

Research Article

Foliar nutrient concentration in first and second rotations of *Pinus patula* at Sao Hill forest plantation in Tanzania

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Foliar nutrients status can be used to determine soil nutrient supplying potential and plant nutrient deficiencies of the site. This study examined the foliar nutrients status of *Pinus patula* in first and second rotation stands in Sao Hill forest plantation, Tanzania. The findings can be used as data base for foliar nutrients status and the basis for improvement of management practices in the plantation. Foliage samples were collected during dry and wet seasons and analysed for macro nutrients. Results showed nutrients concentrations varied with rotations and were mostly within the range of critical levels except Phosphorus which was low implying lower uptake due to low soil Phosphorus availability, probably attributed to low soil pH. Foliar Nitrogen concentration was significantly higher during the wet season compared to other nutrients increased ($p < 0.001$) during the dry season. Foliar Nitrogen, Phosphorus, Potassium, Calcium, Magnesium concentrations and NP ratio were significantly higher ($p < 0.0001$) in PPR1 than PPR2 stands. These contrasting results imply that the increase in rotation negatively affected foliar nutrient concentration in *Pinus patula* at Sao Hill forest. The study recommended improvement in management practices by retaining foliage in the field during harvesting for better nutrient cycling and avoid burning of log trashes.

Key words: Foliar nutrients concentration, nutrient resorption, sao hill, *pinus patula* rotations

INTRODUCTION

Tree leaves and twigs often contain a high proportion of nutrients in the aboveground tree biomass (Jiménez et al., 2015). Specifically, nutrient concentrations in needles have frequently been used for assessing the nutritional status of forest stands in different parts of the world (López-Serrano et al., 2005, Davis et al., 2007, Davis et al., 2010, Albaugh et al., 2010 and Knapp et al., 2016). *Pinus patula* trees originate from Mexico and are widely planted in many parts of the world including Africa. Foliar nutrient concentrations and resorption efficiency of Pines have been used to determine the nutrient-supplying potential of a site and therefore to identify nutrient deficiencies before they appear as visual symptoms and loss in forest productivity (Brockley 2001, Louw et al., 2003, Sánchez-Parada et al., 2018 and Guo et al., 2023).

Forest stand development can cause changes in soil and plant nutrient status and the transition in the kind of nutrient limitation where Nitrogen (N) limitation always occurs in

young forests, while Phosphorus (P) is limited up to old forests, especially in areas with nutrient deficiencies (Yan et al., 2018, Deng et al., 2019 and Guo et al., 2023). Most research has focused on N, P, and Potassium (K) as major limiting nutrients for forest growth, as well as on foliar Calcium (Ca) and Magnesium (Mg) (Lopez-Serrano et al., 2005 and Zhang et al., 2018). Foliage tests provide an integrated index of soil nutrient supply and stand demand (Brockley 2001).

The N:P ratio of green needles has been considered as an indicator for detecting nutrient limitation status of plant growth (Tessier et al., 2003). Koerselman et al., (1996) suggested that N:P values lower than 14 indicate that plant growth is limited by N, while $N:P > 16$ represents P limitation, and N:P values between 14 and 16 indicate that plant growth is co-limited by both N and P. A recent global meta-analysis showed that there is generally a shift from N limitation to P limitation with stand development of forest plantations (Zhang et al., 2022).

The season of foliage sampling has been considered as a source of variations in foliar nutrient levels for *Pinus patula* stands. A study by Sánchez-Parada et al., (2018) in Mexico

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showed critical levels for macro-nutrients N, P, K, Ca, and Mg in percentage as 1.49, 0.13, 0.63, 0.33 and 0.14, respectively. Further, studies conducted in South Africa (SA) for *Pinus patula* stands indicated N exceeding 1.2% as critical for all pines and K concentration of 0.97% was considered sufficient but K with 0.47% was deficient. Louw et al., (2003) in South Africa indicated critical levels in percentage for *Pinus patula* during the wet season as 1.79, 0.12, 0.5, 0.25, 0.10, 0.14 for N, P, K, Ca, Mg and S respectively while NP ratio was 14.9. During dry season the concentrations were 1.24, 0.17, 0.76, 0.17, 0.11 and 0.18 for N, P, K, Ca, Mg and S respectively and NP ratio was 7.6 (Anderson et al., 1993).

