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Research Paper



Combined Effects of Moisture and Defoliation on Growth and Leaf Chlorophyll Content of *Voacanga africana*

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ABSTRACT: This study was aimed at investigating the interactive effect of soil moisture content and defoliation on growth and leaf chlorophyll content of a medicinal plant species Voacanga africana. A greenhouse experiment was conducted where plants were subjected to different levels of soil moisture content (low and high) and defoliation (control, moderate, severe) in a split-plot design with two replications. Soil moisture content was the main plot while the defoliation treatments constituted the sub-plots. Plant growth parameters including height, number of leaves, mid height diameter, root collar diameter, leaf length, leaf area, dry mass and leaf chlorophyll were measured three months after the commencement of treatments. Data were subjected to split-plot ANOVA and where necessary, means separation was conducted with Scheffe's test. There was a significant effect of soil moisture on some parameters. Height, leaf area, shoot biomass, root biomass, and total biomass were suppressed by the low moisture regime. Height was also influenced by defoliation, experiencing a decline from the control to severe defoliation level. Root : shoot ratio and leaf chlorophyll content was neither affected by soil moisture content nor defoliation. Furthermore, there was no significant interactive effect of treatments on any of the parameters examined. Given that the stress imposed by the low moisture regime was mild, it is warranted that a further study with much lower moisture levels that are characteristic of soils within species' ecological range be conducted to determine if the absence of a significant interaction between soil moisture and defoliation is sustained.

KEYWORDS: Biomass, Chlorophyll content, Defoliation, Morphology, Soil moisture, Voacanga africana

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I. INTRODUCTION

Defoliation is defined as the relative number of missing needles or leaves in the assessable crown as compared to a reference tree (Eichhorn and Roskams, 2013). Defoliation can result from damage by branches falling from forest canopies, browsing, arthropod attack, disease, competition and alterations in environmental conditions like temperature and soil moisture content. These biotic and abiotic influencers constitute key determinants of plant functioning, growth, and forest productivity (Seidl et al., 2011; Lavoie, et al., 2019; Ambebe et al., 2024). Folia play an important role in determining photosynthetic potential and growth rate of many plant species. Any modification in plant canopy affects photosynthesis of individual leaves there by influencing growth. Thus, one aspect of tree plant improvement is to maintain a critical leaf number and leaf area for the greatest photosynthetic capacity and most efficient metabolism.

The amount of water needed for proper growth and development varies across plant species and its availability often depends on other factors like temperature, wind current, evapo-transpiration rate and soil texture, among others (Cai et al., 2010; Stock, 2014; Ambebe and Tanwie, 2020). The importance of water to plant growth and functioning cannot be over emphasized. For example, the processes of photosynthesis and translocation of photosynthetic assimilates are strongly dependent on plant water status. Moreover, soil nutrients are often dissolved in water before being taken up by plants. Excess or limited soil moisture availability may affect the plant negatively with visible stress indicators like withering, low biomass production and stunted growth. Similarly, severe low soil moisture stress can cause the death of the entire plant or suppress growth through its damaging effects on the photosynthetic machinery (Cai et al.; 2010; Ambebe and Tanwie, 2020; Mkhanzi, 2023).

Voacanga africana (Apocynaceae) is an understorey tree of the African tropics that is native to Cameroon and other countries where it is found in forests and savannah woodlands (POWO, 2024). The species is highly perceptible in the Western Highlands of Cameroon where its pharmaceutical potential is greatly exploited. Fruit, bark and leaf extracts are used for the treatment of orchitis, ectopic testes and gonorrhoea, respectively, in Cameroonian ethnomedicine (Jiofack et al., 2008; Tan et al., 2000; Hussain et al., 2012, Katjima et al., 2013). Furthermore, the wood is used locally as building material, fuel and for making musical instruments, arrow and knife sheaths. Branches falling from forests canopy often inflict damage on understorey plants (Chazdon, 1991), related to breakages of woody structures and reductions of foliage area. With wide variations in edaphic factors and relief across the landscape of the Western Highlands, soil moisture and defoliation are likely to interact in controlling the developmental aspects of plants. This study was aimed at investigating the effect of soil moisture availability on the response of growth and leaf chlorophyll content of V. africana seedlings to defoliation.

