International Journal of Agricultural Policy and Research Vol.4 (3), pp. 35-45, March 2016
Available online at http://www.journalissues.org/IJAPR/
http://dx.doi.org/10.15739/IJAPR.016.006

ISSN 2350-1561

Original Research Article

Effects of watering regime, organic manuring and mycorrhizal inoculation on the growth and development of Shea butter *(Vitellaria paradoxa* C.F.Gaertn) seedlings

Received 18 January, 2016 Accepted 20 February, 2016 Revised 12 March, 2016 Published 22 March, 2016

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In the Sub-Saharan Africa, the Shea tree (*Vitellaria paradoxa* C.F. Gaertn) has widespread occurrence of Shea Parklands across savanna landscapes. Recent demand growth for Shea butter by cosmetic and confectionary industry obtained from shea tree provides opportunities for income generation thus making significant contribution to rural livelihoods and ecosystem services. However, the Shea butter tree is a valuable resource which may become extinct if domestication is not intensified. Two screen house experiments were conducted sequentially to evaluate the effect of watering regime, organic manure application and mycorrhizal (*Glomus mossae*) inoculation on the growth and development of Shea butter seedlings. After the screen house experiment was terminated, the seedlings were planted on the field to assess their performance and survivability. Eight treatments (once a week watering (W_1), once a week watering plus 2 t ha^{-1} of manure (W_1Mn300), once a week watering plus 100 g of mycorrhiza inoculation (W_1My100) and once a week watering plus 300 g of mycorrhiza inoculation (W_1My300), once in two weeks watering (W_2), once in two weeks watering plus 2 t ha^{-1} of manure (W_2Mn300), once in two weeks watering plus 100 g of mycorrhiza inoculation (W_2My100), once in two weeks watering plus 300 g of mycorrhiza inoculation (W_2My300) were fitted into a Completely Randomizd Design (CRD) replicated three times. On the field, 32 plants 4 each from the 8 treatments were fitted into a randomized complete block design (RCBD) replicated three times. Collectively, the variables measured were plant height, stem diameter, number of leaves, crude leaf area, chlorophyll content, mycorrhiza colonization of roots, leaf fall and plant mortality. Shea butter seedlings treated with 2 t ha^{-1} of manure in combination with either once a week or once in 2 weeks watering exhibited significantly ($p < 0.05$) better growth than seedlings from the other treatments. Seedlings inoculated with 100 g of mychorriza and watered once a week (W_1My100) were comparable to those that received 2 t ha^{-1} of manure and once a week watering (W_1Mn300) in number of Shea plant leaves. The survival ability of seedlings on the field was high (75 %) irrespective of treatment received in the screen house.

Key words: Manuring, vegetative methods, *Vitellaria paradoxa,* environmental degradation, mycorrhiza inoculation

INTRODUCTION

The Shea tree (*Vitellaria paradoxa* C.F. Gaertn., syn. *Butyrospermum paradoxum*, Family: Sapotaceae, is indigenous to the semi-arid zone of sub-Saharan West Africa and commonly seen at high densities on intercropped farms in the parkland landscapes of the savanna (guinea, sudan and sahel savanna) agroecologies of West africa (Lovett and Nazmul Haq 2000; Elias 2012). It is a highly valued tree species with its most notable product being the oil, known as Shea butter (French: beurre de karate) obtained from the seeds traditionally harvested by women in this zone.

The growing importance of this tree species, cutting for fuelwood, the threat of over-harvesting (Elias, 2012), has prompted research into domestication including the clonal propagation of this tree (Raebild et al., 2011).

The wooded parklands, of which Shea trees form a high-percentage, also protects against environmental degradation prevalent in this area and contain substantial carbon stores with vast potential for future sequestration for climate change mitigation (Luedeling and Neufeldt, 2012). In the world market, shea butter is highly valued for use in luxury cosmetic (moisturising creams, sun lotions and soaps) or pharmaceutical products (cholesterol-lowering and anti-arthritic remedies). The main demand for shea, however, is for the production of edible stearin, an exotic speciality fat utilised in the formulation of cocoa butter improvers for chocolate confectionary (Lovett 2010; Luedeling and Neufeldt, 2012). The other edible by-product, shea olein, is often used in vegetable spreads, and the residue remaining after oil extraction is used as biofuel. There is rapid growth in the shea industry over the past decade, the Shea tree is therefore a high foreign exchange earner in the West African savanna in the range of US$ 150 million (Lovett 2010; Lovett and Nazmul Haq, 2000). Current populations of Shea trees result from natural regeneration, protected and wild managed during cycles of cultivation, the wild-managed stands produce an unreliable crop in terms of quantity and quality.