Foliar nutrient concentrations normally correlate with forest site index values (Louw et al., 2003). Crous et al., (2009) noted that foliar N, P and K concentrations were increased by the application of 50 kg P ha⁻¹ (Crous et al., 2008). Common nutritional deficiency symptoms in Pines include foliar chlorosis, browning, wilting, exudation, necrosis, growth stagnation, and death of the whole plant, or portion of it (Tangwa et al., 1988). Deficiencies of nutrients in new foliage can be corrected by the plant through resorption from old foliage (Han et al., 2013). Another estimate of nutrient limitations in foliar is Resorption Efficiency (RE) which is computed from the nutrient content reduction in senesced needle (Chase et al., 2016).

Nutrient Resorption Efficiency (NuRE) plays an additional role because it determines the nutritional quality of leaf litter entering the soil, influencing the mineralization rates and thus the release of nutrients in organic matter to plants and soil microbes (Scalon et al., 2017). The “relative resorption hypothesis” states that plants tend to resorb a greater portion of nutrients that limit their growth (Han et al., 2013), and the ratio of phosphorus RE to nitrogen RE (PRE: NRE) has been used to evaluate relative limitation of plant growth between N and P in forest ecosystems (Killingbeck 1996, Wright et al., 2003, Kobe et al., 2005, Reed et al., 2012, Tully et al., 2013; Yan et al., 2018 and Du et al., 2020).

The number of forest rotations has been noted previously in Tanzania as one of the factors that influence changes in soil properties in areas with *Pinus patula*, *Pinus elliotii* and *Pinus caribaea* (Tangwa et al., 1988), but study on how foliar nutrient

status changed between first and second rotations in Pines is scarce. Understanding nutrient status in needles of *Pinus patula* in areas planted as first and second rotations is important as it can be used to indicate high or low nutrient status in the sites and enable the management of Sao Hill forest Plantation (SHFP) to take corrective measures on time without rely on soil testing which is more expensive (Crous et al., 2009). This research investigated the variation of foliar nutrient status between the first and second rotations of *Pinus patula* in SHFP by examining the concentrations and resorption efficiency of Nitrogen, Phosphorus, Potassium, Calcium, Magnesium and Sulphur in current Green Needles (GN) and old Senescent Needles (SN) of *Pinus patula* stands during dry and wet seasons (Crous et al., 2011).

MATERIALS AND METHODS

Study areas

The study was conducted in the Sao Hill forest plantation located between 8°18'-8° 33' S and 35° 6'-35° 20' E in the Southern highlands of Tanzania. Sao Hill plantation has a total area of 135,903 ha (Tanzania Forest Services Agency, 2019). Mean annual rainfall ranges from 750 to 2010 mm falling between November and April and temperature ranging from 15°C to 25°C per annum. The soil is mainly dystricitiosols in association with orthic Acrisols (Ngaga, 2011). The plantation has four management divisions (I, II, III and IV) (Tanzania Forest Services Agency, 2019) and in every division, one location was selected in this study area. The locations involved were Irundi, Matanana, Nundwe and Kitasengwa in Division I, II, III and IV, respectively. Within each location, two sites with PPR1 and PPR2 were randomly selected (Deligöz et al., 2019). The altitude of the study sites varies from 1298 m meters above sea level (m.a.s.l) at Kitasengwa to 1950 m.a.s.l at Nundwe as shown in Figure 1.

Vegetation includes natural forest with 48,200 ha dominated with *Brachystegia spp*, *Julbernadia spp*, *Albizia spp*, *Uapaca spp*, *Erythrina spp* and *Dombeya rotundifolia* (Kangalawe, 2018) and exotic forest with *Eucalyptus grandis*, *Eucalyptus maidenii*, *Eucalyptus saligna*, *Cupressus lusitanica*, *Pinuspatula*, *Pinus radiata* and *Pinus eliotii* (Ngaga, 2011).

