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EVALUATION OF MINERAL NUTRITION AND GROWTH OF *Eucalyptus Saligna* SEEDLINGS RAISED ON ORGANIC – ENRICHED NURSERY POTTING MEDIA

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ABSTRACT

Forest soil is the convectional growth media used in raising seedlings in tree nurseries in Kenya. Poor growth media in tree nursery establishment has been reported as the major cause of poor seedling establishment in the nursery and after out planting. This study evaluated the potential of forest soil and organic mixtures as potting media for seedling establishment. The experiment was laid out in a completely randomized design with five treatments comprising of: Forest soil (Medium 1); Sawdust mixed with cattle manure in the ratio of 1:1 by weight (Medium 2); Forest soil mixed with cattle manure - sawdust mixture (1:1) in the ratio of 1:1 (Medium 3); Forest soil mixed with cattle manure - sawdust (1:1) mixture in the ratio of 1:2 (Medium 4); and Forest soil mixed with cattle manure - sawdust (1:1) mixture in ratio of 1:3 (Medium 5). The treatments were replicated three times in experimental plots of 100 seedlings per treatment. Seedlings were allowed to grow for 24 weeks while the growth parameters, foliar nutrient concentration and seedling mortality were monitored periodically during the study period. Enrichment of Forest soil with Sawdust and Cattle manure at various ratios increased nutrient concentrations in the media and enhanced growth and dry matter biomass compared to those with forest soil alone thus resulting to faster growth and increased nutrient uptake. There is need to develop strategies for mixing organic materials with forest soil which will improve decomposition and growth of seedlings and thereby reduce mining of forest soil.

Key words: *Cattle manure, Forest soil, Sawdust, Tree seedlings, Nutrient uptake*

INTRODUCTION

Large volumes of soils are extracted from natural forests for raising seedlings in tree and vegetable nurseries in Kenya. Soils collected from forest floor (top most layer about 15cm) have been reported to be deficient in essential plant nutrients such as nitrogen and phosphorus (Vitousek and Sanford 1986). Nutrients in forest soils have been found to decline due to the combined effects of pollution, agricultural activities, forest fires and tree harvesting (Binkley and Fisher, 2012). Due to this decline, seedlings raised in such soils suffer from poor growth and survival in the nursery and after out planting in field. Evans, (1983), recommends addition of small quantities of essential macro elements to supplement nutrient deficiency in the soils during preparation of potting media.

Tree nursery growing media can have sub-optimal nutrient concentrations leading to poor seedling growth, survival and mortality (Mbora, *et al*., 2008, Wightman, 1999). Currently, there is no standard tree seedlings nursery potting media in use in most extension and private tree nurseries in Kenya. Studies by Kungu *et al*., (2008) revealed growing media contribute to physical and chemical conditions that may be inappropriate for quality seedling development hence lack of standard nursery media by small scale farmer nurseries has

resulted to poor establishment of agroforestry practices on farms and in forestry systems. Several studies focusing on enhancing nutrient content of nursery soils and to reduce degradation of the forests have advocated several recommendations. For instance, studies by Kungu *et al.* (2008), in central Kenya showed that compost-based nursery media gave the highest seed germination percentage of 61.7% as compared to farm soil which gave the lowest seed germination percentage of 47.2%. In addition, compost based growing media gave the highest seedlings height and diameter and was therefore recommended as an alternative mixture when developing nursery media. Studies done by Depardieu *et al*., (2016), have shown that sawdust, despite being an inert media, when mixed with other substrate, can form a soilless growing system for producing bare-root seedlings, due to its ability to retain nutrients. Baiyeri, (2003) recommended use of poultry manure in nursery soil mixtures. Mixing forest soil with organic matter has also been shown to enhance water retention in media leading to vigorous growth of seedlings (Oades, 1988); plants had better growth and nutrition while phosphorus uptake was notably enhanced (Ostos *et al*., 2008). Saw dust has largely been considered as a waste product in most forestry system (Onchieku *et al*., 2013). Though it does not harbor microorganisms due to its low nutrient content (Okalebo *et al.*, (2002), it can provide nutrients over a long period if decomposition is managed appropriately by mixing with nutrient rich medium. Cattle manure is an ideal component for mixing with sawdust to enhance its decomposition and supply the immediate and long term plant nutrient requirements. The high concentration of macro nutrients in cattle manure can provide initial nutrients required for decomposition of sawdust and for growth of seedlings (Tanimu *et al*., 2013). Since chemical, physical and composition nature of nursery media regulate seedling growth, and affect rates of organic matter decomposition, it ultimately influences survival of the seedlings after out planting and productivity. There exists minimal data on alternative nursery potting medium of tree seedlings other than the convectional growth media comprising of forest soil. There exists minimal data on alternative nursery potting medium of tree seedlings other than the convectional growth media comprising of forest soil. This study sets out to evaluate alternative nursery media and comparatively assess the growth and performance of *Eucalyptus saligna* seedlings established using forest soil and organic mixtures of sawdust and cattle manure as potting media before transplanting in the field.