In many rural communities of Nigeria Shea butter tree is indiscriminately cut down without regard to its difficult silvicultural nature due to a variety of increasing social, economic, political and environmental pressures (Njiti and Sharpe, 1994; Boffa, 1999). Available statistics show that Shea butter tree is an endangered with a dwindling population in the Sudano-Sahelian regions (Boffa, 1999). Indiscriminate annual bush burning, competition from weeds, and several species of epiphytes drastically reduce yields and destroy many Shea butter trees, thereby further prolonging the long gestation period of vegetative growth, before yield (FAO, 1988). Furthermore the rate of fuel wood consumption across the entire Sudano-Sahelian region is on the increase which is a major factor in the degradation of the land (Odihi, 2003) and a threat to *Vitellaria's* existence. There is an urgent need to improve the silvicultural procedures for Shea butter cultivation in order to increase survivability. The recent demand growth for Shea butter from the international, multi-billion dollar, cosmetic and confectionary sectors, coupled with widespread occurrence of Shea Parklands across savanna landscapes, provides opportunities for income generation and significant contribution to rural livelihoods and ecosystem services (Pouliot, 2012). However, vegetative methods, such as grafting, budding, cuttings and air-layering, have produced varying levels of success (Frimpong et al., 1987; Lovett et al., 1996; Sanou et al., 2004; Yeboah et al., 2010).

Shea butter (*Vitellaria paradoxa)* is of priority for African genetic resources (FAO, 1988) because of its significant economic potential to improve livelihood (Teklehaimanot, 2004; Sanou et al., 2004). Shea butter products were first recognized as an important export for West Africa during the colonial period (Saul et al., 2003).Shea butter tree has multi-purpose value in medicinal and pharmaceutical industries and its fruits have numerous uses.

The cultivation of Shea butter tree is uncommon despite its acclaimed socio- economic values and importance to the local population. This is due to the apparent lack of knowledge about the silvicultural procedures for this species (Djossa et al*.,* 2008 a, b).There has been very scanty research on Shea butter despite its socio-economic value. This may be due to its rather long gestation period. Attempts to domesticate *Vitellaria* for cultivation failed because of its long vegetative growth period of 16 to 20 years before fruiting (Yidana, 2004). This long waiting period is unattractive to both farmers and researchers. In the sudan-sahelian zone of West Africa, people arbitrarily gather the fruits from spontaneous trees to extract butter (Bonkoungou, 1987). The obvious solution to these problems is to bring shea tree under cultivation and intensive management as a tree crop. Irrespective of the long gestation period, the benefits derived from Shea butter tree in Nigeria justify increased scientific attention to the crop through selection, breeding and cultural practices. Generally, *Vitellaria paradoxa* reproduce naturally and could be aided in its reproduction through protection from fire and grazing livestock (Kristensen and Lykke, 2003). Planted *Vitellaria* seedlings may survive to produce high yield and bear fruits more consistently than those in the open bush lands (Yidana, 2004). In a study with *Vitellaria paradoxa* seedlings, Ugese et al*.* (2008) reported over 70 % dry matter allocation to the roots. Savanna species as typified by tamarind and shea butter focus more on early root development as a survival strategy in the harsh savanna environment (Ugese, 2010). In an attempt to integrate *Vitellaria paradoxa* with other trees for both the control of desertification and production of Shea products, improved understandings of its physical and silvicultural characteristics are required. Furthermore, to enhance rapid and successful establishment of Shea plantation, it is necessary to make available to resource poor farmers affordable seedlings with improved vigour, survival and establishment potential. The development of an appropriate silviculture management practice for Shea butter through organic manure, mycorrhizal inoculation and watering to promote seedling vigour is a worthwhile investigation.

Application of single super phosphate (SSP) fertilizer and mycorrhizal inoculation significantly increased height and stem diameter of coffee seedlings (Ibiremo et al*.,* 2011). Mycorrhizal inoculation resulted in consistent and significant improvement of stem diameter and leaf area of coffee seedlings (Ibiremo et al., 2011). However, low root colonization by AM fungi prevented the response by Shea butter trees (Dianda et al*.,* 2009).

Watering regime and mycorrhizal inoculation enhanced growth of gum arabic *Acacia senegal* plants both in the nursery and on the field (Oyun et al*.,* 2010).The work by Dianda et al. (2009) which investigated the response of shea butter seedlings to mycorrhizal inoculation and mineral N and P fertilizers was not conclusive in terms of mycorrhizal effect .The authors recommended further work to elucidate the advantages deriveable from mycorrhizal association for shea seedlings. Due to the ready availability of mycorrhizal fungi and poultry manure which unlike mineral fertilizer increases soil organic matter, it is imperative that the previous treatments evaluated by Oyun et al*.* (2010) and Dianda et al. (2009) be modified by introducing organic manure and ascertain the effects of these treatments on Shea butter, another savanna (Sahelian) economic tree crop.

The objectives of this study are to evaluate (1) the effects of watering regime, mycorrhizal inoculation and organic manure application on growth and development of Shea butter seedlings in the screen house; and (2) the performance and field survival ability of young Shea butter plants under different nursery treatments.

MATERIALS AND METHODS

Location of Study

One screen house experiment was repeated once and one field experiment was conducted once between December 2011 and September 2012 and October 2012 and February 2013, respectively. The 3 experiments were carried out at the Nigerian Institute for Oil Palm Research (NIFOR), Benin City. NIFOR is located on Latitude 6.56 °N and Longitude 5.62° E and is characterized by humid tropical climate and bimodal rainfall pattern. The average annual rainfall is 1761.9 mm with average daily temperature of 32.8 °C.