Study site

II. MATERIALS AND METHODS

The experiment was conducted in a greenhouse at The University of Bamenda (1444 m asl; 5.983° N, 10.250° E), Bambili, North West Region, Cameroon. Bambili is characterized by a rainy season that extends from March to September and a dry season that starts in September and ends in March. Under a changing global climate, however, there have been variations in the timing and duration of the seasons in recent years. Bambili has as mean annual rainfall 2095 mm, temperature 22.51 °C, and relative humidity 75.96 %. With over 350 mm of rainfall each, the wettest months are July, August, and September while the driest is January with 6 mm of precipitation. The warmest month is February with an average temperature of 29.8/16.1 °C (high/low) while August is the coldest with 21.6/15.6 °C (Weather Atlas, 2024).

Plant Material

The study ran from the 7th of March to the 7th of June 2024. Four months old seedlings of Voacanga africana were obtained from the Reforestation Task Force (RETAFO) nursery, Bamenda III Sub-division, North West Region, and transported to the experimental site at the Department of Forestry and Wildlife Technology on the campus of the University of Bamenda. Seedlings were transplanted into polythene bags filled with soil from the compost site. The seedlings were of uniform size and normally developed with no visual sign of damage. They were irrigated immediately after planting.

Experimental Design

The treatments which consisted of two moisture levels (high and low) and 3 defoliation levels (0%, 50% and 70%) were laid out in a split plot design with two replications. The moisture levels constituted the main plot while the defoliation treatments the sub plots. The low and high moisture levels were achieved by irrigating the plants once and thrice a week, respectively, with normal tap water while the defoliation was done artificially using a pair scissors. Defoliation was done once and the seedlings protected from any further abrasions or damage. There were 8 randomly assigned seedlings in each soil moisture and defoliation treatment.

Data collection

At the end of the experiment, three seedlings were randomly selected from each treatment combination for measurements. Number of new leaves was recorded, plant height was measured from the root collar to the tip of the shoot using a ruler, and diameter at the root-collar was measured with a Vernier caliper. The chlorophyll content of the most fully expanded leaf was measured with a SPAD-502 Chlorophyll Meter (Konica-Minolta Co. Ltd., Tokyo, Japan) after which it was harvested and spread out on a graph paper for leaf area determination. The leaf area was noted from the number of 1 cm2 and 0.04 cm2 cells in the region covered by the leaf on the graph paper. The roots of the plant were rinsed free of soil and the seedling dissected into root and shoot. The weight of each fragment was taken with an electronic balance after drying to constant weight in an oven.

Statistical analysis

The data was examined for homogeneity of variance and normality using probability plots and histograms. Split-plot ANOVA was used to examine the effect of moisture, defoliation and their interactions on the growth and leaf chlorophyll content. Where necessary, means were separated using Scheffe's test. The analyses were carried out in Data Desk 6.01 at p = 0.1.

Morphology

III. RESULTS

There was a significant effect of soil moisture on height and leaf area (Table 1). Both traits declined from the high to low moisture regime (Figure 1).

 Table 1: p-values from ANOVA for the effect of soil moisture, defoliation and their interaction on growth of

 Voacanga africana seedlings

Source	Moisture	Defoliation	Moisture × Defoliation
Height	0.0218	0.0778	0.6187
Root-collar diameter	0.1805	0.3863	0.8347
Number of leaves	0.2148	0.3434	0.6694
Leaf area	0.0976	0.5734	0.4172
Shoot mass	0.0994	0.1677	0.4266
Root mass	0.0994	0.6904	0.5059
Total mass	0.076	0.3611	0.56
Root : Shoot mass	0.3468	0.3839	0.1534
Chlorophyll content	0.386	0.8408	0.8374

On the other hand, height was the only morphological parameter that responded to defoliation, decreasing from the control to the highest defoliation level. However, the 50% defoliation treatment did not differ with either the control or 70% defoliation for height (Figure 1). Root-collar diameter and number of new leaves were unaffected by either of soil moisture content or defoliation. Furthermore, the interactions of treatments did not have a significant effect on any of the traits (Table 1, Figure 1).



