Hardening watering interval of East African greenheart (*Warburgia ugandensis*), nursery seedlings in East Mau watershed, Njoro, Kenya

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Abstract

Water has been a scarce resource in agricultural areas and is essential in the establishment of nursery seedlings. This requires the effective and efficient use by farmers of this important resource. Hardening nursery seedlings through the reduction of watering regimes is necessary before transplanting seedlings to the field. This leads to better survival but when the interval of watering is not well established, and this might affect species and locality. An experiment was set up during the dry season just before planting out to determine the best hardening watering interval for East African greenheart (*Warburgia ugandensis*) seedlings in Egerton University, East Mau watershed, Njoro, in Kenya. The experiment was laid down as a Completely randomized design (CRD) with 5 treatments replicated 3 times. Treatments constituted of different watering intervals, which were: twice daily, once daily, 2 days, 4 days and 6 days. These treatments were applied for 60 days on 270 days old seedlings from January to March 2018. Analysis of variance (ANOVA) was used to determine treatment differences while DMRT determined the significantly different treatment means at $p \leq 0.05$. The results revealed that the best growth was shown by twice daily, once daily, 2 days and 4 days intervals for shoot biomass, total plant biomass and total leaf area. However, the recommended watering interval is 4 days since it showed good growth for most of the variables with minimal water use equivalent to 12.5% of the water used by seedling watered twice daily which saves 87.5% of the water applied. The results are applicable in the Kenyan highlands for Warburgia and other leafy succulent plants. However further studies needs to be done for other non-succulent species using different pot sizes and soil mixtures.

Keywords: Nursery, seedlings, Warburgia ugandensis, bedding plant, watering interval

1.0 Introduction

In Africa, *Warburgia ugandensis* is commonly known as East African greenheart, a native species of evergreen trees, mainly found growing in Kenya, Ethiopia and some parts of western

Africa. The leaves are succulent and can be used to flavor curries while the extract has some antimalaria, antifungal and antibacterial properties (Olila *et al*., 2001; Were *et al*., 2010)

Water is an important natural resource vital for plant growth, but there is a growing concern about its availability (Goyne & McIntyre, 2003) in the next future. With the effects of climate change, water is scarce in most geographical zones globally (Morrison *et al*., 2009). Previous permanent water supply sources are now seasonal hence posing serious challenges to nursery establishment and management, especially in the drier regions.

Plants vary greatly in their response to watering intervals depending on edaphic and environmental conditions. Hardening is essential in the nursery through reducing watering of seedling 4-8 weeks before transplanting with the purpose of acclimatization (Inoti, 2001). Changes in water availability can affect seedling resource allocation (Blain and Kellman, 1991) because this affects water uptake and photosynthesis, further modifying growth and survivorship of transplanted seedlings. An experiment was set up 60 days before transplanting to determine the ideal watering interval for Warburgia nursery seedlings during hardening in the eastern Mau catchment of Kenya.

The young growth of seedlings mainly depends on food reserves contained in the cotyledons, and next from soil moisture availability. After depletion of food reserves, seedlings rely on manufactured photosynthetic reserves for growth, development and survivorship (Bargali and Tewari, 2004). Soil moisture is key in nutrient uptake from the growth media to support growth (Shao *et al*., 2008). Many vital processes in plants such as cell division, cell elongation, stem as well as leaf enlargement and chlorophyll formation depend on plant water availability.

Regular watering is required under nursery conditions to produce cost effective seedlings (Inoti, 2001 and Simon *et al*., 2011) because stagnation in growth or subsequent mortality translates into economic losses to nursery owners. Low water supply can result to huge losses because seedlings take long to reach an appropriate size (0.3 m height) for grafting (Mhango *et al*., 2008), transplanting and for sales.

Water is the main constituent of living organisms involved in photosynthesis process thus its availability at acceptable interval affects productivity of many plants. Water stress reduces the

growth of plants in terms of leaf number, leaf area, biomass weight and stomata conductance (Hartmann *et al*., 2005). Root development is more preferred over foliage in arid environments and extensive rooting is a special adaptation for plant survival in these areas (Abbott, 1984; Inoti, 2018).