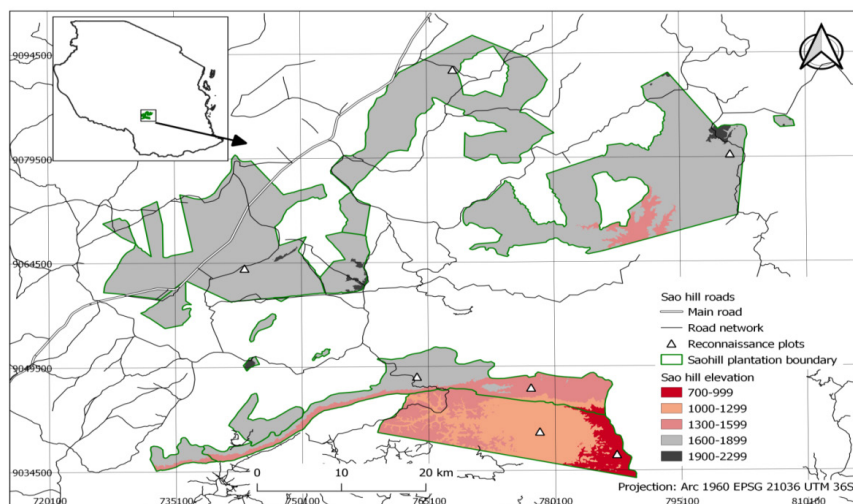


Figure 1. Sketch of SHFP with study sites located at four divisions in Mufindi Iringa, Tanzania.

Note: Sao hill roads (—): Main road; (—): Road network; (Δ): Reconnaissance plots; (□): Saohill plantation boundary; Sao hill elevation: (■): 700-999; (■): 1000-1299; (■): 1300-1599; (■): 1600-1899; (■): 1900-2299.

Rotations

Two rotations PPR1 and PPR2 with trees of 10 years old were involved in this study. The stands in PPR1 were established soon after natural vegetation cleared while the PPR2 was established in land occupied by *Pinus patula* for 25 years and all trees harvested for timber (Maliondo et al., 2005).

Sampling design

Stratified sampling was used where four locations (Blocks) with 8 compartments with two treatments (PPR1 and PPR2) were randomly selected. The sampling units were plots in which eight trees with larger, medium, and lower sizes selected for foliar sampling. Ten (10) quadrant plots measuring 20 m x 20 m (400 m²) per compartment were laid out at different slope positions following the contour at an interval of 100 m along. Total of 80 plots were used for foliar sampling in the study area (Mavimbela et al., 2018).

Foliar sampling, processing, and analysis

From each plot, Green Needles (GN) were sampled from the middle crown position and Senescent Needles (SN) were sampled in the lower crown from eight (8) trees tagged with ribbons. A bulked needle sample from 8 trees pooled into two samples one for GN and another for SN per plot (Okalebo et al., 2002). Total composited samples collected in ten 10 plots per compartment were 10 for GN and 10 for SN during dry season and repeated during rainy season from the same tagged trees (Reuter et al., 1997). Composited needle samples were cleaned by removing dust using wet cotton wool and placed in paper bag and immediately transported to the SUA laboratory for further processing.

In the laboratory needle samples were dried at 70°C for 48 hours, grounded and sieved through 1.5 mm screen. Chemical analysis involved different methods as shown in Table 1.

Table 1. Summary of methods for foliar nutrient analysis.

SN	Test parameters	Methods to determine the parameter	Reference
1	Foliar N	1 g of foliar sample determined by micro-Kjeldahl method.	Okalebo et al., 2002
2	Foliar P	1 g of foliar sample extracted using 6N HCl and 0.1 ml from extract measured colorimetrically for P (Ammonium molybdate method) at 884 wave lengths.	Okalebo et al., 2002
3	Foliar S	1 g of foliar sample extracted by 6N HCl digestion and 1 ml from extract determined turbid metrically for S and was measured using UV-spectrophotometer at 535 wavelengths.	Louw et al., 2003
4	Foliar K, Ca, and Mg	Foliar sample dried at 450°C for 16 hrs and 1g of the ash was dissolved in 6N HCl solution made to 25 mls, and Ca and Mg measured from the extract using atomic absorption spectrophotometer while K measured using flame emission spectrophotometry.	Okalebo et al., 2002

Calculation and statistical analysis

The mean and standard error of different nutrients in the needles and that of tree growth were obtained using descriptive statistics by applying GLM procedures in R packages (Schutz 1990). The Resorption Efficiency (RE) of the elements was calculated using the following equation:

$$RE = 1 - \frac{SN}{GN} \times MLCF \times 100\% \dots \dots (1)$$

Where RE=Resorption Efficiency; GN=concentration of Green Needle; SN=concentration of Senescence Needle; and MLCF=Mass Loss Correction Factor of 0.745 for coniferous (Vergutz et al., 2012).