MATERIALS AND METHODS

Study Area

The study was carried out in South Kinangop Forest in Nyandarua County, Kenya. The forest is part of the Aberdare Forest Reserve and is located at an of latitude S 00^0 43' 28.4", longitude E 036^0 40' 51.7" and elevation of 2545m above sea level. The area receives annual rainfall of about 1590mm distributed bi-modally with peaks in late April and late November and mean annual temperature of 12.3^0C (Jaetzold *et al*., 2006). The forest consists of both natural and planted forest blocks occupying a total area of 6,660 hectares. The common forest plantation species include *Cupressus lustanica, Pinus patula and Eucalyptus saligna*. In the natural forest, common species include; *Juniperus procera Macaranga kilimandscharica, Hagenia abyssinica, Olea Africana, Dombeya torrida, Polyscias kikuyuensis, Prunus Africana, Nuxia congesta, Ekerbagia reuperiana* and *Podocarpus latifolia* The forest blocks are established with seedlings mainly raised in local tree nurseries at the forest station and managed in the field by the community. The soils are deep red, strongly weathered and acidic. The nutrient content of the soils is considered moderate for nitrogen (N), potassium (K) and calcium (Ca), while low in phosphates (P) (Jaetzold *et al*., 2006).

Experimental Layout and Data Collection

Preparation of nursery potting media

The potting media were prepared from mixtures of Forest soil (conventional mixture), and organic materials: Sawdust and Cattle Manure. The five mixtures of soil and organic materials used are described in detail in Table 1. Forest soil was collected from the surface layer of 0-15cm depth from the natural forest stands of *Dombeya torrida* during the dry season of 2015. Five stands of *D. torrida* of approximately $4M^2$ were identified and the top soil dug up and bulked in gunny bags. The soil was mixed thoroughly, air dried under shade and sieved to pass through 8mm sieve to remove

large plant materials and stones, then weighed for media preparation.

Fine sawdust was collected from nearby timber processing sawmills and mainly from *Pinus patula* trees and sorted to remove wood debris. Cattle manure was collected from homesteads near the forest boundary, mixed together to obtain a homogenized bulk sample. The sawdust, forest soil and cattle manure were then weighed according to treatment mixtures based on weight ratios and heaped separately. The heaps of treatment mixtures were covered with polythene sheet and allowed to decompose for three months under shade. After three months, 300g each of the media was weighed into polythene bags measuring 3''by 4''and planted with *Eucalyptus saligna* seedlings pricked from germination seedbed.

Table 1: Treatment description showing the growth media mixtures and rations used in the study

Treatment code	Treatment description	Mixture ratios		
		Forest soil	Sawdust	Cattle manure
Medium 1	Forest soil only	1	0	0
Medium 2	Sawdust mixed with cattle manure in the ratio of 1:1 by weight	1	1	0
Medium 3	Forest soil to cattle manure - sawdust mixture (1:1) in the ratio of 1:1 by weight.	1	0.5	0.5
Medium 4	Forest soil to cattle manure - sawdust (1:1) mixture in the ratio of 1:2 by weight	1	1	1
Medium 5	Forest soil to cattle manure - sawdust (1:1) mixture in ratio of 1:3 by weight	1	1.5	1.5

Tree seedling establishment and management

For seedling germination, fifty (50grams) of *Eucalyptus saligna* seeds were acquired from Kenya Forest Research Institute (KEFRI), Muguga Seed Centre. The seeds were sown in a seed bed of sand, two weeks before the completion of decomposition period of the nursery growth media. After germination the seedlings were allowed to grow for 14 days and on the 15thday after germination, they were pricked out, into polythene tubes. Three hundred (300) polythene tubes of size 3''by 4'' were filled with the growth media (300g) for each of the treatment described in Table 1 and arranged in an experimental unit of 100 seedlings per replicate. The layout comprised of seedlings established in a completely randomized design with three (3) replications. Seedlings were allowed to grow for 24 weeks under normal nursery management operations. Watering was done twice daily during the dry period or once daily. Weed removal was done manually. Seedlings growth parameters including height and root collar diameter, biomass accumulation and mortality were measured at four-week interval.