Treatments and Experimental Design

Eight treatments (W_1, W_1Mn300, W_1My100, W_1My300, W_2, W_2Mn300, W_2My100, and W_2My100) were evaluated. Among the treatments were 2 watering regimes (once a week (W1) and once in two weeks watering (W2), Poultry manure application of 2 t ha^{-1} (Mn300) and mycorrhizal fungi (*Glomus mosseae)* inoculation of 100 g (My100) and 300 g (My300) per plot. The screen house experiments were evaluated under a Completely Randomized Design (CRD) with 3 replications giving a total of 24 plots, whereas the field experiment utilized a randomized complete block design (RCBD) with 3 replications .

The treatments were as follows:

W_1 = Once a week watering

W_1Mn300 = Once a week watering and 2 t ha^{-1} of poultry manure

W_1My100 = Once a week watering and 100 g of mychorrizal inoculation per plot

W_1My300 = Once a week watering and 300 g of mychorrizal inoculation per plot

W_2 = Once in two weeks watering

W_2Mn300 = Once in two weeks watering and 2 t ha^{-1} of poultry manure

W_2My100 = Once in two weeks watering and 100 g of mychorrizal inoculation per plot

W_2My300 = Once in two weeks watering and 300 g of mychorrizal inoculation per plot

Planting Materials

Five months old Shea butter (*V. paradoxa*) seedlings were obtained from the Nigerian Institute of Oil Palm Research (NIFOR) substation Bida, Niger State, Nigeria. Top soil was randomly collected from one site in NIFOR and bulked before filling into black 500 gauge polythene bags (polybags) of 35x30 cm dimension having drainage holes at the bottom. Each polythene bag was filled with 4 kg of top soil. Weeding was carried out regularly by hand picking and hoe weeding within and around the polybags.

Sources of Manure and Mycorrhizal Inoculum

Deep litter poultry manure was obtained from a poultry farm at NIFOR. Arbuscular mycorrhizal was obtained from the International Institute for Tropical agriculture (IITA), Ibadan, Nigeria. Both manure and mycorrhizal were sub sampled and analyzed for their chemical constituents presented in Tables 2 for manure and mycorrhizal, respectively.

Experimental Treatments

Shea butter seedlings were transplanted into the polybags before treatments were imposed. Three hundred ml (300 ml) of water was administered on either a once a week or once in two weeks basis throughout the duration of the experiment. Poultry manure was applied at 300 g per plot which corresponded to 2 t ha^{-1} while crude mycorrhizal inoculum was applied at either 100 g or 300 g per plot. Mycorrhizal placement was below the roots in each plot.

Soil Analysis

A representative sample was obtained from the bulked top soil for routine physical and chemical analysis. In the laboratory, the soil sample was air dried before grinding and sieving. The soil pH was determined in 1:1 soil water suspension ratio and read in a glass electrode pH meter. Organic carbon was determined by the dichromate wet oxidation method of Black (1965) as modified by Allison (1984). The particle size distribution was determined by hydrometer method of (Bouyocous 1951). Exchangeable

Table 1. Physical and chemical properties of the planting soil pre and post planting

Properties	Experiment 1		Experiment 2	
	Pre-planting	Post-planting	Pre-planting	Post-planting
pH	5.98	6.55	6.05	6.75
Organic carbon (%)	1.55	1.44	3.75	1.32
Total nitrogen (g kg^{-1})	0.12	0.21	0.15	0.17
Available P (g kg^{-1})	1.16	7.60	0.50	0.25
Ca (cmol kg^{-1})	2.19	2.85	2.23	3
Mg (cmol kg^{-1})	1.03	1.69	1.21	1.85
Na(cmol kg^{-1})	0.86	1.22	0.91	1.27
K(cmol kg^{-1})	0.12	1.20	0.12	0.82
CEC	4.65	8.32	4.83	8.15
Clay %	4.26	4.30	27	29
Silt%	1.90	1.50	10	12
Sand %	93.83	92.10	56.0	56.0
Textural class	Sandy	Sandy	Sandy clay loam	Sandy clay loam

cations were extracted using IN NH_4OAc, potassium and sodium were determined with flame photometer, whereas calcium and magnesium were determined by EDTA titration. The effective cation exchange capacity (ECEC) was a summation of exchange acidity.

Experimental layout and planting

The experimental field was cleared with the aid of a cutlass and hoe and the debris were packed out of the cleared portion and burnt. The land was tilled slightly to loosen up the soil before transplanting. The plots were marked out to 2 m x 2 m spacing to obtain 24 plots in the experiment. Thirty-two (32) healthy vigorous Shea butter plants from the screen house experiments were trans-planted to the field on the 24 October, 2012 such that there were 4 plants from each of the 8 treatment. Plants were transplanted into holes of 30 cm x 30 cm dimension. The plant with the ball of earth surrounding the roots was placed into the hole after carefully removing the polythene bag .The root system was covered and the soil pressed firmly around it.

Observations and Data Collection

Agronomic variables were collected on a monthly basis for 8 months in the screen house experiment and at 6 months for the field experiment. The variables included plant height (cm), stem girth (cm), crude leaf area (product of length and width) (cm^2), number of leaves, chlorophyll content of leaves, mychorrizal colonization of roots, leaf fall and plant mortality. The height of each plant in the plot was measured with a ruler and recorded. The girth of each plant in the plot was measured with a venier caliper and recorded. The crude Leaf Area (Length and Width) (cm^2) was computed as the product of the length and width of a leaf on each plant was measured and the product represented crude leaf area (Adeyemo et al., 1987) which was recorded. The number of leaves on each plant in the plot was counted and recorded. The chlorophyll concentration in leaf tissues was measured using a chlorophyll content (SPAD) meter (CCM-200 + Apogee).