To get the effects of seasons and rotations in foliar nutrients concentration, data was subjected to Nested linear mixed models analysis using R packages (See et al., 2015). This analysis was carried out using the “lmer” function from the “lme4” package in R. In this model, location was considered as random factors. This model was used to minimize the effect of locations (random factor) on foliar nutrients contents.

RESULTS

The comparisons between foliar nutrient concentrations in PPR1 versus PPR2 were made during wet and dry seasons (Seidel et al., 2022). The mean and p-values were used to indicate results between rotations for foliar Nitrogen (N), Phosphorus (P), Potassium (K), Calcium (Ca), Magnesium (Mg), and Sulphur (S).

Variation of foliar nutrient concentrations between rotations in SHFP

Foliar N, P, K, Ca, Mg, S concentrations and NP ratio differed significantly (p=0.001) between rotations of *Pinus patula* as shown in Table 2.

Results in Table 3 showed that foliar Ca differed significant between rotations (t=-2.39, p<0.05), Mg concentrations in foliar differed significantly (t=7.6, p<0.0001), Foliar K and S were also differed significantly between PPR1 and PPR2 (t=-2.32, p<0.05) and (t=-2.16, p<0.05) respectively (Wallace et al., 1982). The N, P and N:P ratio were also studied but the values obtained did not differed significant between rotations. Foliar Mg was higher in PPR2 than in PPR1 as shown in Figure 2. In contrast, foliar Ca as shown in Figures 3a and 3b K, and S concentrations were significantly higher in PPR1 than in PPR2 as shown in Figures 4a and 4b. In summary, foliar P and K, concentrations decreased with an increase in rotation while N, Ca, Mg, S concentrations and N:P ratio were higher in the second rotation as shown in Table 3.

Seasonal variations of foliar nutrients concentration in SHFP

Seasonal variation of foliar nutrients: The seasonal change from wet to dry has an influence on the concentrations of foliar nutrients in the first and second rotations of *Pinus patula* stands. Foliar Ca, K, P, and N:P ratio differed significantly (p<0.05) between PPR1 and PPR2 during dry and wet seasons (Wu et al., 2020). Foliar P was significantly higher as shown in Figure 5 in dry seasons than in the wet seasons (t=-2.59, p=0.01) both in PPR1 and PPR2. While, Ca was significantly higher as shown in Figure 3a in the wet season (t=3.89,

p=0.0001), as well as N:P ratio (t=3.6, p=0.001) as shown in Figure 6. In contrast, K as shown in Figure 4a was higher in the dry season in PPR1 while Mg and S were higher during dry season in PPR2 as shown in Table 4.

Foliar Nutrients Resorption Efficiency (NuRE)

Results in Table 5 show RE of different nutrients varied significantly between rotations (p<0.0001). Foliar RE of N, P, K, Mg and S were significantly higher in PPR1 than in PPR2

while CaRE was higher in PPR2. These elements in needles were highly resorbed in PPR1 except Ca as shown in Table 5.

Therefore, plants found in soils with poor nutrients have higher NRE and PRE compared to plants in areas with high nutrients (Kobe et al., 2005). The higher NuRE in some nutrient elements in our research implies most of the areas cleared for the establishment of PPR1 at SHFP are poor in some nutrients in the soil.

Table 2. Mean ± SE of foliar nutrients at PPR1 and PPR2 in SHFP.

Rotations	Foliar nutrient concentration (%)						
	N	P	K	Ca	Mg	S	N:P
PPR1	1.27 ± 0.05	0.12 ± 0.01	0.85 ± 0.06	0.25 ± 0.02	0.14 ± 0.01	0.20 ± 0.01	14.5 ± 1.2
PPR2	1.32 ± 0.03	0.1 ± 0.01	0.67 ± 0.02	0.23 ± 0.02	0.17 ± 0.01	0.17 ± 0.01	16.3 ± 1.0

Table 3. Variation in foliar nutrients at PPR1 and PPR2 in SHFP.

