

Tissue Macro Nutrient Contents and Their Relationship with Growth in Two Dominant Eucalyptus Cultivars Across Three Selected Districts of Uganda

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ABSTRACT

Analyzing nutrient levels in plant tissues and comprehending how they are distributed among different parts is essential for implementing efficient nutrient management strategies across various crops. This study aimed to assess the levels of macro nutrients in different tissues of two prevalent Eucalyptus cultivars and their correlation with growth parameters in Kabarole, Mpigi, and Rakai districts. Two dominant Eucalyptus cultivars, *E. grandis* (local) and GU7 (Improved), were examined for nitrogen, phosphorus, potassium, calcium, and magnesium levels in leaves, bark, branches, and wood. Plant growth was evaluated by measuring diameter at breast height, total tree height, tree volume, and biomass per plot. Results revealed that the GU7 cultivar exhibited a higher accumulation of major nutrients in all plant parts compared to *E. grandis* across the three districts, with a descending order of Leaves > Branches > Bark > Wood for N, P, and K. Whereas, Ca and Mg were predominantly stored in the bark for both cultivars, although more significantly in GU7, in the order of Bark > Branches > Wood > Leaves. Furthermore, a positive significant correlation was observed between most nutrient contents in leaves and growth parameters of Eucalyptus cultivars, although no significant relation was found with phosphorus.

KEYWORDS: *Eucalyptus cultivars, Macro nutrients, Plant parts*

1. INTRODUCTION

Eucalyptus stands out as one of the most industrially significant forest species worldwide, boasting diverse applications as a primary resource for various Eucalyptus-derived products (Bayle, 2019). Thriving across tropical and subtropical regions, Eucalyptus demonstrates remarkable adaptability to diverse environments throughout Sub-Saharan Africa (Gonçalves *et al.*, 2017). With its rapid growth rate, substantial wood density, and versatile utility encompassing charcoal production, timber, and fuel wood, among others, Eucalyptus has earned widespread cultivation (Gonçalves *et al.*, 2017). In Uganda, particularly in the Western Highlands, South Western, and Lake Victoria Crescent Agroecological Zones, *E. grandis* and GU cultivars, notably *E. grandis*, dominate cultivation, with GU7 clones

exhibiting notable environmental resilience (Rocha *et al.*, 2016).

Eucalyptus species typically exhibit high nutrient uptake efficiency, capable of extracting substantial nutrients from fertile soils, leading to nutrient accumulation within their plant structures (Kottek *et al.*, 2006). This nutrient accumulation, predominantly nitrogen (N), phosphorus (P), and potassium (K), profoundly influences growth parameters such as height, stem diameter, and leaf area (Grove *et al.*, 1996; Turner *et al.*, 2008; Leite *et al.*, 2011; Schmidt *et al.*, 2020; De Souza, 2022; Taiz, 2015). Understanding the macronutrient distribution across various tissues leaves, branches, bark, and wood of different Eucalyptus species holds paramount importance for forestry management, encompassing

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both productivity and sustainability (De Souza *et al.*, 2022; Taiz and Møller, 2015). Such knowledge facilitates tailored fertilization strategies and nutrient management plans, optimizing resource allocation and fostering sustainable Eucalyptus cultivation.

Plant boasting higher leaf nutrient content often exhibit enhanced photosynthetic efficiency, thereby promoting accelerated growth (De Souza *et al.*, 2022; Taiz and Møller, 2015). Moreover, species with elevated nutrient concentrations in leaves and stems typically display superior nutrient uptake and accumulation capacities, offering a more efficient utilization of fertilizers under similar soil conditions. This nutritional efficiency underscores the potential for rational fertilizer use, contributing to enhanced productivity and sustainability within the Eucalyptus enterprise.

