees, but their role presently is unknown (Venter and Witkowski 2011). Furthermore, there seem to be wide variation in flowering time within the populations, influencing effective population size by creating subpopulations which may eventually lead to genetic drift and poor fruit productivity. It is known that *Adansonia digitata* has hermaphroditic flowers (both male and female parts in the same flower) and outcrossing (Assogbadjo et al. 2006; Sidibe and Williams 2002; Wickens 1982; Venter and Witkowski 2011). Further studies should be done to understand the reproductive biology of the "male" trees and their relationship with the genetic structure of the population. Wright (1976) indicated that it is possible for a tetraploid and diploid to mate and produce a triploid which is sterile, which could be the cause of sterility ("male trees") of some baobab trees today. *Adansonia digitata* is one of the species being promoted for domestication in southern Africa (Akinnifesi et al. 2008). Thus tree seed collection should ensure large number of mother trees collected widely in the population to avoid effects of inbreeding depression in future generations. Dawson et al. (2009) has presented gruesome statistics that most agroforestry nurseries raise seedlings from an average of 6.4 trees and 22% of them use a single mother tree: a precursor for future inbreeding depression, genetic drift and extinction of the species. To enhance fruit production in fruit trees, clonal propagation is currently being promoted (Akinnifesi et al. 2008). Clonal forestry reduces genetic diversity (Dawson et al. 2009). Continued reduction of genetic diversity in Malawi will rapidly result into genetic drift with dire consequences on sustainability of baobab products. This calls for further studies on the right number of clones that should be combined in seed orchard and the design (pattern) of ramets in a plantation to avoid related mating since there is long gene flow in baobab. Wind pollination resulted in successful fruit development but growth was inferior compared to naturally pollinated fruits. This depicts the importance of effective pollination for fruits to develop to the optimum size. The results reflect the importance of maintaining the ecosystem balance which is doubtful at present with continued deforestation and expansion of agriculture in the parklands. The results also point to the need of reviewing the notion that baobabs are exclusively bat pollinated. Hence the call for intensification on studies on reproductive biology of baobab as presented by Raebild et al. (2011). The results also support molecular results where the value of $G_{st}= 0.13$ is a reflection of mixed mating system (Nassar et al. 2003). Fruit productivity is still high in some trees showing effective natural pollination despite habitat destruction in Malawi: perhaps due to long gene flow. Dominant flower visitors in this study were insects of the orders Hymenopter and Coleoptera; generally suspected to be parasites (see Fig.7.2) and not necessarily pollinators. Some trees showed massive abortion of fruits due to pest and disease infestation. High fruit abortion, disease infestation and pest predation calls for intensive study to understand the biology of both the *Adansonia digitata* and the associated pests and diseases. Domestication initiatives will likely favour selection of genotypes resistant to pests and diseases.

Fig. 7. 2 Insects that hatched out of the mature baobab fruits collected in Karonga

7.6 Socio-economic value of Adansonia digitata (Baobab) products in Malawi

This study has revealed that baobab has important multiple social roles in the form of food, fibre and medicinal purposes and source of income. Economically, the results have shown potential of commercialisation of baobab provided the government can support the entrepreneurs with financial support for product development and procurement of advanced machinery. In addition, in order to support the industry, there is need to establish sustainable resource base by instituting proper management of the natural resource including promotion of domestication. It is reported that integration of fruit production into forest management can enhance financial security among local communities (Syampungani, 2009). Rural communities should therefore be trained in the propagation of the species and encouraged to protect natural regenerates. Notwithstanding, in order to maintain sustainable supply of baobab products and improve marketing at national level, there is need for enabling government policies (including land tenure) to transform informal markets to formal sector.