Mean	Foliar macro nutrients concentrations						
	Ca	Mg	K	P	S	N	N:P
	0.24 ± 0.02	0.16 ± 0.01	0.71 ± 0.02	0.11 ± 0.01	0.18 ± 0.01	1.30 ± 0.03	15.6 ± 1.0
P-value	0.0179*	2.63-12***	0.0216*	0.2856	0.0325 *	0.8154	0.2559
t-statistic	-2.392	7.605	-2.321	-1.071	-2.158	0.234	1.14
Status	*	***	*	NS	*	NS	NS

Note: ***: 0; **: 0.001; *: 0.01; . *: 0.05; 1: 0.1.

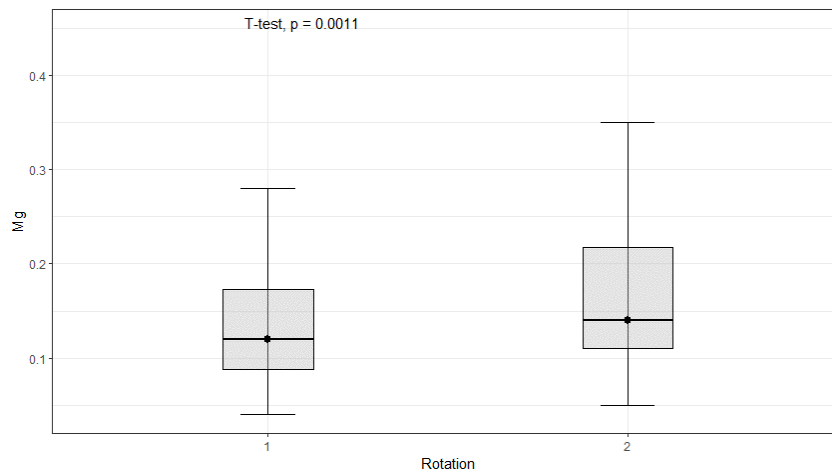


Figure 2. Foliar magnesium concentration at different rotations in SHFP.

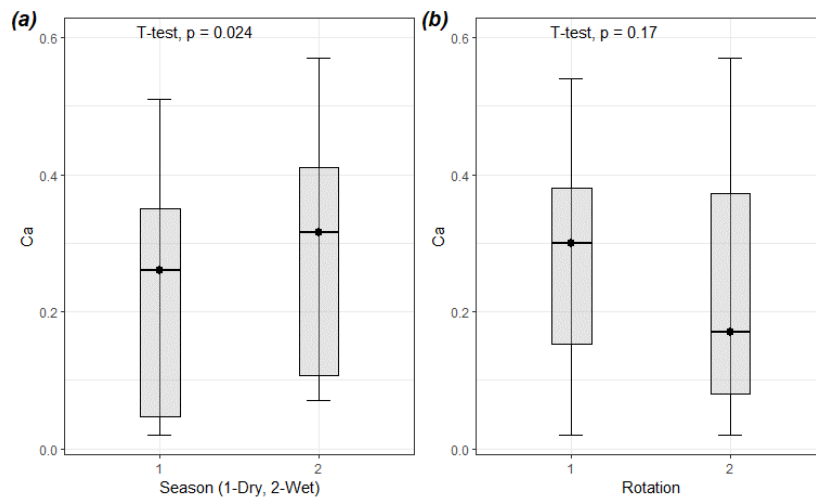


Figure 3. Foliar Calcium in (a): Different seasons and; (b): Rotations in SHFP.