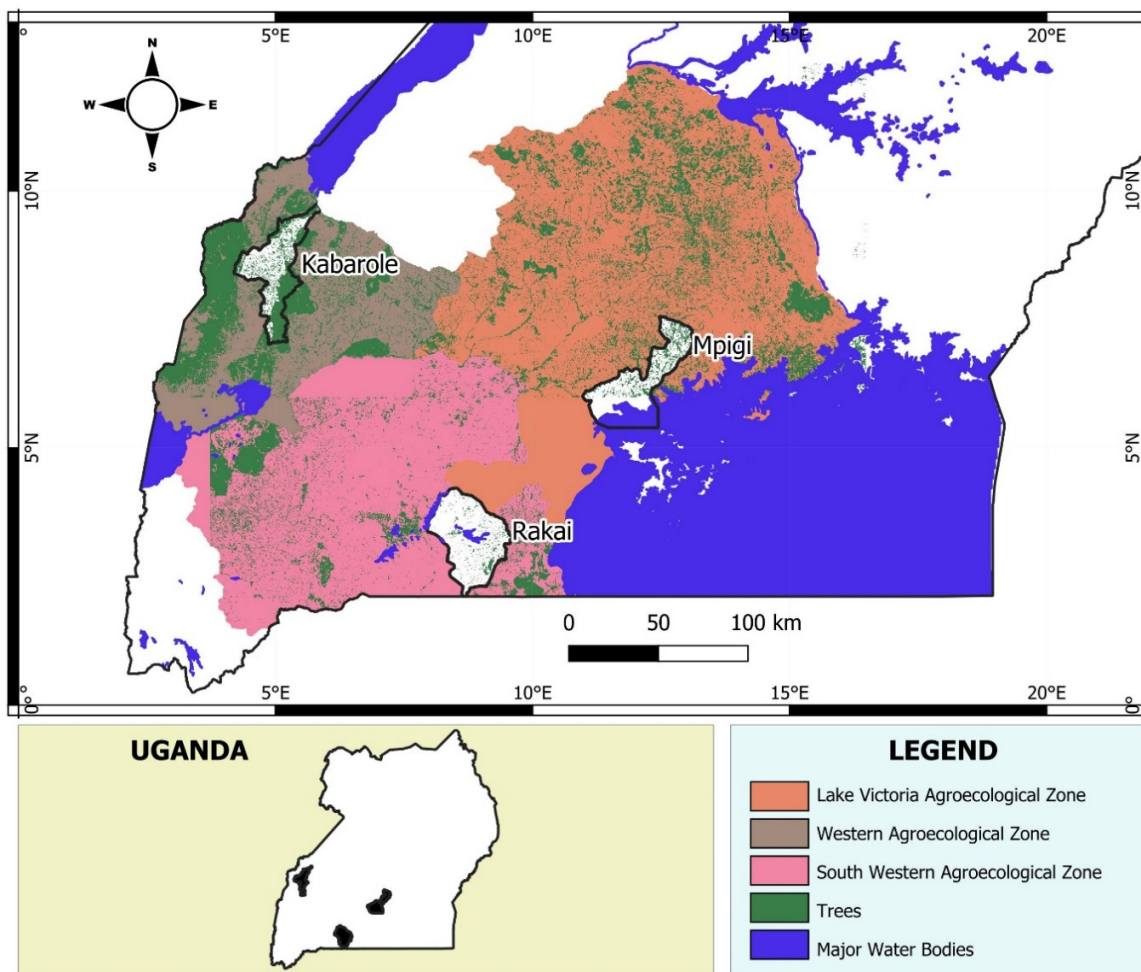
Given these considerations, this study aims to (i) assess macronutrient levels in various tissues of the predominant Eucalyptus species under examination and (ii) correlate these findings with tree growth

parameters to identify critical nutrients for optimizing yields and fostering a sustainable production system.

2. Materials and Methods

2.1. Description of the study area

The study was conducted in three districts which include Mpigi, Rakai and Kabarole that are part of the three agroecological zones that is Lake Victoria Crescent, South western and Western agroecological zones. The three districts were selected due to their increasing trend of Eucalyptus production compared to other agroecological zones in Uganda. The geographical location of the representative districts includes Mpigi (latitude: 0°04'60.00'' and 32°00'0.00 E 0.2274°, Rakai (latitude: 0°39'59.99'' and longitude: 31°24'59.99'') and Kabarole (latitude: 10°27'0" and 39°07'0" longitude. The altitude in Kabarole district ranges between 626 m - 2,919 m, Mpigi district is 1,124 m - 1,344 m, and Rakai district 1,150 m - 1,541 m above the sea level. The total annual precipitation received per study district include 1,431 mm, 183 mm and 1,500 mm for Mpigi, Rakai and Kabarole respectively.



Source (Nankya *et al.*, 2024).

Figure 1: Map representing study districts in the three agroecological zones. The green color represents trees of approximately 15 m height or higher in 2022 as derived from Land use/Land cover raster file built from Sentinel-2 of resolution 10 m (Karra *et al.*, 2021).