7.7 Conclusion

Naturally growing baobabs in Malawi have revealed relatively low genetic diversity and the status should be improved or maintained so that it does not decline any further. In addition, human induced decline in genetic diversity has also been revealed. An infusion of genetic materials from West Africa, East Africa and Southern Africa is proposed to expand the genetic diversity. Southern Africa, including Malawi, can benefit from range wide baobab seed collection done in Sahelian West Africa as reported by Raebild (2011). In Malawi, the government needs to institute measures so that populations in Mwanza and Salima should have the genetic diversity restored most likely through domestication. Due to existence of moderate genetic differentiation among populations, prudent handling of seed distribution should be followed to avoid inbreeding depression in future generations. In addition, further field testing is needed to characterise the provenances and families with respect to adaptation and to understand heritability of the fruit traits. Adoption of clones as ideotypes should only be promoted widely after testing clonal plasticity (evaluating genotype x environmental interaction). The silvicultural zonation, as prescribed by Hardcastle (1978), as a tool for guiding seed distribution should be reviewed taking into account results of the present molecular studies. Domestication should ensure that only seed of broad genetic base is used in agroforestry for sustainable supply of baobab products and improved livelihood of rural communities. Research in species reproductive biology and its association with pest and diseases should be intensified. This study only sampled a small distribution of baobab geographical distribution; calling for future studies to cover a broader region of southern Africa. In order for rural communities to maximise benefits from baobab, people should be encouraged to organise themselves in groups (cooperatives) to collect and produce high value products inform of fruit pulp, wine and oil from seed for export. Furthermore seed which at present is discarded could be made into beverages for domestic and international trade. Leaf products could also be promoted at local and national level to improve diets especially supply of vitamin A in rural communities. Root tubers should also be promoted as novel products in the field of nutrition. Most importantly, enabling government policies that support growth of trade to formal sector should be instituted and land and tree tenure should be reviewed to enhance domestication.

7.8 References

Akinnifesi FK, Sileshi G, Ajayi OC, Chirwa PW, Kwesiga FR, Harawa R (2008) Contributions of Agroforestry Research and development to Livelihood of Smallholder Farmers in Southern Africa:2. Fruit, Medicinal, Fuelwood and Fodder Tree Systems. Agricultural Journal 3 (1): 76-88

Assogbadjo AE, Kyndt T, Sinsin B, Gheysen G, Van Damme P (2006) Patterns of Genetic and Morphometric Diversity in Baobab (*Adansonia digitata*) Populations Across Different Climatic Zones of Benin (West Africa). Annals of Botany 97: 819-830

Assogbadjo AE, Kyndt T, Chadare FJ, Sinsin B, Gheysen G, Eyog-Matig O, Van Damme P (2009) Genetic fingerprinting using AFLP cannot distinguish traditionally classified baobab morphotypes. Agroforest Syst, 75: 157-165

Chirwa M, Chithila V, Kayambazinthu D, Dohse C (2006). Distribution and Population Structures of *Adansonia digitata* in Malawi. FRIM, Zomba, Malawi

Chung MY, Nason, JD, Chung GM (2004) Spatial genetic structure in populations of the Terrestrial orchid *Cephalanthera longibracteata* (Orchidaceae). American Journal of Botany 91(1): 52-57

Dawson IK, Lengkeek A, Weber JC, Jamnaclass R (2009) Managing genetic variation in tropical trees: Linking knowledge with action in agroforestry ecosystems for improved conservation and enhanced livelihoods. Biodivers Conserv 18: 969-986

De Smedt S, Alaerts K, Kouyaté AM, Van Damme P, Potters G, Samson R (2011) Phenotypic variation of baobab (Adansonia digitata L.) fruit traits in Mali. Agroforest Systems 82: 87-97

Dhillion SS, Gustad G (2004) Local management practices influence the viability of the baobab (Adansonia digitata Linn.) in different land use types, Cinzana, Mali. Agriculture, Ecosystems and Environment 101: 85-103

Esselman EJ, Crawford DJ, Brauner S, Stuessy TF, Anderson GJ, Silva OM (2000) RAPD marker diversity within and divergence among species of *Dendroseris* (Asteraceae: Lactuceae). American Journal of Botany 87(4): 591-596