Measurement of height, root collar diameter, biomass and mortality

Seedling shoot height, root collar diameter, biomass and mortality were measured at an interval of 4 weeks for a period of 24 weeks (to nearest 1 mm). A total of 75 randomly sampled seedlings treatment (5 from each replicate) was sampled during each sampling period, whereby 15 seedlings were selected randomly from each treatment for destructive sampling. The seedlings height (cm) and root collar diameter (mm) were measured and then the seedlings above ground biomass cut and oven dried at 70°C for 24 hours for biomass (g) determination and nutrient concentration in the tissue. Seedling mortality was determined by the total number of dead or weathered seedlings within each sampling period.

Nutrient analysis of the nursery media

For determination of nutrient content in the potting media, 500g sample of each of the potting media treatments was taken for qualitative analysis. The media were subdivided into three portions, air-dried, sieved through a 2 mm sieve and oven dried at 80^0C before element analysis at Soil Testing and Plant Analysis Laboratory – Kenya Agricultural Research Organization (KARLO Muguga). pH was determined in water (Anderson and Ingram 1993), while total N and C were determined using a Carlo Erba CNHS analyzer. Available P was determined calorimetrically after extraction with Melich' s reagent (l0g soil in 100 ml 0.025 N H_2SO_4/0.05 N. HCL, exchangeable cations K, Mg and Ca were determined after extraction by shaking for 2h with 1 M ammonium acetate (5 g in 100 ml) (Anderson and Ingram 1993). Available K was determined by flame photometry, while Mg and Ca were determined by atomic absorption spectrophotometry.

Nutrient analysis in Plant tissue

Dried leaves and stems from the 8-week and 24 week harvests were ground in a Wiley mill and analyzed for concentrations of N, P, K, Ca, Mg. Nitrogen was determined by Auto analyzer, and for other elements P, K, Ca, and Mg plant material was wet-ashed with concentrated H_2O_2/H_2SO_4 solution and nutrients were determined as for the potting media.

Nutrient uptake was estimated as: -

*Uptake from seedling biomass = nutrient content in foliar * Biomass yield (g/pot)/100*

Statistical Analysis

All data collected were subjected to statistical analysis and analysis of variance using SPSS, Version 20 statistical package. The nutrient uptake of plant tissue was calculated from nutrient concentrations and corresponding tissue dry weights for each treatment. Since the experiment was laid out in complete randomized design, variations over treatment were studied using one-way ANOVA model with average growth parameters as the dependent variable. Treatments were considered as the fixed effect and within treatment variation as the error term. Critical value of $P>0.05$ was used to determine statistical significance.

RESULTS

Fertility of the Potting Media

The mixture of forest soil, cattle manure and sawdust had the highest amount of total nitrogen content of 5.6g/kg, while the lowest amount of total nitrogen of 5.3g/kg was recorded in Medium 2 (Table 2). Percentage total carbon ranged between 17.82 g/kg to 36.8g/kg in Medium 3 and Medium 2 respectively. PH (in water) of the media was acidic (pH 4.93) in Medium 1 while in Medium 5, the media was a weak base (pH8.05).

Table 2: Characterization of the nursery potting media at the start and the end of the nursery experiment

At start of experiment (Baseline)

Treatment	pH (water)	Org C (g/kg)	Nitrogen g/kg	Phosphorus g/kg	Exch. K ppm	Exch. Ca ppm
Medium 1	4.93	18.09	5.5	0.26	0.97	1.93
Medium 2	6.04	36.80	5.3	0.90	*7.30*	4.64
Medium 3	6.30	17.82	5.6	0.58	5.22	2.66
Medium 4	6.75	20.20	5.4	0.36	5.15	2.81
Medium 5	8.05	18.15	5.4	0.87	7.08	3.21
LSD (P<0.05)	*	*ns*	*ns*	*Ns*	*Ns*	*Ns*