2.2. Soil types in the three districts

The soil composition within Mpigi district primarily comprises ferralitic red or yellow sandy to clay loams, commonly referred to as latosols, with a pH range of 5.5 to 6. Despite some leaching, these soils generally demonstrate favorable drainage properties, as detailed in the 2023 annual report of the Mpigi district local government. In Rakai district, more than three-quarters of the soil composition is ferralitic, indicating an advanced stage of weathering with minimal remaining weatherable mineral reserves, as documented by Wortmann and Kaizzi et al. (2019). Although certain heavy clay variants exhibit moderate fertility, sandy soils in this region notably lack essential nutrients. Additional soil types present in the area include lithosols, characterized by young, stony, or rocky surfaces and lacking well-defined horizons as well as alluvial and lacustrine sands and alluvial clays.

Conversely, Kabarole districts boasts favorable soil conditions for agricultural purposes, with geological divisions indicating that approximately 90% of the district is covered by black loam (volcanic) soil. However, specific areas such as Busoro and portions of Hakibaale sub-counties feature red sandy clay loams occasionally underlined by laterites as outlined in the Kabarole District Master plan of 2018.

2.3. Assess the levels of macro nutrients in different tissues of two prevalent Eucalyptus cultivars and their correlation with growth parameters in the study districts

2.3.1. Data collection and Variables Assessed

Plant data collection was performed in June 2023. Different plant samples were picked from a total of 9 farms per district with five farms of each treatment (Cultivar: GU7 and *E. grandis*) which were replicated three times making a total of 27 replications across three districts from which data was collected. From each replicate, 2 trees were sampled of the different plant parts which included the bark, wood, branches, and leaves. The sub samples were measured of the fresh weights which were recorded, labeled and taken to the laboratory for analysis of target macro nutrients.

2.3.2. Measurement of plant growth variables

The plant growth variables were determined in the two selected trees from each plot and these included: diameter at breast height at 1.30 m (DBH, cm), tree height (HT, m) and wood volume (VOL, m³). A tape measure was used to determine the diameter the DBH at breast height. The plant height was measured using a Haglof hypsometer model ECII. For determination of VOL, the trees were cubed using the Smalian method (Campos and Leite, 2013). A disc with a thickness of 3–5 cm was removed from each of the trees cubed, at a position of 1.3 m from ground level. VOL was then estimated according to equation (1) adopted from (Sharma, 2021).

The wood was then separated from the bark using a ball pointed knife, allowing samples of wood and bark to be obtained for the analysis of target macro nutrients. Leaves were also collected from these same trees. To sample the leaves, around 300 g of leaves were collected from each tree, taken from the middle third of the canopy of the tree including the branches (Malavolta *et al.*, 1997). All sampled parts (leaves, branches, bark, and wood) were oven dried at 65 °C until a constant dry mass was obtained.

$$V = \beta D^2 H \quad \text{Equation 1}$$

V = Total volume (inside bark, m³) of a tree where, $\beta = \pi/4$, D = Diameter at breast height, H = Total tree height (m).

2.3.3. Laboratory analysis

The Macro nutrients: nitrogen (N) (g kg⁻¹), phosphorus (P) (g kg⁻¹), potassium (K) (g kg⁻¹), calcium (Ca) (g kg⁻¹) and magnesium (Mg) (g kg⁻¹), were determined in the bark, leaves, branches and wood using wet digestion with HNO₃ + HClO₄ (3:1). Acid digestion consists of completely oxidizing the organic matter in the plant's tissues using acids and high temperature, with the aid of a digester block (Miyazawa *et al.*, 2009). After digestion of the extract, the phosphorus was determined using the vanadate yellow spectrophotometry method, where H₂PO₄⁻ reacts with MoO₄²⁻ and VO₃²⁻ leading to formation of a yellow-colored complex with light absorption in the 420 nm region following laboratory procedures of Miyazawa *et al.*, (2009) whereas Ca, K and Mg were determined using atomic absorption spectrophotometry method.

Meanwhile, nitrogen content was determined by obtaining the dry matter which was weighed (0.1 g) using the Kjeldahl digestion, distillation, and titration technique. The material was then digested using H₂SO₄ + H₂O₂ in a block digester; distilled using a semi-micro Kjeldahl distiller; and titrated using an automatic titrator. NH⁴⁺ was obtained via digestion with H₂SO₄, distilled in an alkaline medium, so that the condensed NH⁴⁺ is collected in the H₃BO₃ solution and titrated with the HCl solution (Miyazawa *et al.*, 2009).

2.4. Statistical Analysis

Initially, an analysis of variance (F test) was carried out and the Duncan's Multiple Range Test was applied to group the eucalyptus species according to the variables evaluated. A significance level of 5% probability was adopted. Consequently, Pearson's correlation coefficients were calculated by examining the covariances, using data from the eighteen assessed traits. From the matrix, a Pearson two tailed test at $p \leq 0.05$ to study the correlation of plant tissue nutrient concentration on growth parameters was performed. Statistical analysis was performed using the *car* package of the R software 3.8.1 version (Team, 2021).