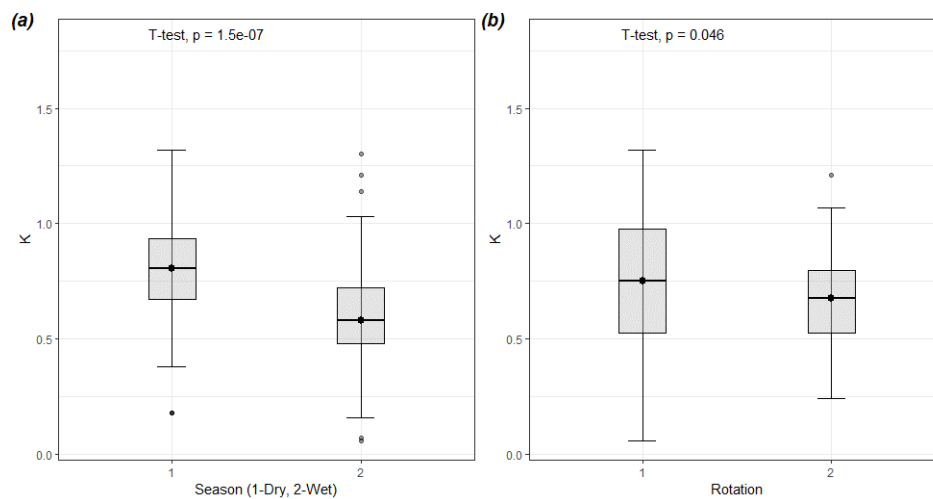


Figure 4. Foliar Potassium (a): Different seasons and; (b): Rotations at SHFP.

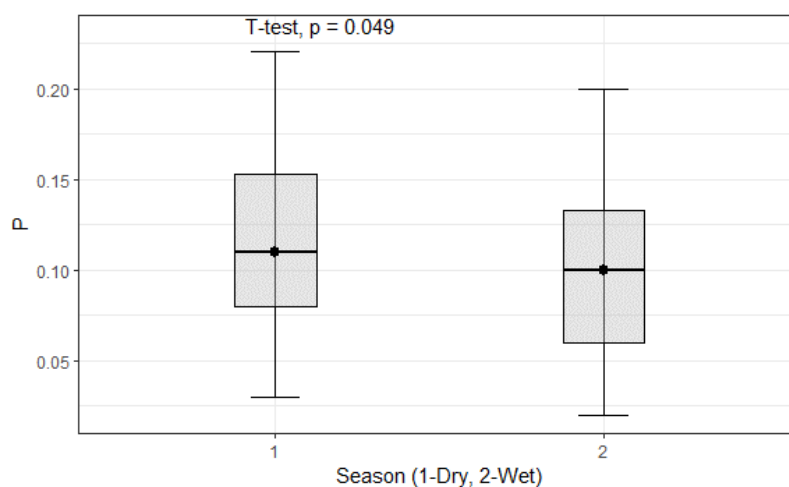


Figure 5. Foliar Phosphorus in different seasons at SHFP.

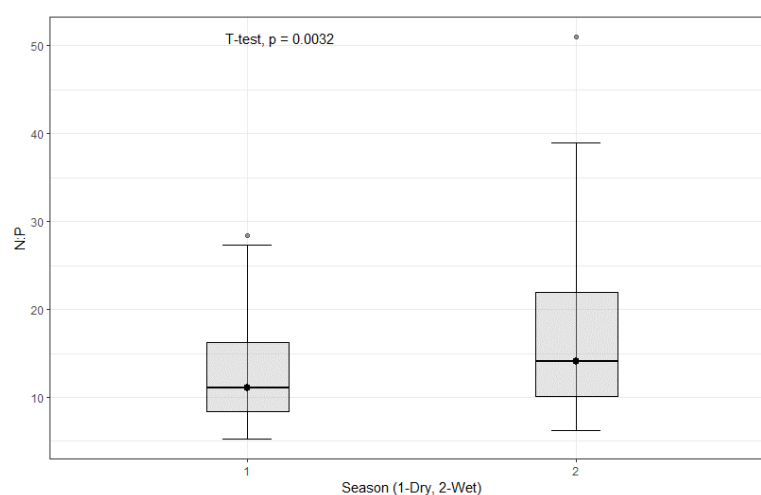


Figure 6. Foliar N:P ratio in different seasons at SHFP.

Table 4. Mean and variation of foliar nutrients in PPR1 and PPR2 during dry and wet seasons in SHFP.