Franzel S, Akinnifesi KF, Ham C (2008) Setting priorities among Indigenous Fruit Tree Species in Africa: Examples from Southern, Eastern and Western African Regions. In: Akinnifesi FK, Leakey RRB, Ajayi OC, Sileshi G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds). Indigenous Fruit Trees in the Tropics. Domestication, Utilisation and Commercialization. Wallingford, UK, pp1-27

Hardcastle PD (1978) A preliminary silvicultural classification of Malawi. Forestry Research Institute of Malawi. Forestry Research Record No. 57

Heywood VH, Watson RT (1995) Global Biodiversity Assessment. Cambridge University Press, United Kingdom

Giang LH, Hong PN, Tuan MS, Harada K (2003) Genetic variation of *Avicennia marina* (Forsk.) Vierh. (Avicenniaceae) in Vietnam revealed by microsatellite and AFLP markers. Genet System 78: 399-407

Gruenwald J, Galiza I (2005) *Adansonia digitata* L. Baobab. Market Brief in the European Union for selected natural ingredients derived from native species. United Nations Conference on Trade and development (UNCTAD)

International Centre for Underutilized Crops (ICUC) (2002) Fruits for the Future. Baobab. Factsheet No.4. March, 2002. ICUC, Southampton, UK

Jama BA, Mohamed AM, Mulatya J, Njui AN (2008) Comparing the “Big Five”: Aframework for the sustainable management of indigenous fruit trees in the dry lands of East and Central Africa. Ecological Indicators 8: 170-179

Jones TH, Stean DA, Jones RC, Pilbeam D, Vaillancourt RE, Potts BM (2006) Effects of domestication on genetic diversity in *Eucalyptus globules*. Forest Ecology and Management 234:78-84

Kalinganire A, Weber JC, Uwamariya A, Kone B (2008) Improving Rural Livelihoods through Domestication of Indigenous Fruit Trees in the Parklands of the Sahel. In: Akinnifesi FK, Leakey RRB, Ajayi OC, Sileshi G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds). Indigenous Fruit Trees in the Tropics. Domestication, Utilisation and Commercialization. Wallingford, UK, pp186-203

Kaushik N, Kumar K, Kumar S, Kaushik N, Roy S (2007) Genetic variability and divergence studies in seed traits and oil content of Jatropha (*Jatropha curcas* L.) accessions. Biomass and Bioenergy 31: 297-502

Kitzmiller JH (1990) Managing Genetic Diversity in a Tree Improvement Programme. Forest Ecology and Management 35: 131-149

Kyndt T, Assogbadjo AE, Hardy, OJ, Kakaïa RG, Sinsin, B, Van Damme P, Gheysen G (2009) Spatial genetic structuring of baobab (*Adansonia digitata*, Malvaceae) in the traditional Agroforestry systems of West Africa. American Journal of Botany 96(5):950-957

Laushman RH (1993) Population genetics of hydrophilous angiosperms. Aquatic Botany, 44: 147-158

Maranz S, Niang A, Kalinganire A, Konaté D, Kaya B (2007) Potential to harness superior nutritional qualities of exotic baobabs if local adaptation can be conferred through grafting. Agroforest Systems. DOI 10.1007/s10457-007-9093-2

Nassar JM, Hamrick JL, Fleming TH (2003) Population genetic structure of Venezuelan Chiropterophilous Columnar Cacti (Cactaceae). American Journal of Botany 90(11): 1628-1637

Pautasso M (2009) Geographical genetics and the conservation of forest trees. Perspective in Plant Ecology, Evolution and Systematics 11: 157-189

Pocktsy JL, Lumaret R, Mayne D, Mohamed VA, Abutaba YIM, Sagna M, Raoseta S, Danthu P (2009) Chloroplast DNA phylogeography suggest a West African centre of
origin for the baobab, *Adansonia digitata* L (Bombacoideae, Malvaceae). Molecular Ecology 18, 1707-1715