Leaf fall and plant mortality

This variable was obtained by counting the number of leaves that fell from each plant in each plot. The number of dead Shea butter seedlings in each plot was counted and recorded.

Statistical Analysis

The data collected were subjected to analysis of variance (ANOVA) to test the significance of treatment means using the Statistical Analysis System (SAS) software package version 9.00 (2000). The means were separated using Duncan's Multiple Range Test (DMRT) (Duncan, 1955).

RESULTS

Physical and Chemical Properties of the soil Pre and Post Experimentation

The results of the soil analysis (Table 1) indicated that the chemical properties were affected by the treatments applied. The soil pH before treatment application was slightly acidic but at termination of experiment the pH was raised to near neutral thus creating suitable soil condition for nutrient availability. Similarly, the soil organic carbon was higher before than after the experiment for both cases. The textural classification of the soil used for the experiment is sandy clay loam. Generally, total N was higher after than before the experiment being 0.12 and 0.21 g/kg and 0.15 and 0.17 g/kg for Experiments 1and 2, respectively. Available P was 1.16 before and 7.60 mg/kg after in Expt. 1 but 0.50 before and 0.17 mg/kg after in Expt. 2. Soil CEC was 78.9 % and 68.7 % higher after than before the experiment for Exp 1and 2, respectively(Table1).

Table 2. Nutrient status of Poultry manure and Mycorrhizal innoculum used in the study

Nutrient	Poultry manure	Mycorrhizal fungi Inoculum
N(g kg^{-1})	2.00	7.00
P (g kg^{-1})	3.08	7.50
K (cmol kg^{-1})	2.54	0.04
Ca (cmol kg^{-1})	1.99	1.42
Mg (cmol kg^{-1})	1.28	0.41
Na(cmol kg^{-1})	0.57	0.25

Mineral Composition of Poultry Manure and Mycorrhizal Inoculum

Table 2 shows that the poultry manure had a higher percentage of calcium (Ca), magnesium (Mg), sodium (Na) and potassium (K) than mycorrhizal which was higher in phosphorus (P) and nitrogen (N).

Growth and development of Shea butter seedlings in the screenhouse

Effect of treatments on plant height was highly significant at 2 and 4 months after transplanting (MAT) (Table 3). Watering once a week in combination with 2 t ha^{-1} of manure (W_1M_n300) was statistically superior to watering once in two weeks with 2 t ha^{-1}. Treatments progressively increased the height of Shea butter seedlings across the months with the tallest height (75.55) cm was recorded at 6 MAT for once a week watering in combination with 2 t ha^{-1} of manure (W_1M_n300). However, Shea butter had its shortest height of 11.00 cm at 8 MAT in response to once in two weeks watering W_2. Watering once a week in combination with 100 g of mycorrhizal W_1M_Y100 produced taller seedlings than seedlings inoculated with 300 g of mycorhizal inoculum.

Effect of treatments on number of leaves of shea butter seedlings at 2, 4 and 6 months after transplanting (MAT) (Table 4, combined analysis of the treatments). At 2 MAT, once a week watering and 100 g of mycorrhizal inoculation (W_1M_y100) was similar to once a week watering (W_1) and once in two weeks watering and 300 g of mycorrhizal inoculation (W_2M_y300).

At 4 MAT once a week watering and 2 t ha^{-1} of manure (W_1M_n300) was significantly better than once a week watering and 300 g of mycorrhizal inoculation(W_1M_y300) and once in 2 weeks watering (W_2). At 6 MAT, once a week watering and 100 g of mycorrhizal inoculation (W_1My100) was significantly better than once in 2 weeks watering (W_2) and once in 2 weeks plus 2 t ha^{-1} manure applications (W_2M_n300). Observation also showed that Shea butter had the highest number of leaves (11) at 6 MAT in Expt.1. This treatment was once a week watering and 100 g of mycorrhizal inoculation (W_1M_y100) (Table 5).

At 2 months after transplanting (MAT) once a week watering in combination with 2 t ha^{-1} of manure (W_1M_n300) the plant crude leaf area was superior to once a week watering plus 300 g of mycorrhizal, once in two weeks watering $W_2$and once in two weeks watering plus 2 t ha^{-1} of manure but similar to the other treatments (Table 6). At 4 MAT, once a week watering plus 2 t ha^{-1} of manure (W_2M_n300) was similar to once a week watering (W1) but superior to the other treatments. At 6 MAT, once a week watering plus 2 tha^{-1} of manure (W_1Mn300) was statistically superior to once a week watering plus 300 g of mycorrhizal (W_1MY300), once in two weeks watering (W_2), once in two weeks watering plus 100 g of mycorrhizal (W_2M_Y100) and once in two weeks watering plus 300 g of mycorrhizal (W_2M_Y300) but at par with the other treatments. At 8 MAT, once a week watering in combination with 2 tha^{-1} of manure (W1Mn300) was statistically superior to once a week watering plus 300 g of mycorrhizal (W_1MY300), once in two weeks watering (W_2), once in two weeks watering plus 100 g of mycorrhizal (W_2M_Y100) and once in two weeks watering plus 300 g of mycorrhizal (W_2M_Y300).