At the end of the experiment (24 weeks)

Treatment	pH (water)	Org C (g/kg)	Nitrogen g/kg	Phosphorus g/kg	Exch. K ppm	Exch. Ca ppm
Medium 1	5.16	10.39	7.3	0.16	0.18	1.84
Medium 2	6.07	9.83	8.5	0.26	1.15	3.24
Medium 3	6.30	12.36	8.5	0.38	1.02	3.76
Medium 4	6.48	13.20	8.3	0.51	0.99	4.50
Medium 5	6.56	25.00	10.8	0.93	0.69	5.82
LSD (P<0.05)	*	*ns*	*ns*	*Ns*	*Ns*	*Ns*

* means significant at P<0.05; *ns* means not significant *(Modified from Ashiono et al., 2017).*

Carbon to nitrogen ratios of the potting media was high (range 24:1 - 37.4:1) at the beginning of the experiment and declined during the experimental period (Figure 1). At week 24, the five potting media under consideration had C: N ratios of between 8:1 to 15.9:1 with Medium 5 and Medium 2 recording the lowest C: N of 8:1 and 11.5:1 respectively. A carbon to nitrogen ratio of 20:1 is considered optimum for decomposition of organic residues (Zhu, 2007) and therefore in this study decomposition was at the maximum from week 16 when C: ratio was below 20: 1 for all treatments.

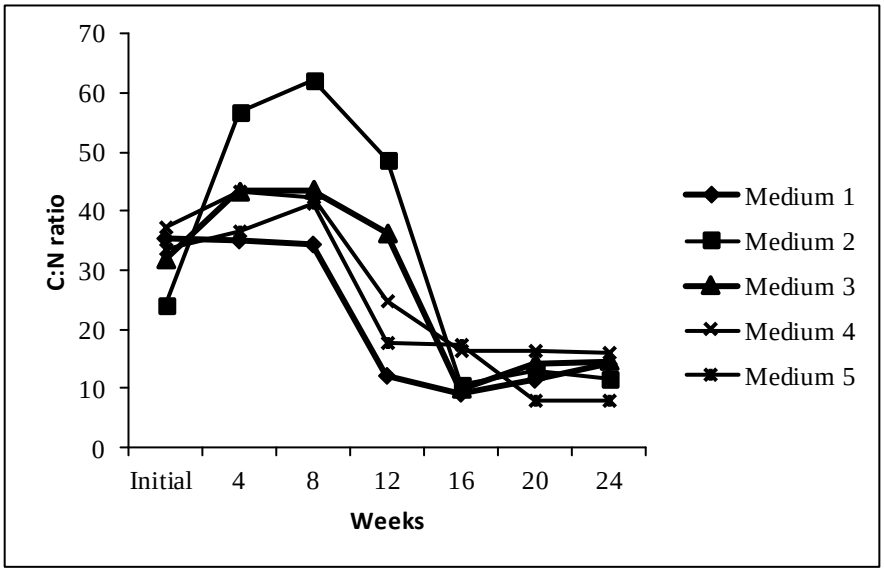

Figure 1: Carbon: Nitrogen ratio of the potting media during the seedling growing period

Determination of Seedling Growth Parameters

Height of seedlings grown in different potting media during the growing period is shown in Table 3. Shoot height of the seedlings at the end of the growing period was highest in Medium 2 followed by Medium 3 and Medium 1 with height values of 13.15cm, 11.78cm, and 11.34cm respectively. All seedlings at week 24 had reached transplantable height of above 10cm which is recommended by Lamprecht, (1989). Growth increased steadily reaching a diameter above 5.0 mm in all treatments at week 24 as shown in Table 3. Medium 5 which comprised of Forest soil mixed with cattle manure - sawdust (1:1) mixture in ratio of 1:3 had the fastest growth as compared to other treatments recording a root collar diameter of 9.55mm at week 24.

Seedling growth and biomass was enhanced by mixing forest soils with sawdust and cattle manure. The highest biomass yield at the end of the experimental period was recorded in Medium 5 and Medium 2 with 19.1g and 18.88g dry biomass respectively. However, these differences were not statistically different among the potting media treatments at 5% level with respect to biomass, shoot height and root collar diameter.