Water is an important factor in dry land forestry and it is vital in tree growth and development in the tropics (Awodola and Nwoboshi, 1993). According to Miller *et al.* (1999), for each ton of vegetative growth, hundreds of tons of water may be consumed by the growing plant especially in arid sites. As observed by Awodola (1984), the decrease in relative water content affects physiological processes and plant growth. Earlier works by Huang *et al.* (1985) reported that root to shoot ratio to be 3.5 times higher in water stressed plants compared to those with no stress.

2.0 Material and methods

The seeds were collected from mature trees at Egerton University in Njoro.

2.1 Study site

The investigation was conducted at Agroforestry tree nursery, Egerton University, Njoro, in Kenya, within the eastern Mau water-catchment. The University is located in Njoro, a small community approximately 25 kilometers southwest of the town of Nakuru. This is located approximately 182 kilometers, northwestern Nairobi. The study site 0°22'11.0"S Latitude, 35°55'58.0"E Longitude and an altitude of 2,238 m. The area falls in agro ecological zone Lower Highland 3. The experimental site receives annual rainfall of 1200 mm where the distribution is bimodal with long period between April and August and short period between October and December. The temperature ranges from 10.2 to 22.0°C (Ngetich *et al*., 2014) while the soils are mollic andosols (Kinyanjui, 1979). The experimental period received low rainfall especially from January to February (11.5 mm) which were coupled with moderately high temperatures (21.9 ^{0}C) (Table 1).

Table 1: Rainfall and temperature during the experimental period (from January to March 2018)

Months/parameters recorded	January 2018	February 2018	March 2018
Total rainfall (mm)	11	12.3	194
No. of rainy days	3	4	19

Average temperature (^{0}C)	21.2	22.6	19.6
Maximum temperature (^{0}C)	22.9	23.7	22.3
Minimum temperature (^{0}C)	17.6	21.0	15.8
Source: Department of Water Engineering, at Egerton University, in Njoro			

2.2 Experimental design

The experiment was laid down in a Completely randomized design (CRD) with 5 treatments replicated 3 times. The treatments were different watering intervals which were: twice daily, once daily, 2 days, 4 days and 6 days. The amount of water applied in each pot per watering was 200 milliliters. Forest soil used as the potting media had a chemical composition of 1.05% N, 0.15% P, 0.89 % K, 1.42% Ca and 0.25% Mg, while the polythene pot size was 9x20 cm, in width and length respectively. The pot was elliptical in shape and contained a volume of 1,272 cm^3. The seedlings were raised in the nursery for 270 days before being randomly selected for the experiment. Each treatment consisted of 10 plants per replicate which were then raised in the nursery for 60 days. Out of these, 7 seedlings were randomly sampled for assessment. The experiment was carried out from Mid January to Mid March 2018, during the dry period.
The dependent variables were: height, root collar diameter, third internode length, number of leaves, leaf length and width, leaf area, seedling sturdiness quotient, root length, plant and root biomass. Seedling sturdiness quotient (SSQ) = shoot height (cm) divided by root collar diameter (mm) and smaller the quotient the better sturdiness.

2.3 Data analysis

Data of the measured variables was subjected to statistical analysis using ANOVA model procedures of Genstat statistical package (2013). Variations between the treatment means were compared using Duncan's Multiple Range Test (DMRT) at $P \leq 0.05$. Furthermore, the

coefficient of variation (CV %) was calculated to reveal the relative measure of variation that existed within the data.

3.0 Results and discussion

Seedling growth was relatively uniform for all the treatments ranging from twice daily to 4 days interval. However, the 6 days watering interval compromised the growth for most of the shoot, foliage and root variables. According to Levy and Krikum (1983), low water levels in plants below a critical level usually triggers changes in all structures sometimes leading to the death of the plants. This is further supported by earlier observations by Awodola (1984) and Farah (1996), who reported that water scarcity reduces growth, yield and other physiological processes.