Growth and development of shea butter seedlings on the field

The performance of Shea butter plants in terms of (plant height, stem girth, number of leaves, crude leaf area in the field, at 6 months after transplanting (MAT) is presented in Table 7. The stem girth and number of leaves were significantly different depending on the treatments previously received. In respect of stem girth, watering once a week and 300 g of mycorrhizal inoculation (W_1M_y300) was statistically superior to once a week watering and 2 t ha^{-1} of manure (W_2M_n300) , once in two weeks watering (W_2), once in two weeks watering plus 2 t ha^{-1} of manure ($W1M_n300$) and once in two weeks watering plus 300 g of mycorrhizal (W_2M_y300).On the other hand, watering once in 2 weeks in combination with 100 g of mycorrhizal (W_2M_y100) exerted a significant effect on the number of leaves in the field. Plants that had received this treatment had more leaves than those previously treated with once a week watering and 2 t ha^{-1} of manure (W_1Mn300), once a week watering in combination with 100 g of mycorrhizal (W_1My100) and once in two weeks watering plus 2 t ha^{-1} of manure (W_2M_n300). In terms of number of leaves of Shea butter plants the highest value (9) was recorded by seedling that had received (W_2M_y100) treatment.

Table 3. Effect of treatment on plant height of shea butter in experiment 1, 2 and combine analysis at 2, 4, 6, and 8 months after transplanting (MAT)

	2MAT			4MAT			6MAT			8MAT		
Treatment	**Exp 1.**	**Exp 2**	**Comb.**	**Exp 1.**	**Exp 2**	**Comb.**	**Exp 1.**	**Exp 2**	**Comb.**	**Exp 1.**	**Exp 2**	**Comb.**
W_1	14.70	14.67^{c}	14.68^{abcd}	18.00	19.40^{abcd}	18.70^{ab}	23.17	25.33	60.01	23.60	23.67	23.33
W_1M_n300	14.33	22.40^{a}	18.37^{a}	17.317	25.67^{ab}	21.42^{ab}	22.73	26.40	75.55	23.47	25.73	24.60
W_1M_y100	17.33	15.27^{c}	16.30^{ab}	18.70	18.33^{bcd}	18.52^{ab}	24.70	23.00	58.16	27.17	25.23	26.30
W_1M_y300	11.40	13.20^{c}	12.30^{bcde}	12.37	22.87^{abc}	17.62^{abc}	14.90	24.83	59.14	14.97	25.13	20.05
W_2	11.57	11.33^{c}	11.45^{de}	13.7	19.50^{abcd}	16.28^{bc}	14.57	22.00	41.02	11.00	24.27	17.63
W_2M_n300	11.00	11.67^{c}	11.33^{e}	12.23	12.40^{de}	12.32^{d}	40.13	17.17	29.99	16.77	20.10	18.43
W_2M_y100	17.33	15.50^{c}	16.42^{ab}	14.20	17.83^{bcd}	16.02^{abc}	24.23	24.83	52.59	20.67	29.23	24.95
W_2M_y300	13.90	13.57^{c}	13.73^{bcde}	14.13	18.23^{bcd}	16.18^{bc}	21.73	26.50	61.70	22.20	23.30	22.75
Significance	Ns	**	**	ns	*	*	ns	ns	ns	ns	ns	ns
SE		1.72	1.45		2.82	2.02						

Means in a column followed by the same letters are not significantly different using Duncan's Multiple Range Test (DMRT) at 5 % level.W1= once a week watering, W2= once in two weeks watering, Mn300= 2 t ha^{-1}manure , My 100=100 g per plot of mycorrhizal inoculation , My300= 300 g per plot of mcorrhizal , Comb=Combined analysis, SE=Standard error of mean and MAT=Months after transplanting,Exp=Experiment

Table 4: Effect of treatment on Stem girth of shea butter in experiment 1, 2 and combine analysis at 2, 4, 6, and 8 months after transplanting (MAT)

	2 MAT			4 MAT			6 MAT			8 MAT		
Treatment	**Exp 1.**	**Exp 2**	**Comb.**	**Exp 1.**	**Exp 2**	**Comb.**	**Exp 1.**	**Exp 2**	**Comb.**	**Exp 1.**	**Exp 2**	**Comb.**
W_1	5.67	1.87^{bcd}	2.05	1.6^{abc}	2.00	1.80^{abc}	1.90	2.50	2.20	1.93	2.13	2.03
W_1M_n300	5.00	2.67^{a}	2.03	2.10^{ab}	2.70	2.40^{a}	2.23	2.45	2.4	2.17	2.63	2.40
W_1M_y100	10.33	1.83^{bcd}	2.00	2.17^{ab}	2.20	2.18^{ab}	2.63	2.57	2.57	2.567	2.2	2.43
W_1M_y300	3.00	1.73^{bcd}	1.77	1.30^{dc}	2.07	1.68^{bc}	1.67	2.00	2.00	1.37	2.07	1.72
W_2	5.33	1.57$^{d\ c}$	1.60	1.30^{dc}	2.10	1.70^{bc}	1.67	2.12	2.12	0.97	2.10	1.53
W_2M_n300	3.67	2.23^{abc}	2.20	1.87^{abc}	2.13	2.00^{abc}	1.27	1.85	1.85	1.47	2.13	1.80
W_2M_y100	5.33	1.80^{bcd}	1.95	1.47^{a}	2.30	1.88^{abc}	2.37	2.23	2.23	1.87	2.30	2.08
W_2M_y300	7.33	1.97^{bcd}	1.82	1.87^{a}	1.90	1.88^{abc}	2.13	2.32	2.32	2.07	1.90	1.98
Significance	Ns	*	Ns	*	Ns	*	ns	ns	ns	ns	ns	ns
SE		0.22		0.26		0.22						