3. Results and Discussions

3.1. Effect of eucalyptus cultivars on plant tissue nutrient composition in the three districts under study

In general, findings (illustrated in figure 2 and table summarised in table 1) suggests that nutrient distribution varies across plant parts and among different Eucalyptus cultivars. This observation aligns with the outcomes of Otavio *et al.* (2023), who explored macro and micronutrient concentrations in various plant parts of different Eucalyptus species in Brazil. Their research revealed interspecies variation across all nutrients, resulting in elevated levels of specific nutrients within the tissue. These outcomes are consistent with prior investigations by Taiz *et al.* (2015) and De Souza *et al.* (2022).

Furthermore, findings (figure 2; table 1) also revealed that nitrogen (N), phosphorus (P), and potassium (K) were predominantly accumulated in the leaves of both cultivars across the three districts, with more pronounced significance ($P < 0.05$) noted in the leaves of GU7. This trend aligns with observations made by Otavio *et al.* (2023) during their research in Brazil on Eucalyptus *urograndis*, where a tendency to store nutrients primarily in leaves was evident. Notably, their study did not indicate substantial levels of macronutrients in the wood and bark, but rather highlighted elevated macronutrient contents in the leaves, suggesting a preferential allocation of nutrients to foliage. Practically, this allocation strategy ensures a stoichiometric balance conducive to photosynthesis, potentially promoting increased wood volume and greater diameter at breast height.

Tissue Nitrogen, Phosphorus and Potassium followed a similar trend (Figure 2a, b, c; table 1) in a descending order of Leaves > branches > bark > wood across Eucalyptus cultivars in the three districts. Similar results were obtained by Márcio *et al.*, (2017) during studies on level of nutrient concentrations for *E. Saligna* and *E. urograndis* species and found out that all macronutrients (N, P and K), except Ca and Mg, the concentration gradient followed the order wood < branches < bark < leaves. Results also showed that Cultivar GU7 significantly ($P < 0.05$) accumulated more N in leaves compared to *E. grandis* in Kabarole and Rakai districts unlike for Mpigi. This is possibly because GU7 cultivar matures very fast requiring more N to increase biomass. Although, generally both cultivars accumulated more macro nutrients in leaves across the three districts. Results obtained by Marcela *et al.*, (2009) indicated that different cultivars can vary in accumulation of nutrients in aboveground biomass. For instance, their study found out that Nutrient content of *E. grandis* presented little variation compared with that of *E. camaldulensis* in Brazil.

The findings once again indicated that GU7 retained significant ($P < 0.05$) levels of phosphorus (P) (Figure 2b) and potassium (K) (Figure 4c) within the twigs and wood in Rakai compared to Eucalyptus *grandis*. Various factors, ranging from ecological to genetic, may contribute to the variation in nutrient concentrations among different species. For instance, a study conducted in Brazil by Otavio *et al.* (2023) demonstrated an 80% variance in nutrient content in leaves, wood, and bark between *E. grandis* and *E. urograndis*.

Additionally, GU7 stored notable quantities of potassium (K) (Figure 2c) across different plant parts compared to *E. grandis* in Kabarole, as opposed to other districts. Environmental factors can act as filters that determine which individuals with specific "response trait" attributes can thrive in forest communities (Keddy, 1992), including those intended for commercial forestry. Plants exhibit different response traits to environmental factors, potentially explaining the variation observed across districts.

Previous research by Leite *et al.* (2011) revealed variability in nutrient concentration in eucalyptus across different plant tissues, primarily concerning nitrogen (N), phosphorus (P), and potassium (K). Nutrient contents tend to decrease in a gradient from leaves to bark, branches, belowground structures, and wood, consistent with the findings of this study. Eucalyptus trees redistribute and store nutrients in their organs as they perform vital functions (Schmidt *et al.*, 2020), leading to varying amounts and concentrations of elements in different tree parts (Akmakjian *et al.*, 2021).

