Effects of Planting Density on the Growth rate of *Euphorbia tirucalli* (L.) under Different Agro-ecological Zones in Tanzania

Hamisi. Y. Nchimbi
Department of Conservation Biology, University of Dodoma, P.O. Box 338 Dodoma, Tanzania
nchimbis9@gmail.com

ABSTRACT
The growth rate of *Euphorbia tirucalli L.* under different agro-ecological zones of Tanzania was determined using standard growth analysis procedures. It was presumed that, increase in planting density results in a significant increase in the growth rate of *E. tirucalli* under different agro-ecological zones. Assessment of the effects of planting density on growth rate of *Euphorbia tirucalli* was carried out through plant heights and above ground biomass at intervals of three months for 12 months. Results showed that the biomass and height growth rates increased significantly between 0.004g/g^{-1}day^{-1} and over 0.0006g/g^{-1}day^{-1} and 0.0040 cm.cm $^{-}$1.day^{-}1 and over 0.0100 cm.cm $^{-}$1.day $^{-}$1 respectively, for *E. tirucalli* cuttings planted in different agro-ecological zones. Also, the cuttings planted in the Dodoma agro-ecological zone had significantly higher biomass and height growth rates than those planted in the Mbeya and Dar es Salaam agro-ecological zones respectively. However, the biomass growth rates were not significant during the period between planting and the first harvest irrespective of the planting density or agro-ecological zone. Results further demonstrated that throughout the one year period for which the study plants were grown, their performance in biomass and height growth rates were highest at the closest planting densities (129,600 plants per hectare). The study concludes that, the increase in planting density results in a significant increase in the growth rate of *E. tirucalli*. It also recommends for Dodoma being a better zone for higher growth rates of the plants when compared with the Mbeya and Dar es Salaam.

Keywords *– Agro-ecological zone, Euphorbia tirucalli, growth rate, planting densities*

I. INTRODUCTION
Euphorbia tirucalli L. is a shrub or a tree native of America but has been naturalized and grows freely in tropical areas, all parts of India and East African countries including Tanzania, where it is used as a medicinal and energy plant (VanDamme, 1990; FARA, 2008; Kumar, 2009; Julius and Patrick, 2011b). *Euphorbia tirucalli* contains pencil-like branches from which it derives the name, the pencil-tree. *E. tirucalli* is generally evergreen and are rarely fed on by herbivores. When wounded, *E. tirucalli* produces white latex which contains substances having medicinal and energetic properties (Julius and Patrick, (2011a and b). Several studies have shown that *E. tirucalli* grows vigorously once established. According Orwa *et al.* (2009) *E. tirucalli* plants grown using cuttings of 3 inches (7.62cm) attained a height of 2 feet (60.96cm) in one year (Van Damme, 1999; Taylor, 2005; Loke *et al.,* 2011). Yet, it was recognized that *E. tirucalli* plants grown for periods of 6 – 7 months attained a height of about 4 feet (121.92cm) (Van Damme, 1999; Van Damme, 2001; Loke *et al.,* 2011). Further reports showed that under ideal conditions *E. tirucalli* cuttings can grow to a height of 72 cm in 6 months. Calvin, (1980) also noted that a 5 cm *E. tirucalli* cutting can attain more than 50 cm height in the first growing season. However, there is insufficient information on the effect of planting densities on the growth rates under which the reported heights of *E. tirucalli* were determined. Apart from the information on *E. tirucalli* height growth rate described above, the effect of planting density on *E. tirucalli* biomass production was also evaluated by various authors. Studies revealed that under a planting density of 14,000 plants /hectare, *E. tirucalli* can yield 22–25 tonnes of dry biomass per hectare (Van Damme, 2001; Kumar, 2009). Also *E. tirucalli* can yield 2.3 tonnes of dry biomass per hectare when grown at a density of 10,000 plants/hectare with 6 harvests. Again it was also reported *E. tirucalli* yields of 14 tonnes of dry biomass per hectare after one year when planted at a density of 10,000 - 20,000 plants per hectare (Saw *et al.*, 1989; Van Damme, 2001). Furthermore it was demonstrated *E. tirucalli* biomass can produce 20 dry metric tonnes per hectare per year at a density of 80,000 plants (Saw *et al.*, 1989). These results show that variations in biomass yields of *E. tirucalli* in diffrent areas reflect the effect of planting density. But, there was insufficiency of information on the effect of such reported planting densities on the growth rate of *E.*

tirucalli. Thus, assessing the effects of planting density on growth rates of *Euphorbia tirucalli (L.)* under different agro-ecological zones in Tanzania was crucial in order to obtain information that relate to the environment of Tanzania. Therefore, the present study was carried out in February to August 2012 to determine effects of planting density on the growth rate of *E. tirucalli* in Dar es Salaam, Dodoma and Mbeya agro-ecological zones of Tanzania.