Means in a column followed by the same letters are not significantly different using Duncan's Multiple Range Test (DMRT) at 5 % level.W_1= once a week watering, W2= once in two weeks watering, Mn300= 2 t ha^{-1}manure , My 100=100 g per plot of mycorrhizal inoculation , My300= 300 g per plot of mycorrhizal , Comb=Combined analysis, SE=Standard error of mean and MAT=Months after transplanting, Exp=Experiment.

DISCUSSION

Effect of watering regime

The results of this study showed that the growth variables of Shea butter seedlings responded significantly to watering regimes imposed. These treatments increased plant height, stem girth, number of leaves and crude leaf area. This observation is consistent with report by Majumdar (2010), who stated that when adequate water is available, plant cells remain turgid and plants retain their form and structure. Weekly watering was statistically superior to once in 2 weeks watering. This is at odds with the report of Oyun et al.(2010),

Table 5: Effect of treatment on number of leaves of shea butter in experiment 1, 2 and combine analysis at 2, 4, 6, and 8 months after transplanting.

	2MAT			4MAT			6MAT			8MAT		
Treatment	**Exp 1**	**Exp 2**	**Comb.**	**Exp 1**	**Exp 2**	**Comb.**	**Exp 1**	**Exp 2**	**Comb.**	**Exp 1**	**Exp 2**	**Comb.**
W_1	6	7	6^{ab}	2	7^{a}	2^{abc}	5^{bc}	10^{a}	7^{abc}	6	9	8^{ab}
W_1M_n300	5	5	5^{bcd}	2	5^{ab}	2^{a}	6^{bc}	10^{a}	8^{abc}	5	9	7^{ab}
W_1M_y100	10	8	9^{a}	2	7^{a}	2^{ab}	11^{a}	6^{abc}	9^{a}	8	6	7^{ab}
W_1M_y300	3	5	4^{bcd}	1	3^{b}	2^{b}	4^{bc}	7^{ab}	6^{abd}	4	8	7^{ab}
W_2	5	3	3^{cd}	1	3^{b}	2^{bc}	3^{bc}	7	5^{bcd}	4	7	5^{bc}
W_2M_n300	4	2	3^{d}	2	2^{b}	2^{abc}	4^{bc}	5^{bc}	4^{dc}	5	5	5^{bc}
W_2M_y100	5	4	5^{bcd}	2	2^{b}	2^{abc}	6^{bc}	7^{abc}	7^{abc}	9	5	7^{ab}
W_2M_y300	7	5	6^{ab}	2	6^{ab}	2^{abc}	6^{ab}	10^{a}	8^{abc}	8	6	7^{ab}
Significance SE	Ns	ns	**	ns	*	**	*	*	*	ns	ns	*
			1.02		1.20	0.84	1.67	1.37	1.19			1.16

Means in a column followed by the same letters are not significantly different using Duncan's Multiple Range Test (DMRT) at 5 % level.W_1= once a week watering, W2= once in two weeks watering, Mn300= 2 t ha^{-1}manure, My 100=100 g per plot of mycorrhizal inoculation , My300= 300 g per plot of mcorrhizal , Comb=Combined analysis, SE=Standard error of mean and MAT=Months after transplanting,Exp=Experiment

Table 6 Effect of treatment on Crude Leaf Area of shea butter at 2, 4, 6, and 8 months after transplanting (Experiment 1, 2 and combine analysis)

	2MAT			4MAT			6MAT			8MAT		
Treatment	**Exp 1.**	**Exp 2**	**Comb.**	**Exp 1.**	**Exp 2**	**Comb.**	**Exp 1.**	**Exp 2**	**Comb.**	**Exp 1.**	**Exp 2**	**Comb.**
W_1	40.08	37.08	38.58^{abcd}	62.67^{ab}	75.93^{abcd}	69.30^{ab}	98.05^{a}	67.00	82.53	86.77^{ab}	77.62	82.20
W_1M_n300	39.1	43.34	41.23^{abc}	82.62^{a}	87.17^{abc}	84.90^{a}	98.44^{a}	72.58	85.51	93.92^{a}	91.77	92.85
W_1M_y100	31.42	30.70	35.33^{abcd}	44.13^{bcd}	40.51^{bcd}	42.32^{bc}	66.06^{ab}	46.33	56.20	85.67^{ab}	79.41	82.54
W_1M_y300	39.97	26.90	20.84^{de}	15.55^{d}	58.98^{bcd}	37.32^{bc}	29.08^{cd}	99.17	64.13	29.57^{c}	86.13	57.85
W_2	14.79	19.78	21.59^{de}	25.03^{cd}	38.71^{bcd}	31.87^{c}	30.61^{cd}	66.83	48.72	26.87^{c}	69.80	48.34
W_2M_n300	10.94	16.37	17.39^{e}	27.50^{bcd}	34.9^{cd}	31.20^{c}	21.81^{ab}	30.67	26.24	35.85^{c}	49.70	42.78
W_2M_y100	23.90	42.84	45.80^{ab}	35.110^{bcd}	59.53^{bcd}	47.32^{bc}	55.40^{bc}	64.67	60.03	46.48^{bc}	95.23	70.86
W_2M_y300	48.7	39.33	28.37^{bcde}	30.93^{bcd}	47.44^{bcd}	39.19^{bc}	54.57^{bc}	87.67	71.12	50.55^{ab}	79.87	65.86
Significance	Ns	ns	**	*	*	**	**	ns	ns	*	ns	ns
SE			5.92	12.31	18.56	11.75	13.51			14.72		