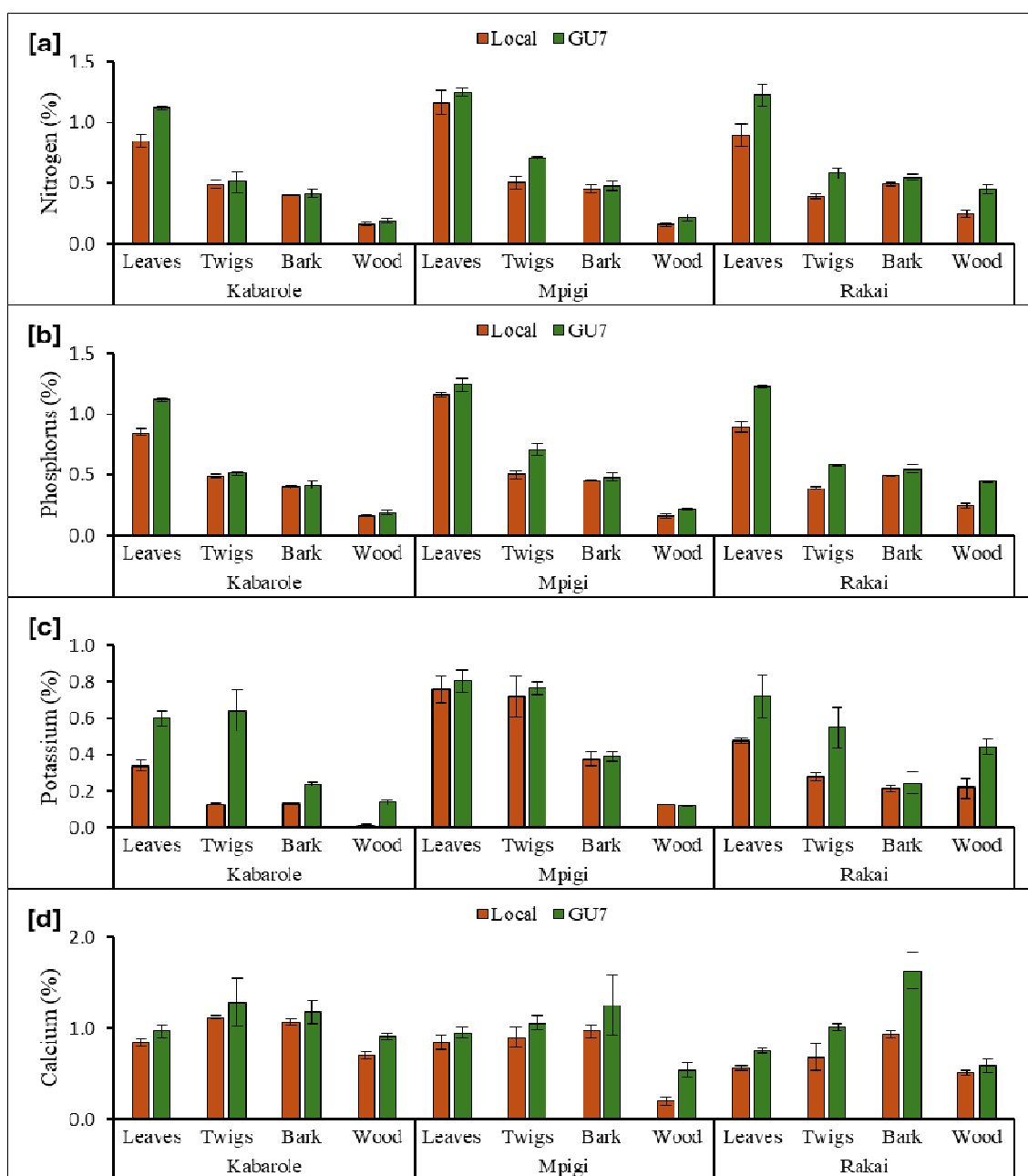
Calcium accumulation in the three districts followed a distinct pattern (table 1; figure 2 d) across plant parts for both cultivars. In Rakai and Mpigi, the order was bark > branches > leaves > wood, while in Kabarole, it differed slightly as branches > bark > leaves > wood. Similar findings were reported by Otavio *et al.* (2023), indicating

that eucalyptus bark is rich in calcium content across various Eucalyptus species in Brazil. However, there may be variations in certain species, resulting in higher levels of specific nutrients in tissues.

Furthermore, high metal contents in tissues can confer protection against herbivory and microorganisms, enhancing plant defense against pests and diseases (Cabot *et al.*, 2019; Lahlali *et al.*, 2022; Hörger *et al.*, 2013). For example, Eucalyptus saligna exhibits resistance to xylophagous agents, such as wood wasps, attributed to copper (Cu), manganese (Mn), and zinc (Zn) contents in bark and sapwood (Cabot *et al.*, 2019; Lahlali *et al.*, 2022).