3.1 Effect of watering interval on shoot growth of *Warburgia ugandensis* nursery seedlings

Results showed that shoot biomass and total plant biomass for twice daily, once daily, 2 days and 4 days interval were significantly ($P \leq 0.05$) higher compared with 6 days interval (Table 2). These findings corroborate with studies by Abo El-Khei (2000) who reported that water stress can lead to low shoot dry weight. Water deficiency imposes huge reductions in crop yield through diminished leaf carbon fixation and general growth inhibition (Chaves & Oliveira, 2004).

More recent studies by Daba and Tadese (2017) also concluded that *Moringa oleifera* and *Grevillea robusta* watered twice daily with 1.5 liters and *Cordia africana* watered twice after one day with 2 liters per plot ensures good growth performance of those tree seedlings species. These findings therefore have implications on water wastage, reduced labor costs and maximizing profitability of tree seedlings production.

Table 2: Effect of watering interval on shoot growth of *Warburgia ugandensis* nursery seedlings

Watering interval	Bedding plant height	Third internode length	Shoot biomass	Total bedding plant biomass	Seedling sturdiness quotient
	cm	mm	g	g	unit
6 days	44.03[b]	8.67	8.47[b]	13.20[b]	378.70[b]

4 days	46.83[ab]	10.00	10.57[a]	16.37[a]	439.30[ab]
2 days	46.30[ab]	9.33	10.27[a]	16.47[a]	423.30[ab]
Once daily	48.50[a]	10.33	10.60[a]	17.27[a]	480.70[a]
Twice daily	48.00[a]	11.00	10.87[a]	16.87[a]	469.00[a]
CV%	3.6	12.0	8.8	9.1	8.1
SED	1.37	0.97	0.73	1.19	28.93

Means with different superscript letters within the columns differ significantly using DMRT at P≤0.05.

According to Olaoye and Oyun (2019), two or three times seedlings watering per 7 days of *Terminalia ivorensis, Terminalia superba, Cleistopholis patens and Mansonia altisima* is very good for improving the physiological growth which can enhance the domestication and further cultivation of these seedlings in their environment. Excessive water encourages the growth of microorganisms such as bacteria and fungi which can cause disease in the seedlings. Similarly, Oyun *et al.* (2010) reported that, watering twice in 7 days is more suitable for seedlings of *Acacia senegal* in the nursery. This is true since daily watering produces fragile and succulent seedlings that are unable to survive the harsh field conditions. This corroborates with earlier observations by Awodola (1984) and Huang *et al.* (1985).

In contrast, height and seedling sturdiness quotient (SSQ) for twice (48 cm and 469 respectively) and once daily (48.5 cm and 480.7 respectively) were significantly higher compared with 6 days interval (44.03 cm and 378.7 respectively) which was the lowest. The lowest SSQ shows the best sturdiness and survival in the field, which tends to decrease with reduction in watering.

3.2 Effect of watering interval on foliage growth of *Warburgia ugandensis* nursery seedlings

Watering twice daily showed the highest number of leaves (16.23) and total leaf area (436.7 cm^2) which were significantly ($P \leq 0.05$) higher compared with all the other treatments except once daily interval (14.93 and 418 cm^2 respectively) (Table 3). On the other hand, 6 days interval showed significantly the lowest total leaf area (289.3 cm^2) compared with all the other treatments. These findings are in agreement with earlier studies by McMaster and Smike (1988) who explained that during vegetative growth, phyllochron decreases under water stress leading to leaves becoming smaller, which consequently results in low leaf area index.