		Foliar macro nutrients concentrations						
Seasons		Ca	Mg	K	P	S	N	N:P
PPR2	Dry	0.2 ± 0.03	0.18 ± 0.01	0.78 ± 0.02	0.11 ± 0.01	0.2 ± 0.01	1.26 ± 0.05	13.6 ± 1.1
	Wet	0.26 ± 0.03	0.17 ± 0.01	0.55 ± 0.02	0.1 ± 0.01	0.15 ± 0.01	1.37 ± 0.05	17.84 ± 1.64
PPR1	Dry	0.24 ± 0.02	0.14 ± 0.01	0.85 ± 0.06	0.12 ± 0.01	0.19 ± 0.01	1.27 ± 0.04	13.05 ± 1.2
	Wet	0.29 ± 0.02	0.13 ± 0.01	0.66 ± 0.04	0.1 ± 0.01	0.16 ± 0.01	1.35 ± 0.05	17.76 ± 1.98

P-value	0.000146***	0.0845	2.37-08***	0.01053*	0.7139	0.0823	0.000422***
t-Stat	3.895	-1.736	-5.888	-2.589	0.367	1.749	4
Status	***	NS	***	*	NS	NS	***

Note: ***: 0; **: 0.001; *: 0.01; . *: 0.05; 1: 0.1.

Table 5. Mean \pm SE and variation in NuRE of PPR1 and PPR2 foliar in SHFP.

Nutrients	N	P	K	Ca	Mg	S
Mean PPR1	38.81 \pm 0.95	42.24 \pm 2.05	43.46 \pm 2.69	39.99 \pm 2.05	46.43 \pm 1.83	43.88 \pm 1.39
Mean PPR2	36.87 \pm 0.78	36.69 \pm 1.55	36.07 \pm 1.79	40.37 \pm 1.45	41.21 \pm 1.43	42.52 \pm 1.75
t-stat	-57.4	-28.59	-22.87	-30.95	-35.04	-37.37
P-values	1.77-66***	9.97-44***	6.87-37***	3.14-46***	3.36-50***	2.79-52***
Status	***	***	***	***	***	***

Note: ***: Denotes significant difference at $p < 0.0001$; SE: Standard Error; R: Resorption.

DISCUSSION

Foliar nutrients concentration of PPR1 and PPR2 in SHFP

Foliar N obtained in this study was higher in PPR2 than PPR1 but the difference was not significant and N was lower than the critical levels of 1.9% reported in Swaziland, Louw et al., (2003). Sánchez-Parada et al., (2018) in Mexico for *Pinus patula*, and was within the lower critical level of 1.0%-1.3% indicated by Sánchez-Parada et al., (2018). This implies that there is a deficiency of N in all studied sites at SHFP. The lower level of N in foliar of both PPR1 and PPR2 in both seasons might be caused by the nature of the soil, previous land use, previous vegetation type, soil moisture availability and resorption efficiency of needles.

The values of foliar P were higher in PPR1 than PPR2 but the differences were not significant. The obtained values are within the critical level of 0.12 to 0.17% indicated by Louw et al., (2003) and 0.13% reported by Sánchez-Parada et al., (2018) in Mexico hence sufficient except in PPR2 with p values lower than the stated critical level in previous studies. The lower foliar P in PPR2 might be due to low soil pH, presence of clay soil and previous land use.

Foliar K was higher in PPR1 than PPR2 and differed significantly. The K in this study was above the critical levels of 0.63% stated by Sánchez-Parada et al., (2018) and above 0.76% by Louw et al., (2003) therefore, foliar K is sufficient in all sites under study. The lower values of K obtained during the wet season might be due to dilution factor in the soil and plant tissues since K is very soluble and therefore highly mobile in plant tissues and is almost completely provided by the weathering of soil parental material.

Foliar Ca was higher in PPR1 than PPR2 and differed significantly. In our research Ca was below the critical value of 0.33% reported by Sánchez-Parada et al., (2018) during the dry season, also above 0.25% reported by Louw et al., (2003) during the wet season, hence sufficient. The lower level of Ca during the dry season might be due to soil pH (acidity), texture (Sandy and clay) and the presence of high Mn, K and Na in the foliage.

Foliar Mg was higher in PPR2 than PPR1 and differed significantly but was within the critical level of 0.14% reported by Sánchez-Parada et al., (2018) in Mexico and 0.11% indicated by Louw et al., (2003) in South Africa in both seasons, hence

sufficient.