Means in a column followed by the same letters are not significantly different using Duncan's Multiple Range Test (DMRT) at 5 % level.
W_1= once a week watering, W2= once in two weeks watering, Mn300= 2 t ha^{-1}manure , My 100=100 g per plot of mycorrhizal inoculation , My300= 300 g per plot of mcorrhizal , CoTmb=Combined analysis, SE=Standard error of mean and MAT=Months after transplanting,Exp=Experiment

who observed the opposite with *Acasia senegal* seedlings. This may be attributed to the differences between the tree species studied and the prevailing environmental conditions under which the studies were conducted.

Weekly application of water (W_1) was more profitable to Shea butter seedlings than the once in two weeks watering. This implies that the seedlings require consistently humid soil to function effectively. The once in two weeks watering (W_2) would have presented longer periods of dryness than the once a week application of water. This should be borne in mind especially when shea butter is grown in its traditional sahel region where moisture scarcity is the norm.

Table 7. Effect of treatments on (plant height, stem girth, number of leaf, crude leaf area of shea butter seedlings) in the field, 6 month after transplanting

Treatment	Plant height(cm)	Stem girth(cm)	Number of leaves	Crude leaf area (cm^2)
W_1	27.00	2.67^{ab}	6^{abcd}	63.71
W_1M_n300	25.87	1.70^{b}	3^{cd}	62.67
W_1M_y100	26.60	2.90^{ab}	3^{cd}	111.63
W_1M_y300	20.67	4.20^{a}	7^{abc}	78.50
W_2	28.33	1.23^{b}	5^{abcd}	92.92
W_2M_n300	25.53	2.00^{b}	4^{bcd}	45.44
W_2M_y100	36.37	2.67^{ab}	9^{a}	154.84
W_2M_y300	27.13	1.10^{b}	8^{ab}	76.81
Significance	ns	*	*	ns
SE		0.65	1.58	

Means in a column followed by the same letters are not significantly different using Duncan's Multiple Range Test (DMRT) at 5 % level.W_1= once a week watering, W2= once in two weeks watering, Mn300= 2 t ha^{-1}manure , My 100=100 g per plot of mycorrhizal inoculation , My300= 300 g per plot of mcorrhizal , Comb=Combined analysis, SE=Standard error of mean and MAT=Months after transplanting,Exp=Experiment

Effect of Mycorrhizal Inoculation

The significant effect of mycorrhizal inoculation on the vegetative variables of Shea butter seedlings in the screen house was fewer than that of poultry manure on Shea butter seedlings. This agrees with other studies conducted on mycorrhizal inoculation of trees which showed variable results on the growth of the plants (Rao et al., 1990., Ikram et al., 1991; Dania and Fagbola, 2007). Ba et al. (2000) observed that the strain *G.intraradices* was invasive but not efficient in increasing growth of three sahelian fruit trees. The authors obtained relatively low efficiency of *G.intraradices* strains using a larger set of fruit tree species from the Sahel area. The effects of mycorrhizal inoculation on shea butter seedlings were similar to those of several other sahelian fruit trees that were mycorrhized which showed inherently no increases in growth due to AM fungal inoculation (Ba et al.,2000). The growth response of shea butter seedlings in this study was promoted by mycorrhizal inoculation unlike in the aforementioned studies. This is because plants inoculated with mycorrhizal exhibited growth factors similar to their fertilized counterparts in some cases. The reason may be attributed to mycorrhizal root colonization which was relatively high (59.0%) for plants inoculated with 300 g of mycorrhizal compared to 12 % colonization reported by Dianda et al.(2009) which did not increase plant growth. In another study, plant height and stem diameter of coffee seedlings were significantly increased by SSP and mycorrhizal inoculation (Ibiremo et al.2011) which corroborates the findings in the study. Response by shea butter seedlings to mycorrhizal inoculation may depend on the type of fungi inoculated.

In mycorrhizal-Acacia relationships, plant responses were greater from endomycorrhizal than ectomycorrhizal inoculation (Oyun et al., 2010). In addition, Hajibaland et al. (2009) observed differences in responsiveness when both *Glomus mosseae* and *Glomus intraradices* were inoculated on rice.