2. MATERIALS AND METHODS

2.1 The study areas

The study was carried out in three different agro-ecological zones of Tanzania (Mohamed, 2016), *viz.,* Eastern zone: Dar es Salaam, Kinzudi village in Goba (Mbezi), Central zone: Dodoma, Ibihwa village in Bahi District and the Southern highlands zone which includes Mbeya, Iyela Ward (Mbeya City).

2.2. Assessment of the Growth Rate of *E. tirucalli* Cuttings under Different Planting Densities

E. tirucalli cuttings, each measuring 30cm and having a mean circumference of 5cm, were planted in plots measuring 10m x 10m each in a randomized complete block design (Plate 2.1; A, B and C) at the three study sites. Prior to planting the cuttings, each plot was subdivided into 16 sub-plots measuring 2.5 x 2.5m each.

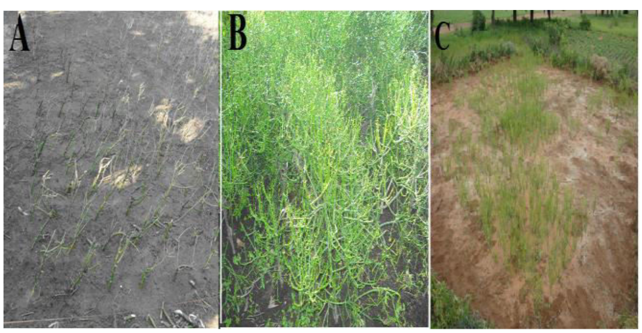

Plate 2.1: Planted *E. tirucalli* cuttings. A: 2.5m x 2.5m sub-plot one day after planting in Mbeya. B: 2.5m x 2.5m sub-plot six months after planting in Mbeya. C: 10m x 10m plot three months after planting in Dodoma showing canopies of different densities.

In order to determine the best planting density for optimum growth rate of the study plant, the cuttings were planted at different spacing of 0.25 x 0.25m, 0.5 x 0.5m, 1 x 1m and 2 x 2m making densities of 81, 25, 9 and 4 cuttings per sub-plot respectively. These densities per sub-plot were respectively equivalent to densities of 129,600, 40,000, 14,400 and 6,400 plants per hectare. Each planting density was replicated 4 times. The parameters used to assess the growth rate were plants above ground biomass and heights. Despite the fact that biomass in general, includes the above-ground and below-ground living mass (Lu, 2006) in this study only the above-ground biomass was considered for convenience. Determination of underground biomass can be cumbersome and time consuming and often yields inaccurate results; so in the present study it was not attempted due to limited time and monetary resources.

The measurements of biomass of the plants were done by randomly harvesting 25% of all *E .tirucalli* trees from the peripheral areas of the treatment plots cut at ground level in each sub-plot. Thus, 1, 2, 6 and 20 plants were harvested from each of the densities of 129,600, 40,000, 14,400 and 6,400 plants per hectare at each harvesting time, respectively. The harvests were carried out at intervals of three months for the duration of 12 months. At each harvest a 15-cm long stem bark strip was stripped from a stretch of each harvested stem, air dried for two days to prevent mold formation and then carefully placed in a labeled re-sealable type of plastic bag. All samples were then transferred to the Chemical and Mining Laboratory at the College of Engineering and Technology, University of Dar es Salaam. In the laboratory, all the harvested plant samples were oven-dried to a constant weight at 70°C and the mean dry weight of each plant was determined by dividing the total dry weight by all the plants making up the sample group from each sub-plot.

The mean plant dry weight thus obtained was used to determine the mean relative biomass growth rate (RBGR). The rate of growth of *E. tirucalli* was determined as Mean Relative Biomass Growth Rate (RBGR) using the formula derived by Fisher (1921) and modified by Lewis and Greene (1970), Kvet *et al* (1971) and Hunt (1982; 1990) i.e.

$$\textit{Mean Relative Biomass Growth rate (RBGR)} = \frac{1}{t_2 - t_1}\int_{t_1}^{t_2} \frac{1}{W} * \frac{dW}{dt} * dt$$

$$= \frac{1}{t_2 - t_1}\int_{w_1}^{w_2} \frac{dW}{W} * dt$$

$$= \frac{LnW_2 - LnW_1}{t_2 - t_1}\int_{w_1}^{w_2} g.g^{-1} * day^{-1} \ldots\ldots\ldots\ldots\ldots (2.2)$$