Table 3: Effect of different potting media on biomass, height and root collar diameter of seedlings

Treatment	Height (cm)		Root collar diameter (mm)		Dry Biomass (g)	
	4 weeks	24 weeks	4 weeks	24 weeks	4 weeks	24 weeks
Medium 1	1.43	11.34	0.2	5.29	0.59	17.77
Medium 2	0.25	13.15	0.22	6.15	0.57	18.89
Medium 3	0.32	11.78	0.21	7.60	0.12	16.87
Medium 4	0.32	10.86	0.22	7.67	0.08	16.68
Medium 5	0.24	10.03	0.21	9.55	0.22	19.10
LSD (P<0.05)	*ns*	*ns*	*ns*	*Ns*	*Ns*	*Ns*

Seedling mortality was determined by counting the total number of dead or weathered seedlings during the growing period. On the average, death of seedlings in the treatments was minimal during the study period. Mortality of seedlings was highest in Medium 2 (6.67%) followed by Medium 4 (5.67%) and the lowest in Medium 1 (2%) as shown in Table 4.

Table 4: Cumulative seedling mortality in different growth media during the growing period (at 24 weeks)

Treatment	Initial count per treatment	Number of dead seedlings at week 24	Percentage mortality (%)
Medium 1	300	6	2.0
Medium 2	300	20	6.67
Medium 3	300	15	5.0
Medium 4	300	17	5.67
Medium 5	300	13	4.33