Magnesium followed a consistent trend (table 1; figure 2 e) across both cultivars in the three districts: bark > branches > wood > leaves. Similar to nitrogen (N), phosphorus (P), potassium (K), and calcium (Ca), GU7 accumulated more magnesium (Mg) than *E. grandis* in various plant parts across the districts. Studies by Otavio *et al.* (2023) in Brazil also observed higher iron (Fe) content in the bark of *E. grandis* and *E. urograndis*, suggesting a potential parallel trend with magnesium accumulation. Remobilizing nutrients from the bark significantly contributes to the biochemical cycle in Eucalyptus trees.

It is essential to assess the suitability of specific eucalyptus tree types in a particular location concerning nutrient availability before planting, as indicated by the results of this study. Different eucalyptus cultivars exhibit variations in nutrient uptake and utilisation which can contribute to more sustainable forest production practices.



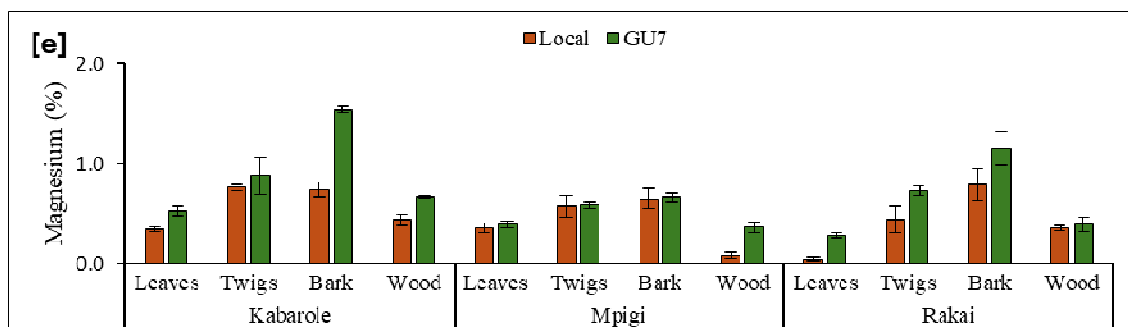


Figure 2: Effect of eucalyptus cultivars in different ecological zones on plant tissue nutrient composition. [a] Nitrogen, [b] Phosphorus, [c] Potassium, [d] Calcium, [e] Magnesium. Error bars indicate standard error.

Table 1: Mean biomass (kg/plot), diameter at breast height (cm), total plant height (m), volume (m³) of eucalyptus varieties under different ecological zones.

Ecological Zones (E)	Varieties (V)	Biomass (kg/plot)	DAB (cm)	Height (m)	Volume (m ³)
Kabarole	Local	1.31 ^c	13.73 ^c	13.00 ^c	0.081 ^c
	GU7	6.36 ^a	25.40 ^a	25.23 ^{ab}	0.545 ^{ab}
Mpigi	Local	3.66 ^b	19.60 ^b	23.10 ^b	0.456 ^b
	GU7	5.51 ^a	27.72 ^a	28.80 ^a	0.725 ^a
Rakai	Local	1.96 ^{bc}	9.63 ^c	13.33 ^c	0.064 ^c
	GU7	2.45 ^{bc}	13.21 ^c	16.30 ^c	0.159 ^c
F-Test					
E	Na	**	***	***	***
V	Na	***	***	***	***
E X V	Na	**	Ns	Ns	*

DAB, Diameter at Breast Height; Na, not applicable; ns, not significant; * Significant ($p \leq 0.05$); ** Significant ($p < 0.01$); *** Significant ($p < 0.001$). Means within columns followed by the same letter do not differ significantly at $p < 0.05$ by Duncan's Multiple tests.

3.2. Variation of yield components in Eucalyptus varieties across the three districts under study

The findings (Figure 3) indicated noticeable differences in growth parameters among the three districts. Specifically, (Figure 3, a) demonstrated a significant ($p < 0.05$) contrast in biomass yield between the two Eucalyptus cultivars, with cultivar GU7 accumulating significantly more biomass than *E. grandis* (Local) in Kabarole and Mpigi. The findings align with those from Camila *et al.* (2016), who demonstrated variations among clones regarding biomass production and allocation, with cultivar GU7 standing out as a distinct clone in this regard.

In both Kabarole and Mpigi districts, a consistent pattern (figure 3 c, b) emerged across the two cultivars, with cultivar GU7 exhibiting significantly larger diameter and taller height ($p < 0.05$) compared to the local cultivar (*E. grandis*), unlike in Rakai. These findings are consistent with previous studies by Camila *et al.* (2016) and Otavio *et al.* (2023), which also reported significant variations in growth parameters among different Eucalyptus genotypes examined in their research.