Mwase WF, Bjornstad A, Stedje B, Bokosi JM, Kwapata MB (2006) Genetic diversity of *Uapaka kirkiana* Muel. Arg. Populations as revealed by amplified fragment length polymorphism (AFLPs). African Journal of Biotechnology Vol. 5 (13): 1205-1213

Raebild A, Larsen AS, Jensen JS, Ouedraogo M, De Groote S, Van Damme P, Bayala J, Diallo BO, Kalinganire HAS, Kjaer ED (2011) Advances in domestication of indigenous fruit trees in the West African Sahel. New Forests 41: 297-315

Ramadhani, T and Schmidt, E, 2008. Marketing of Indigenous Fruits in Southern Africa. In: Akinnifesi, F.K, Leakey, R.R.B, Ajay, O.C, Sileshi, Tchoundjeu, Z, Matakala, P and Kwesiga, F.R (eds). Indigenous Fruit Trees in the Tropics. Domestication, Utilisation and Commercialization. Wallingford, UK, pp224-236

Saka JDK, Kadzere I, Ndabikunze BK, Akinnifesi FK, Tiisekwa BPM (2008) Product Development Nutritional Value, Processing and Utilization of Indigenous Fruits from the Miombo Ecosystem. In: Akinnifesi, F.K, Leakey, R.R.B, Ajay, O.C, Sileshi, Tchoundjeu, Z, Matakala, P and Kwesiga, F.R (eds). Indigenous Fruit Trees in the Tropics. Domestication, Utilisation and Commercialization. Wallingford, UK, pp288-309

Saka KJD, Msonthi JD (1994). Nutritional value of edible fruits of indigenous wild trees in Malawi. Forest Ecology and Management 64 (1994) 245-248

Scharberg PG, De Hayes DH, Hawley GY, Nijensohn SE (2008) Antropogenic alterations of genetic diversity within tree populations: Implications for forest ecosystem resilience. Forest Ecology and Management 256: 855-862

Sidibe M, Williams JT (2002) Baobab. *Adansonia digitata.* International Centre for Underutilized Crops, Southampton, UK

Sniezko RA, Stewart HTL (1989) Range-Wide Provenance variation in growth and nutrition of Acacia albida seedlings propagated in Zimbabwe. Forest Ecology and Management, 27: 179-197

Sreekumar VB, Renuka C (2006). Assessment of genetic diversity in *Calamus thwaitesii* BECC. (Arecaceae) using RAPD markers. Biochemical Systematics and Ecology 34: 397-405

Syampungani S (2009) Vegetation change analysis and ecological recovery of the Copperbelt Miombo woodland of Zambia. Dissertation, University of Stellenbosch, South Africa

Teklehaimanot Z (2008). The Role of Indigenous Fruit Trees in Sustainable Dryland Agriculture in Eastern Africa. In: Akinnifesi FK, Leakey RRB, Ajayi OC, Sileshi G, Tchoundjeu Z, Matakala P, Kwesiga FR (eds). Indigenous Fruit Trees in the Tropics. Domestication, Utilisation and Commercialization. Wallingford, UK, pp204-223

Venter SM, Witkowski ETF (2011) Baobab (*Adansonia digitata* L.) fruit production in communal and conservation land-use types in Southern Africa. Forest Ecology and Management 261: 630-639

Verma S, Rana TS (2011) Genetic diversity, within and among the wild populations of *Murraya koenigii* (L.) Spreng., as revealed by ISSR analysis, Biochemical Systematics and Ecology 39: 139-144

Wickens GE (1982) The Baobab-Africa's upside-down tree. Kew Bulletin, Vol. 37, No.2: 173-209

Wilson RT (1988) Vital Statistics of the baobab (*Adansonia digitata*). African Journal of Ecology , Volume 26: 197-206

Wright JW (1976) Introduction to Forest Genetics. Academic Press, New York.

Zobel B, Talbert J (1984) Applied Forestry Tree Improvement. John Wiley & Sons, New York. U.S.A