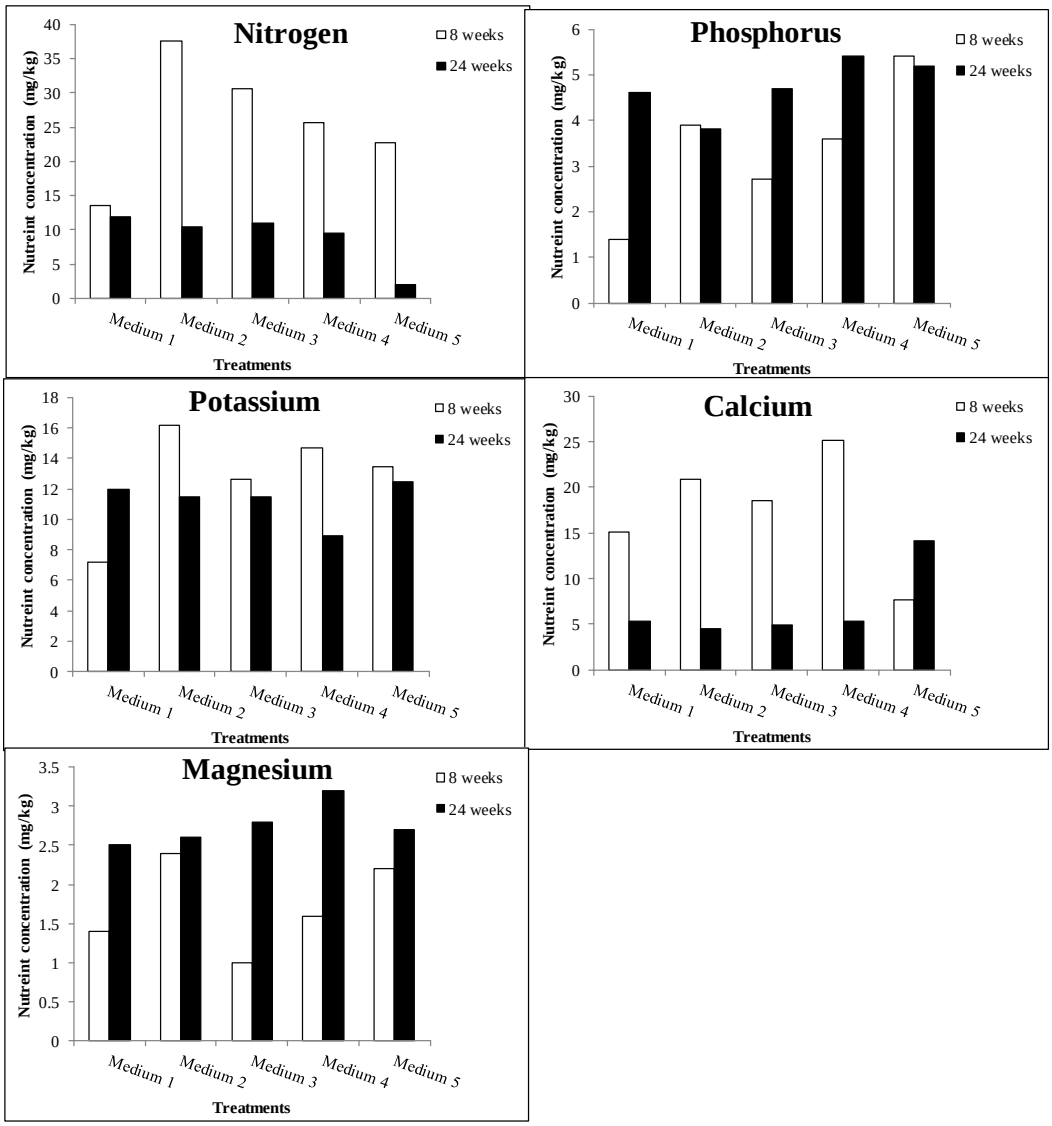

Figure 2. Foliar nutrient concentrations in *Eucalyptus saligna* seedlings grown in enriched forest soil as potting media

Foliar Nutrients analysis

Foliar nutrient concentrations varied with treatments in the potting media (Figure 2). Foliar nitrogen was significantly reduced in seedlings grown in Medium 1 (Forest soil alone) at week 24 compared with other potting media which were enriched with cattle manure and sawdust. Nitrogen ranged between 13.5 – 37.7 g/kg for Medium 1 and Medium 2 respectively in week 8, and declined to 2 – 12mg/kg in Medium 1 and Medium 5 respectively. Foliar phosphorus was however highest in the 24 week (3.8 – 5.4mg/kg). The level of Ca concentration was high in week 8 (range 7.7 - 25.2 mg/kg) and declined appreciably in week 24 (4.5 – 14.2 mg/kg) while Mg ranged between 1.0 - 2.4 mg/kg in week 8 and 2.5 – 3.8 mg/kg in week 24. Nutrient concentrations of all the above-mentioned elements were low in seedlings established in forest soil alone.

Effect of Enrichment of Potting Media on Seedling Nutrient Uptake

Nutrient uptake in the foliage of *Eucalyptus saligna* seedlings is shown in Figure 3. Plant tissue analysis varied with the potting media whereby N uptake in the foliar increased steadily during the growing period and was highest in week 20 ranging between 5.49 to 10.79mg/kg. Phosphorus uptake in the seedling biomass was highest in Medium 4 and lowest in Medium 2 while Potassium uptake was

highest in Medium 5. Low levels of calcium uptake were recorded in all treatments and showed a trend similar to nitrogen during the growing period of the seedlings.