The Growth Rate of *E. tirucalli* was also determined as Relative Increase in Height per day - an index of growth of the cuttings, using the formula proposed by Grotkopp and Rejmanek *et al.,* (2007). i.e,

$$Mean\ Relative\ Height\ Growth\ rate\ (RHGR) = \frac{Log_e L_2 - Log_e L_1}{T_2 - T_1}\ cm.cm^{-1} * day^{-1} \ldots\ldots (2.2)$$

2.3 STATISTICAL ANALYSES

Differences in the relative growth rate of *E. tirucalli* at different planting densities were determined using One-way analysis of variance (ANOVA) using SPSS v.15 with post-hoc tests.

3. RESULTS AND DISCUSSION

Results on effects of planting densities on the growth rates of *E. tirucalli* under different agro-ecological zones of Tanzania showed that mean plant weight (g) and height (cm) as well as the relative growth rate in weight ($g.g^{-1}day^{-1}$) and height (cm/day) are presented in Tables 3.1 - 3.4 respectively (Appendix A). Both mean plant dry weight (g) and height (cm) showed increasing trend with increases in harvesting time or days of harvest from the first harvest (day 90) to the fourth harvest (day 360) across different planting densities in all agro-ecological zones. The plant weight and height across different planting densities increased from the lowest to the highest planting densities in all sub-plots established in different agro-ecological zones of Dodoma, Mbeya and Dare es Salaam (Figures 3.1-3.6; Appendix E). Also, the results showed that at any one harvest time the weight and height values of *E. tirucallli* plants grown in the Dodoma site were significantly higher than those of the plants grown in the Mbeya and Dar es Salaam sites (see Figures 3.1-3.6).

Results of the relative biomass growth rates (RBGR) (Figure 3.7-3.9; Appendix C) and relative height growth rates (RHGR) (Figure 3.10-3.12; Appendix D) suggest that, the density of planting or the spacing between the cuttings is an important factor influencing the growth *E. tirucalli.* The relative biomass growth rate and the relative height growth rate increased with increases in planting density in different agro-ecological zones. An increase in the relative biomass growth rate was observed between the first (at day 90) and second harvests (at day 180) thereby indicating the rapid early establishment of *E. tirucalli* cuttings and a high accumulation of biomass in relation to the pre-existing biomass. Thereafter the relative growth rate increased at a relatively reduced rate towards the third (day 270) and fourth (day 360) harvests. The declining trend between the first and the last harvests at all spacing indicated that in all cases the growth increments relative to the pre-existing plant heights were rather small. Also, the results showed that the *E. tirucalli* cuttings planted in the Dodoma agro-ecological zone had significantly higher growth rates in terms of both biomass and height than the cuttings planted in the Mbeya and Dar es Salaam agro-ecological zones in that order (see Figures 3.7 - 3.12; Appendix D). In view of all cases, the results show that throughout the one year period for which the plants were grown, the performance of *E. tirucalli* in terms of both relative biomass growth rate and relative height growth rate was highest at the closest planting densities.
Furthermore, the results of the statistical analysis of variance (One-way ANOVA) for the relative growth rate of *Euphorbia tirucalli* showed that the differences in the relative growth rate in terms of both biomass and height with planting density and agro-ecological zones were significantly different ($p < 0.05$) respectively (see respective Tables 3.50 and 3.60; Appendix B), except for differences in the relative biomass growth rates which were not statistically significant different during the period between planting and the first harvest irrespective of the planting density or agro-ecological zone in which the planting had been done.

Generally, the findings of the present study showed that, *E. tirucalli* growth interms of height and biomass at all planting densities in the different agro-ecological zones increased significantly. With regard to plant height, the Relative Growth Rate (RGR) was found to increase continuously but at a decreasing rate from the first harvest (90 days after planting) to the fourth harvest (360 days after planting). This shows that the proportions of height increments relative to the denominator heights were small as plant height increased. The height increase was due to the fact that, as *E. tirucalli* plants grew older there was a reduction in the light intensity captured by the plants because of increased interplant shading (canopy cover). Insufficient light reduced the rate of photosynthesis which decreased with increases in time after the first harvest in all sub-plots under different agro-ecological zones. This created increased interplant competition for light and space that caused plants to grow taller (development of long and thin stems) in search for light and so their net heights increased with time. The increase in plant's height under increased shading was also reported by earlier studies on *Lilium longiflorum* and Easter lilies (Kohl and Nelson, 1963; Heins *et al.,* 1982).