Effect of Organic Manure

Manure application significantly affected the vegetative parameters of Shea butter seedlings due to adequate mineralization of the poultry manure. Shea butter seedlings treated with 2 t ha^{-1} of manure in combination with once a week watering (W_1Mn300) exhibited significantly better growth in plant height, stem girth, number of leaves and crude leaf area than seedlings from other treatments. A similar trend of response was earlier observed by Tiwari et al. (2009) who reported that the inclusion of manure to the fertilization schedule improved the organic carbon status and available N,P,K and S in the soil which sustained soil health and improved plant growth. The significant response of Shea butter seedlings to the application of poultry manure in the screen house was consistent with findings of Mando et al. (2005) who stated that soil organic matter and crop performance was better maintained using organic materials with low C:N ratio such as manure than those with high C:N ratio such as straw.

However, inorganic fertilization stimulated plant height, collar diameter and dry weights of Shea butter seedlings compared to control treatments (Dianda et al.,2009).

Stem girth and number of leaves of Shea butter seedlings on the field

An important implication of the physio-chemical soil analysis is that the initially low soil nutrients increased after organic manure was applied, The soil pH rose from slightly acidic (5.98) to near neutral (6.55) which is conducive for adequate crop growth. This confirms the findings of Akande et al. (2003) that application of organic materials ameliorate slightly acidic tropical soils to improve crop production. The application of the organic materials also increased soil available P and total nitrogen showing the efficiency of organic materials as sources of both nitrogen and phosphorus to the soil. Organic manure combined with inorganic fertilizer also offers a promising combined source of nutrition for shea butter seedlings growth and survival in the field. This is because shea butter seedlings responded positively to inorganic fertilization (Dianda et al., 2009), although the ratio of N: P was crucial to the response observed.

Leaf fall, chlorophyll concentration, seedling mortality on the field

Growth performance and survival of Shea butter seedlings on the field was good and stable. Plants previously treated with once a week watering plus 300 g of mycorrhizal produced wider stem girth in the field while the number of leaves of Shea butter seedlings was significantly increased in plants that received once in 2 weeks watering plus 100 g of mycorrhizal (W_2My100). This may be due to the activity of the mycorrhizal fungi in enabling the shea seedling to explore a greater volume of soil for increased nutrient absorption which is in line with the report of Smith et al. (1997). This is consistent with report by Ibiremo et al. (2011) that mycorrhizal inoculation increased the stem diameter of tree crops. It is probable that mycorrhizal root colonization increased the ability of the seedlings to take up nutrients which increased their vegetative growth. Levy and Kirkum (1983) and Read and Boyed (1986) noted that mycorrhizal inoculation increased soil water extraction and root hydraulic conductivity in plants. Generally, these findings imply that Shea butter seedlings should be treated with manure and mycorrhizal to increase their chances of survival on the field. These findings also support earlier observations by Oyun et al. (2010) that mycorrhizal inoculation and watering twice weekly enhanced growth and seedling survival of *Acasia senegal* seedlings.

Leaf fall, chlorophyll concentration, mychorrhizal colonization and plant mortality on the field was similar for Shea butter plants irrespective of earlier treatments received. The implication of this is that Shea butter plants are able to overcome initial deficiencies and perform satisfactorily on the field provided root growth which in this case was enhanced by mycorrhizal is adequate. The leaf chlorophyll content of shea seedlings inoculated with mycorrhizal was not significantly higher than that of the non-inoculated seedlings. This is in contrast with Bello (2005), who stated that mycorrhizal inoculation enhanced total chlorophyll content. However, the 300 g mycorrhizal inoculated seedlings had higher chlorophyll content than the 100 g mycorrhizal seedlings. This infers that higher mycorrhizal inoculation is needed to increase chlorophyll concentration of Shea butter seedlings. Root colonization of shea seedlings treated with once a week watering plus 300 g of mycorrhizal (W_1My300) was highest at 59 %.This further confirms that higher levels of mycorrhizal inoculation will furnish better results for Shea butter plants. On the field, 75 % of the plants survived. This buttresses the hardiness of Shea butter plants. It also connotes that initial treatment of seedlings may not affect their survivability provided adequate cultural protocols are carried out to effectively protect the plants on the field. Furthermore, survival of Shea butter seedlings planted in the field could be enhanced by securing them with wire collar and protecting them from bush fires (Kristensen and Lykke 2003).

Conclusion

The recent demand growth for Shea butter from the international cosmetic and confectionary industry, a plant species with widespread occurrence of in parklands across savanna landscapes, provides opportunities for income generation, thus making significant contribution to rural livelihoods and ecosystem services. The experiment revealed that watering once a week in combination with 2 t ha^{-1} of manure application (W_1Mn100) influenced ($p < 0.05$) plant height of Shea butter seedlings better than the other treatments. Application of 2 t ha^{-1} of manure in combination with either once a week or once in 2 weeks watering (W_1M_y300 and W_2M_y100) produced the highest Shea butter stem girth and leaf area. Mycorrhizal inoculation (100 or 300 g) was comparable to manure application (2 t ha^{-1}) in the ability to increase number of leaves for Shea butter seedlings. Seedlings previously treated with 100 g of mycorrhizal inoculation combined with once in 2 weeks watering (W_2My100) produced the highest number of leaves in the field. Shea butter seedlings should be watered once a week or more and supplied with 2 t ha^{-1} of poultry manure along with 300 g of arbuscular mycorrhizal to enhance growth and development.

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