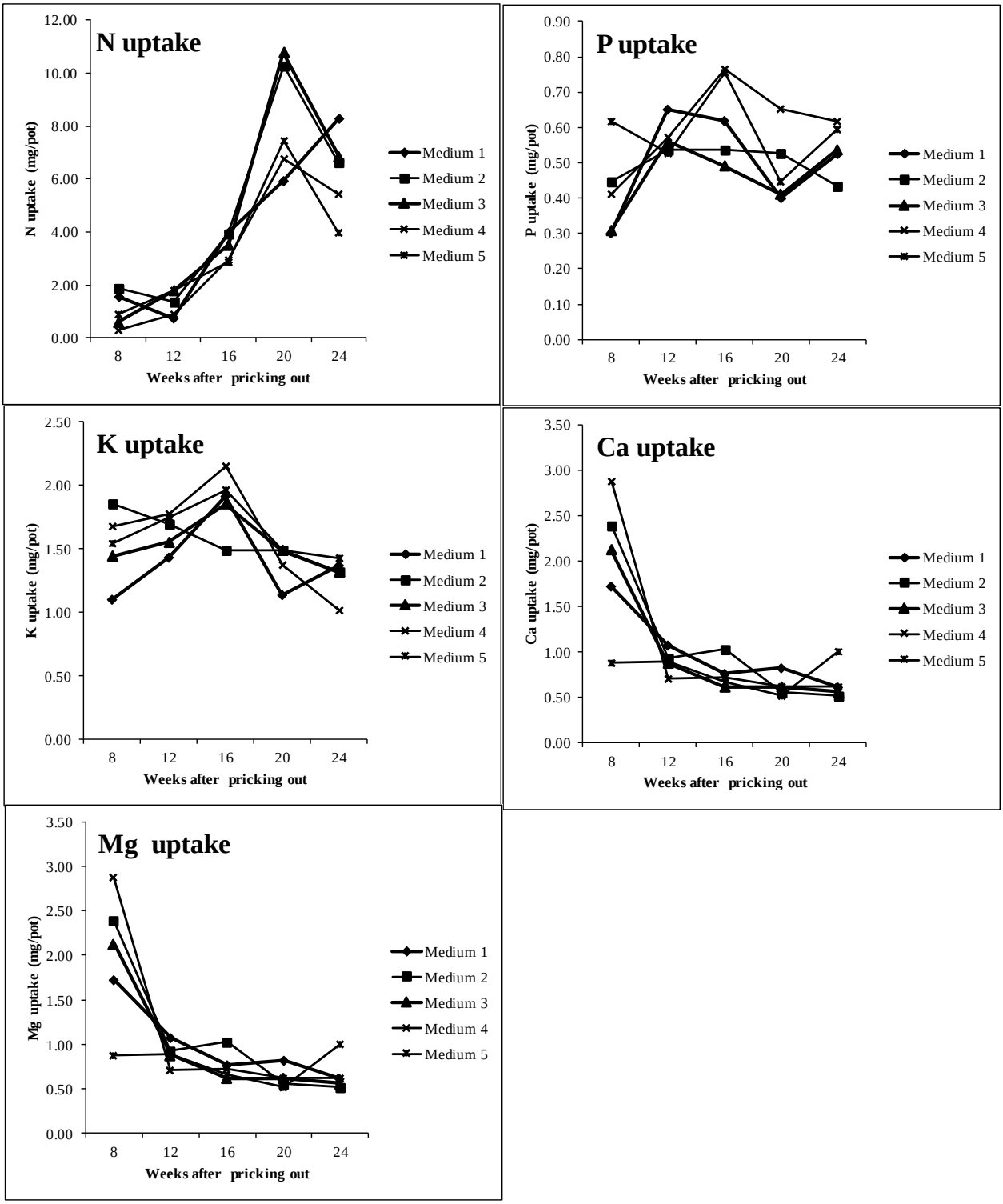

Figure 3: Nutrient uptake by *Eucalyptus saligna* seedlings grown in enriched potting media

DISCUSSION

Nutrient Concentration of the Potting Media and its Effect on Seedling Growth

The results of analysis of the potting media prior to and after the experimental period showed that the content of percentage organic carbon and nitrogen were quite moderate and above the critical level of 0.2% as reported by Okalebo *et al*., (2002), and Landon, (2014). There was a general increase in the contents of total nitrogen in potting media during the study period. Treatments with cattle manure in the mixture had high levels of nitrogen compared to those with forest soil alone. However, the available P content was lower than the critical levels considered optimum by Okalebo *et al*., (2002).

The low levels of P and Mg reported in this study are similar to the findings of Singh and Bhati (2003) on Eucalyptus seedlings when irrigated with municipal effluent in Indian deserts. This is attributed in part to P fixation which is common in tropical soils and partly due to the partial sterilization of soil during preparation of the nursery media which increased microbial activities thereby favoring increased microbial degradation of soil organic matter for more release of nutrients (Fabião, *et al*., 2002).

The slow effect of N at the start of the experiment is mainly found with recalcitrant organic matter with a high C: N ratio as was the case with Medium 4 which had forest soil mixed with sawdust and cattle manure in the ratio of 1:2 by weight with a high C: N ratio of 37:1. This could be attributed to immobilization, which causes loss of N from the system and a reduction in the soil ability to mineralize N (Mendham *et al*., 2003).

Organic residue impact on soil fertility will depend not only on the quality (Parton *et al*, 1994) and quantity of the residues (Tian *et al*., 1992) and the prevailing climatic conditions (Vanlauwe *et al*., 1995) but also on the way they are managed from different sources (Adetunji, 1996, Cadisch and Giller, 1997). The need for proper residue use and management have been stressed by many researchers and therefore if forest establishment is to be improved by expanding the nursery potting media resources, there is need to consider handling of the materials and development of modules to establish the right mixtures which will enhance decomposition and eventual growth of the seedlings.