Table 3: Effect of watering interval on leaf growth of *Warburgia ugandensis* nursery seedlings

Watering interval	Number of leaves	Leaf length	Leaf width	Single leaf area	Total leaf area
		mm	mm	cm^2	cm^2
6 days	11.6^{c}	109.30	29.67	24.90^{c}	289.30^{d}
4 days	13.47^{bc}	114.30	32.33	25.77^{c}	347.70^{c}
2 days	13.27^{bc}	116.00	31.00	27.80^{ab}	369.00^{bc}
Once daily	14.93^{ab}	123.70	33.00	28.00^{a}	418.00^{ab}
Twice daily	16.23^{a}	115.30	32.67	26.90^{b}	436.70^{a}
CV%	8.1	8.1	8.0	2.0	8.4
SED	0.93	7.64	2.07	0.44	25.5

Means with different superscript letters within the columns differ significantly using DMRT at P≤0.05.

However, once daily interval showed the highest single leaf area (28 cm^2) which was significantly higher compared with all the other treatments except 2 days interval (27.8 cm^2). Hartmann *et al*. (2005) reported that water stress decreases the growth of plants in terms of leaf number, leaf area and stomata conductance which is consistent with the current findings.

3.3 Effect of watering interval on root growth of *Warburgia ugandensis* seedlings

Watering once daily showed the highest root collar diameter (9.9 mm) and root length (24.93 cm) which were significantly higher compared with 6 days interval (8.6 mm and 21.33 cm respectively) (Table 4).

Table 4: Effect of watering interval on root growth of *Warburgia ugandensis* nursery seedlings

Watering interval	Root collar diameter	Root length	Root biomass
	mm	cm	g
6 days	8.60^{b}	21.33^{b}	4.73
4 days	9.37^{ab}	22.93^{ab}	6.00
2 days	9.13^{ab}	22.77^{ab}	5.80
Once daily	9.90^{a}	24.93^{a}	6.67
Twice daily	9.77^{a}	23.73^{ab}	6.20
CV%	5.5	7.2	20.5
SED	0.42	1.36	0.98

Means with different superscript letters within the columns differ significantly using DMRT at P≤0.05.

Further results showed that watering twice daily tends to compromise growth of roots in the highlands and this was observed by once daily showing the highest growth for the variables measured though not significant. According to Komer *et al.* (1999), excess water in plant cells may retard physiological processes. For example, stomata conductance, a numerical measure of the maximum rate of passage of water vapour for transpiration or carbon dioxide through the stomata for photosynthesis is influenced by the soil-water balance (Komer *et al.*, 1999). Simon *et*

al. (2011) reported 2 day watering interval for *Persia americana* and *Vangueria infausta* showing the highest root collar diameter growth while Daba and Tadese (2017) also recorded similar results with *Cordia africana* after 1 day interval.

Studies by ElHadi *et al* (2013) showed that moderate stress (9 days watering interval) facilitated seedling height development compared to frequent irrigation (3 and 6 days). While longer period (12 days) resulted in negative impact on the seedling development by affecting the water potential. With moderate stress, the carbohydrates are transported to the root system. The root system benefits from this supply by being under less stress compared to the shoot, leading to better growth (Hsiao 1973). Moderate stress also has minute effect in carbon uptake Nitrogen and mineral uptake by the actively growing root system may concentrate in the shoot.
The results of the current research showed that the watering interval of 4 days was the most ideal for Warburgia seedlings raised in the highlands. This is consistent with other recent studies by Inoti (2018) who recommended 4 days interval for jojoba seedlings while Sale (2015) also recommended 3 and 5 days interval for *Parkia biglobosa* in dry areas. Similar work carried out on *Acacia senegal* by Isah *et al.* (2013) indicated that the species performed better when watered once in three days and this reflects its capability to cope with drought stress. More recent studies by Inoti and Cherop (2022) recommended 2 days watering interval in *Prunus africana* seedlings which is a non- succulent species in the Kenyan highlands.

4.0 Conclusion and recommendations

Four days watering interval showed good growth with economical water use and this is essential during the period of seedling hardening. The results can be applied in the Kenyan highlands for *Warburgia ugandensis* and other succulent leaf plants. However more studies needs to be done for other species using different pot sizes and soil mixtures.

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Competing interest

The authors wish to declare no competing interest as pertains to the current paper

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