Likewise, the Relative biomass Growth Rates increased between the first harvest (90 days after planting) and the second harvest (180 days after planting) and, thereafter declined (increased growth at a decreasing rate) during subsequent harvests. The high Relative Biomass Growth Rate during the early stages of growth (first three months) implies that the amount of biomass increment relative to the existing biomass was high during those early stages of growth which accelerated photosynthetic activity. Also, there was less shading and competition between plants for light and other resources. A declining trend in the relative biomass growth rate from the second harvest all the way to the final harvests implies that the amount of *E. tirucalli* biomass increase relative to the biomass existing at previous harvests was low. The decline in RBGR was also because during the period between planting and the first harvest the sprouts were initially small and yet to develop the profusely branching shoot system which was later responsible for causing the shading effect that reduced photosynthetic activity with significant reduction in the amount of gross primary production that coupled with the effects of respiration, inevitably resulted in reduced net primary production as the plants grew older.

According to South (1995) *E. tirucalli* is among the trees in which growth occurs very rapidly during the first few months after establishment of the cuttings. It grows according to the variable interest law, which states that the amount of growth made by a plant in a unit of time is a percentage of the size of the plant at the beginning of the period and this percentage changes as the plant increases in size often the percentage declines as plant size increases. Thus, the growth pattern exhibited by *E. tirucalli* concurs with the postulates of Medawar, (1941) and Causton, (1983) who reported that, most trees during their first year of growth show an ontogenetic drift in Mean Relative Growth Rate (RGR) as their size increases meaning that, many organisms exhibit a declining Mean Relative Growth Rate (RGR) over time. Furthermore, Britt *et al.,* (1991) and van den Driessche, (1992) concluded that, smaller plants had higher RGR than larger ones. Also, van den Driessche, (1992) and Britt *et al*. (1991) pointed out that young plants often grow exponentially over a period of several years and then decline, and consequently the Relative Growth Rate decreases with time.

Despite the overall increase in growth in terms of biomass and height shown by *E. tirucalli* plants grown at all densities, the closer spacing showed the highest increase in the Relative Growth Rate while the wider spacing showed relatively lower growth rates. This probably indicates that, there was higher nutrients uptake in higher plant densities than lower plant densities. Greater uptake of these nutrients at higher plant densities was probably due to higher biomass production produced at higher plant densities as compared to lower plant densities. Higher biomass production generally result in high biological activity in the soil and on the soil surface which permit an effective uptake of nutrients by plant roots and activate interaction of organic matter with microorganisms. This increases nutrients transfer efficiency between soil and the plant, resulting in almost perfect physical and hydric conditions for plant growth. Furthermore, the closer spacing (i.e., 129,600 plants per hectare) *E. tirucalli* plants grown in all the agro-ecological zones attained canopy closure which was essential to cause higher solar radiation interception and higher growth rates. This again was probably a response of the plants to increased shading by neighboring plants at the closer spacing which forced the individual plants to branch profusely so as to maximize the surface area for light interception. According to Loke *et al,* (2011), at the age of 6 - 12 months or more the petroleum plants (*E. tirucalli* plants) require lots of light and will generate too much shade. The growth rate results showed similar trends at all spacing under different agro-ecological zones. Thus, it is probable that, at the closer spacing competition for light would have been very severe if the *E. tirucalli* plants had been left to grow for a period longer than one year.

The results of the present study showed that both Relative Growth Rate (RGR) in terms of height (RHGR) and Biomass (RBGR) increased with increasing planting densities. This was probably because, there was higher nutrients uptake in higher plant densities than lower plant densities which was due to higher biomass production produced at higher plant densities as compared to lower plant densities. This results to high biological activity in the soil and on the soil surface which permit an effective uptake of nutrients by plant roots. The rapid increase in the Relative Height Growth Rate recorded in the present study for *E. tirucalli* plants grown at high densities are similar to those obtained by Amaglo *et al.* (2006) who observed that increasing plant density accelerates the rate of plant growth hence the increased heights at closer spacing in *Moringa oleifera*. Also, the increase in the biomass of *E. tirucalli* with increasing planting density obtained in this study is similar to the results reported by Norman (1992) and Foidl *et al*. (2001) which showed that total biomass production per unit area increases with increased planting density in tropical crops such as *Moringa oleifera*. According to Killi and Ozdemir (2001), increasing plant density results in an increase in the biological yield of plants due to the increase in the amount of photosynthetically active radiation which is intercepted by the crop.