Nutrient Uptake of Seedlings

Incorporation of highly lignified sawdust have been found to lead to N immobilization and reduced biomass yield (Akinyemi and Adebayo, 1985). The contrary findings in this study might be due to the low amounts of sawdust in the treatments which was also mixed with cattle manure in Medium 2, Medium 3, Medium 4 and Medium 5 thereby signifying the potential of mixing cattle manure with sawdust as an alternative enrichment media to forest soil. With respect to treatments amended with cattle manure and sawdust, organic manure additions are known to release large amounts of exchangeable bases which improved the soil reaction thereby affecting the changes in PH of the potting media as observed in this study (Hargrove and Thomas 1981; Keeler *et al*., 2009).

There was increased plant uptake of N in treatments that were enriched with cattle manure and sawdust. The higher production of dry matter though not significantly different in the forest soil and cattle manure treatments compared to forest soil alone would be due to the higher levels of soil available N and P in the media. Calcium and Magnesium availabilities were not affected by microbial decomposition and therefore their level in this study was shown to decline during the experimental period. Soil calcium is one of the macronutrients adversely affected in tropical forest soil. High rainfall amount encourage leaching of the basic cations, such as calcium from upper horizons thereby making them unavailable to plants. Similar findings showing decline in exchangeable calcium were reported in forest soils (Thimonier *et al*., 2001 and Lawrence *et al*., 1997). From these results, the enrichment of forest soil with sawdust and cattle manure enhanced nutrient uptake by the seedlings and hence growth and dry matter yield. However, the optimum ratio of mixing sawdust and other organic amendments ought to be determined before addition to forest soil. The ratios of the organic mixtures would depend on the C: N ratio of the materials. Organic residues with high wide C: N ratios such as sawdust should be applied at lower ratios, while other materials with medium to low C:

N ratios such as cattle manure could be incorporated at higher ratios (Cadisch and Giller, 1997).

This study established that seedlings growth parameters (height and root collar diameter) were enhanced when forest soil was mixed with other organic materials at various ratios. Slow growth at the initial growth (0-8 weeks) could be regarded as establishment stage where roots start to grow and new leaves start to develop. At juvenile growth, phosphorus is important for root growth and the treatments had sufficient available phosphates according to Yost *et al*., (1987). From week 8 to week 24, seedlings showed rapid growth of beyond 10cm in all treatments. Rapid growth could be associated with uptake of available nutrients in the growth media thus attaining a height of 10cm height, which according to Lamprecht (1989) was ideal height for out planting. This suggests that there were readily available nutrients in forest soil and cattle manure mixtures which provided initial nutrient requirement for growth as reported by Mendham *et al*., (2003) and Tanimu *et al*., (2013).

Biomass accumulation in seedlings was an indication of how seedlings were able to grow and utilize soil nutrients from a given medium. Wightman, (1999) indicated that higher gain in root biomass of seedlings while in a nursery can be a predictor of good survival in the field (Duryea and Landis, 2012). Increased seedling biomass with various potting media comprising of mixtures of forest soil, sawdust and cattle manure at various ratios provide evidence for enhanced nutrition of the media when several organic materials are used as alternative source of soil nutrients.

Seedling mortality was low in this study possibly due to enhanced nursery management practices and low seedbed and pot density. Therefore, there was improvement in growth and survival of seedlings after pricking out. Better understanding and implementation of nursery cultural practices to improve seedling quality should enable establishment of quality seedlings after transplanting and improved future growth of forest stands.

The application of plant nutrient testing in forest and conservation nurseries is meant to assist in determination of seedling quality and out planting success. Seedling height, root collar diameter and biomass are special attributes in determination of seedling quality that can be used to predict survival in the field. The weak but positive correlation between the three parameters (height, biomass and root collar diameter) with foliar nutrient concentration indicated that seedlings responded to enhanced nutrient supply in the potting media and thus these results demonstrate the potential for developing new strategies for growing seedlings in the nursery.

CONCLUSION

Use of appropriate ratios of potting media is important in enhancing the nutrition of the media and in establishment and management of seedlings in the nursery. Thus mixing forest soil with cattle manure with or without sawdust gave the best results on seedlings performance in the nursery as per growth parameters measured and enhanced their nutrition, therefore could be considered as good predictor of survival and performance in the field.

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