This time, the findings (figure 3d, table 2) indicated a noteworthy ($p < 0.05$) contrast in plant volume among the three districts examined, specifically between the two Eucalyptus cultivars. These outcomes align with the research of Camila *et al.* (2016) and Otavio *et al.* (2023), who similarly observed variances in plant volume within their respective studies on Eucalyptus species.

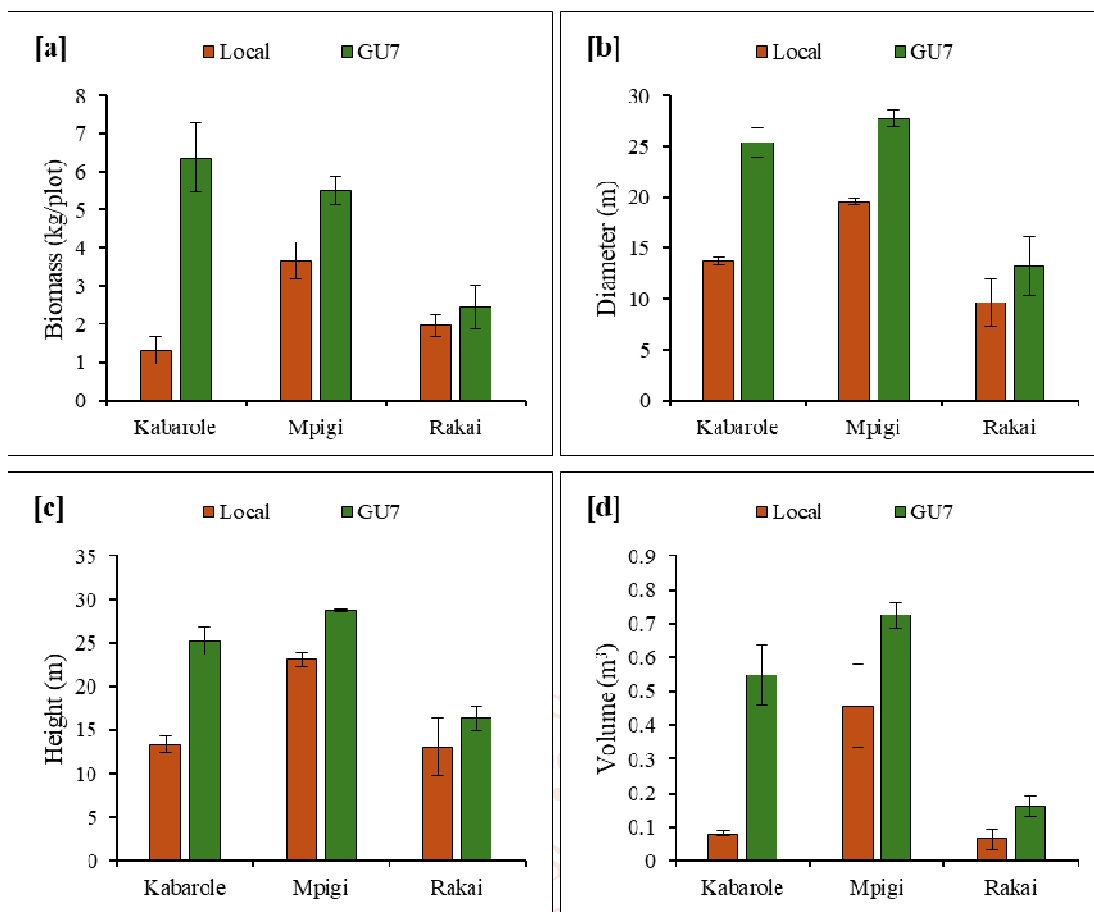


Figure 3: Variation of yield components in Eucalyptus varieties under different ecological zones. [a] Total biomass in a plot, [b] Diameter to breast height, [c] Total plant height, [d] Tree volume. Error bars indicate standard error.

Table 1: Effect of eucalyptus cultivars in different ecological zones on plant tissue nutrient composition