Since, findings of the present study suggest that as the plant population per unit area increases, the total biomass and height increments also increase. However, a point is reached where plants begin to compete for essential growth factors like sunlight interception, nutrients and water uptake (Sangakkara *et al*., 2004). The effect of increasing competition is to decrease the concentration of growth factors available to each competing plant individual. But the increase in plant density does not affect ecological performance of individual plants while the plant density stays below the threshold but when this threshold is exceeded, competition between plants ensues. The fact that in the present study the growth rate of *E. tirucalli* plants kept on increasing with increasing plant density implies that the threshold for plant competition that would slow down the growth rate considerably had not been reached at the density levels studied. Findings of this study show that, the closer spacing i.e., 129,600 plants per hectare, in all the agro-ecological zones facilitated early canopy closure and might be considered as the optimum planting density for achieving high growth rate for a one year growth period considered in the present study. Early canopy closure being the determinant of optimum planting density was reported by Hossain and Rahman, (2011) in soybean varieties who concluded that, plant population that facilitates canopy closure at early reproductive stages could be considered optimum for achieving higher growth rates. Therefore, the results of the present study confirm that "planting density has a significant effect on *E. tirucalli* growth rate under different agro-ecological zones". Results of the present study show that, *E. tirucalli* growth rates were highest in the Dodoma agro-ecological zone intermediate in the Mbeya agro-ecological zone and lowest in the Dar es Salaam agro-ecological zone. This was probably because, there was higher nutrients uptake by *E. tirucalli* plants grown in Dodoma agro-ecological zone than Mbeya (intermediate) and Dar es Salaam (lowest) agro-ecological zones due to higher biomass production. Also, it is because *E. tirucalli* has been described as a hard plant which can survive under a variety of climatic regimes ranging from semi-arid to mesic climatic conditions (Calvin, 1980; Duke, 1983; FAO, 2012). This ability of *E. tirucalli* to survive in a variety of climatic conditions and particularly in the semi-arid conditions is due to its succulent nature which enables it to reserve water in its tissues for use during drought periods. As such it exists more or less independent of water supply from the soil during the peak of the dry season and its physiological activities proceeds as normal. Furthermore, the use of phylloclades instead of leaves for photosynthesis gives an extra advantage to *E. tirucalli* to be able to combine both CO_2-fixation in the leaves with Crassulacean Acid Metabolism (CAM) in the stem and phylloclades. The phylloclades and stem open their stomata and absorb carbon dioxide at night when it is cool thereby minimizing water loss through the stomata and increasing water use efficiency. According to Van Damme, (1989; 2001) this mechanism offers an additional ability of *E. tirucalli* plants to increase their metabolic rates and thereby maximize their growth rates and yields. The small leaves of *E. tirucalli* are preferentially used for photosynthesis in normal situations during the rainy season whereas the green stem takes over when the leaves fall off in arid conditions and a combination of both pathways increases the plant's water use efficiency since the small leaves have a high affinity for carbon dioxide but tend to use water less efficiently (Van Damme, 2001). Herrera (2009), Kluge *et al*. (2001) and Van Damme (1989 and 2001) reported that the extent of succulence has been positively correlated to both colonization of increasingly arid habitats and an increased contribution of CAM activity to total carbon gain. Von Willert *et al*. (1990) argued that, succulence is a survival strategy for plants inhabiting arid and semi-arid areas.

4. CONCLUSION

From the findings of the study, the following conclusions were made.
The growth rates of *E. tirucalli* were found to increase with increasing planting density and it appeared that, the increase in planting density did not affect the ecological performance of individual *E. tirucalli* plants for the whole period of the growth experiment. This probably indicates that the total time (12 months) used to carry out growth experiment did not allow plants to go above the level at which competition would have affected them. However, at the highest planting densities (i.e., 129,600 plants per hectare) it was observed that the rate of *E. tirucalli* branching was very high in all agro-ecological zones in this study. This illustrates that, for the higher density plots (129,600 plants per hectare) competition might have been so severe if the plants would have been left to grow for a period longer than a year which might have caused enormous competition for light thereby lowering the plant's productivity. Hence, the threshold density at which intraspecific competition would limit the growth rate of the species was probably not reached.

Nevertheless, the closer spacing i.e., 129,600 plants per hectare, in all the agro-ecological zones facilitated early canopy closure and might be considered as the optimum planting density for achieving high growth rate for a one year growth period considered in the present study. Findings of the present study conclude that, the increase in planting density results in a significant increase in the growth rate of *E. tirucalli* under different agro-ecological zones. Also, for further growth experiments to be carried out using *E. tirucalli* trees, considering that the study plant can grow and survive in any soil type, Dodoma agro-ecological zone is the most favourable area for the growth of *E. tirucalli* followed by Mbeya and the least was the Dar es Salaam agro-ecological zone.

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