Foliar S was higher in PPR1 than PPR2 and differed significantly. The values of S are within the critical level of 0.14%-0.18% reported by Louw et al., (2003) for *Pinus patula*, hence sufficient in foliar. The N:P ratios of 13 in PPR1 and 15 in PPR2 did not differ but were high compared with (11.4) reported by Sánchez-Parada et al., in Mexico; also, higher than 7.6 during the dry and 14.87 in the wet season reported by Louw et al., (2003) in South Africa. This indicates that the P requirement by *Pinus patula* is higher hence the need for P supplements in SHFP.

Resorption efficiency of foliar nutrients in PPR1 and PPR2 at SHFP

Resorption Efficiency (RE) in this study varied from one element to another. Foliar N, P, K Mg and S were highly resorbed in PPR1 except Ca resorbed more in PPR2 which implies the concentrations of these nutrients in green needles of PPR1 are lower, using the conserved from old needles and according to Han et al., (2013) and Guo et al., (2023) high resorption is an indicator of nutrient deficiency in plants. Our results concur with the study by Guo et al., (2023) who recorded high RE in foliar of Pines in areas with low foliar nutrients. Therefore, PPR1 needles have lower nutrients which lead to high resorption. An increase in nutrient resorption efficiency has been noted by other scholars as a sign of nutrient limiting in plants (Han et al., 2013, Yan et al., 2018 and Du et al., 2020). Previous studies indicated nutrient RE to correlate negatively with soil nutrients and some plants growing in infertile soil did not always show higher nutrient RE (Gerdol et al., 2019, Wang et al., 2021 and Guo et al., 2023).

Variations of foliar nutrients for *Pinus patula* in sao hill forest plantation

Inter-element variation in foliar of PPR1 and PPR2: Macronutrients studied in foliar varied in their contents from PPR1 to PPR2. Significant increase in foliar N and Mg and decrease in P, K, Ca and S from PPR1 to PPR2 might be caused by changes in soil pH, soil organic carbon and management practices during the first rotation, mycorrhizal association and ability of root to absorb nutrients.

Seasonal variation in foliar nutrient concentration for PPR1 and PPR2 in SHFP

Foliar nutrients were varied with the season of sampling, where high concentrations of P, K, S and Mg were recorded

in PPR1 and PPR2 during the dry season. The reasons for less concentration of foliar nutrients during the wet season are the dilution factor and the involvement of elements in different parts of plant tissues. These results are consistent with the study by Louw et al., (2003) who recorded the same trend of high foliar nutrients in needles of *Pinus patula* during dry or dormant season for P, K and Mg.

Variation in resorption efficiency of foliar nutrients at PPR1 and PPR2 in SHFP

Rotations affect the NuRE where RE of N, P, K, Mg and S decreased significantly from PPR1 to PPR2 and Ca increased. The reasons for the observed changes might be when forest cleared, and log trash burnt caused the loss of some nutrients in the soil; or the inability of root to absorb nutrients due to low moisture content of the soil and compaction during harvesting. The results of NuRE in this research are in agreement with studies by Kobe et al., (2005) and Reed et al., (2012) who indicated plants grown in soils with poor nutrients have higher nitrogen RE compared to plants in an area with high nutrients and the findings also agreed with study by Killingbeck (1996) and Yan et al., (2018) who indicated higher nitrogen RE and phosphorus RE in foliar in areas with limitations of N and P in the soil.

CONCLUSION

Foliar nutrients concentrations were within the critical levels for *Pinus patula* but decreased from PPR1 to PPR2 and they are sufficient for most studied nutrients. Foliar P in PPR2 was below the recommended critical levels hence deficiency in the study area. Higher foliar concentrations of most elements were recorded during the dry season except N and Ca. The resorption efficiency of foliar nutrients was higher in PPR1 indicating that nutrients especially N, P, K, Mg and S are deficient in the needles of PPR1. We recommend improvement in management practices by retaining the foliage in the field during harvesting for better nutrient cycling. Further research is needed to relate foliar nutrients with tree growth, and soil physical and chemical properties in forest plantations within Tanzania to ascertain foliar and soil nutrient status after the second rotation.

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