Ecological Zone (E)	Varieties (V)	Plant Part (P)	N (%)	P (%)	K (%)	Ca (%)	Mg (%)
Kabarole	Local	Leaves	0.85 ^b	0.14 ^{ghi}	0.34 ^{ghi}	0.85 ^{defghijk}	0.35 ^{gh}
		Twigs	0.49 ^{de}	0.18 ^{defghi}	0.13 ^{jk}	1.12 ^{bcde}	0.76 ^{cd}
		Bark	0.40 ^e	0.22 ^{abcdefg}	0.13 ^{jk}	1.06 ^{bcdef}	0.74 ^{cd}
		Wood	0.16 ^f	0.14 ^{ghi}	0.01 ^k	0.71 ^{fghijk}	0.44 ^{efgh}
	GU7	Leaves	1.11 ^a	0.25 ^{abcde}	0.60 ^{bcde}	0.97 ^{bcdefg}	0.53 ^{defgh}
		Twigs	0.51 ^{de}	0.26 ^{abcd}	0.64 ^{abcd}	1.30 ^b	0.89 ^c
		Bark	0.41 ^e	0.29 ^{ab}	0.24 ^{ij}	1.18 ^{bcd}	1.54 ^a
		Wood	0.19 ^f	0.23 ^{abcdef}	0.14 ^{jk}	0.91 ^{cdefghi}	0.67 ^{cde}
Mpigi	Local	Leaves	1.16 ^a	0.19 ^{cdefghi}	0.76 ^{ab}	0.85 ^{defghijk}	0.35 ^{gh}
		Twigs	0.50 ^{de}	0.16 ^{fghi}	0.72 ^{abc}	0.90 ^{cdefghij}	0.57 ^{defg}
		Bark	0.45 ^{de}	0.14 ^{ghi}	0.38 ^{fghi}	0.97 ^{bcdefg}	0.65 ^{cdef}
		Wood	0.16 ^f	0.13 ^{hi}	0.13 ^{jk}	0.20 ^l	0.08 ⁱ
	GU7	Leaves	1.25 ^a	0.26 ^{abcd}	0.81 ^a	0.95 ^{bcdefg}	0.39 ^{fgh}
		Twigs	0.71 ^c	0.27 ^{abc}	0.77 ^{ab}	1.06 ^{bcdef}	0.59 ^{defg}
		Bark	0.48 ^{de}	0.18 ^{defghi}	0.39 ^{fghi}	1.25 ^{bc}	0.67 ^{cde}
		Wood	0.22 ^f	0.14 ^{ghi}	0.12 ^{jk}	0.54 ^{jk}	0.37 ^{gh}
Rakai	Local	Leaves	0.89 ^b	0.21 ^{bcdefgh}	0.48 ^{defg}	0.56 ^{ijk}	0.05 ⁱ
		Twigs	0.39 ^e	0.11 ⁱ	0.28 ^{hij}	0.68 ^{ghijk}	0.44 ^{efgh}
		Bark	0.49 ^{de}	0.11 ⁱ	0.21 ^{ij}	0.93 ^{cdefgh}	0.79 ^{cd}
		Wood	0.25 ^f	0.17 ^{efghi}	0.22 ^{ij}	0.52 ^k	0.36 ^{gh}

intensity of the colour (blue for positive and red for negative correlation) between a pair of parameters, respectively. Value nearing to one indicate a strong correlation, and a value closer to zero indicates a less relationship between the two variables. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$ indicate significant correlation between pair of traits.

4. Conclusions And Recommendations

4.1. Conclusion

Eucalyptus cultivar GU7 exhibited the highest level of nutrient accumulation in all plant parts compared to *E. grandis* across the three districts, in a descending order; Leaves>Branches>Bark>wood for N, P and K.

Calcium and Mg is dominantly stored in the bark for both cultivars although significantly in GU7 cultivar in the order; Bark>Branches>Wood>Leaves.

Eucalyptus breeders could take advantage of these relationships to manipulate quality traits while improving yield of the different genotypes.

There was a significant strong positive relationship between most nutrient accumulation in leaves and plant growth parameters of Eucalyptus especially for the GU7 cultivar. This indicates that Eucalyptus effectively utilized the available nutrients for photosynthesis and overall growths which serves as an indicator for optimizing nutrient management strategies in plantations to enhance productivity.

4.2. Recommendations

Eucalyptus growers are advised to restrict the exportation of litter from their plantations in order to minimize nutrient depletion. Additionally, it's suggested that they reduce the practice of de-barking stems during harvesting, a common practice that can result in the loss of vital nutrients such as calcium (Ca) and magnesium (Mg), crucial for plant growth and wood quality. Furthermore, it's recommended that leaves be left in place to decrease the depletion of nitrogen (N), phosphorus (P), and potassium (K), with particular emphasis on preserving leaf N and K, which are highly susceptible to leaching.

Moreover, a deeper understating on nutrient and dynamics within plant tissues, as well as their growth-related characteristics across different forest species, is essential for effective fertilization management. This understanding can inform the selection of more suitable species for specific environments, ultimately leading to improved nutrient utilization efficiency and greater sustainability in the forestry sector through rational and optimal nutrient management practices.

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