Figure 1: Effect of soil moisture content and defoliation on morphology. Values are mean \pm SE. The upper- and lower-case letters above the means indicate the effect of soil moisture content and defoliation, respectively. The absence of letter labels above the means indicates non-significance. Means underneath different letters are significantly different.

Biomass

Shoot, root, and total biomass were significantly influenced by soil moisture content but not defoliation (Table 1). The pattern of response to this factor was similar to that exhibited by the morphological parameters (Figure 2). In contrast, root : shoot ratio was unresponsive to treatments either in isolation or combination. In fact, the combination of soil moisture content and defoliation had no significant effect on any of the biomass attributes (Table 1).



Figure 2: Effect of soil moisture content and defoliation on biomass. Values are mean \pm SE. See caption of Figure 1 for other explanations.

Leaf chlorophyll content

There was no significant main or interactive effect of treatments on leaf chlorophyll content (Table 1, Figure 3).



Figure 3: Effect of soil moisture content and defoliation on biomass. Values are mean ± SE. See caption of Figure 1 for other explanations.

IV. DISCUSSION

This study revealed that, high soil moisture content had a significant positive influence on height, leaf area and biomass accumulation. Tanwie and Ambebe (2024) also reported similar results with increments of soil moisture content on the growth of *Canarium swcheinfurthii*. Plants require water for various physiological processes such as photosynthesis, nutrient uptake, and cell expansion. When soil moisture content is low, plants are not able to take up enough water to support these processes, leading to reduced growth. In response to water shortage, plants may also close their stomata to reduce water loss through transpiration which limits the uptake of carbon dioxide photosynthesis. Additionally, low soil moisture content can stress plants and make them more susceptible to diseases, pests and other abiotic stress, further impacting growth (Gutezeite, 2006; Cavagnaro, 2016; Vassanthini and Premanandarajah, 2018; Chadha et al., 2019).

Defoliation can reduce the area available for the capture of radiation for photosynthetic carbon gain. However, the absence of a decline in biomass following defoliation may be explained by a sustained carbohydrate supply due to photosynthetic compensation. A typical metabolic response to defoliation is an increase in photosynthetic rate of the remaining leaves (Ambebe et al., 2020). However, the effects of defoliation can be attenuated by other biotic factors like sunlight, nutrient availability and soil moisture content (Su et al., 2023, Ambebe et al., 2024). This study was carried out in a greenhouse with controlled conditions. This explains why defoliation did not significantly influence growth parameters in this study as other resources were available and were directed towards re-growing leaves, recovering from the damage caused by defoliation and an ultimate increase in biomass accumulation. As such, number of new leaves did not differ among defoliation treatments. The lack of a soil moisture effect on root : shoot ratio suggests that the low moisture regime subjected the plants to just mild moisture stress.

The leaf chlorophyll content of *V. africana* was not significantly affected by soil moisture levels, defoliation or their interaction. This could be explained by the fact that, the amount of chlorophyll in leaf tissue depends on environmental factors such as drought stress, nutrient availability, heat change and change in light intensity (Palta,1990; Ambebe and Tanwie, 2020). Since the study site was under a controlled condition, these environmental factors were at their optimum and therefore leaf chlorophyll content of *V. africana* was not significantly affected.

V. CONCLUSION

Increasing soil moisture positively affected the growth and biomass accumulation of *V. africana*. Overall, growth did not decline under defoliation due to photosynthetic compensation. There was no significant interactive effect of treatments on any of the traits examined. Given that the stress imposed by the low moisture regime was mild, it is recommended that a further study be conducted with the much lower moisture levels that are characteristic of soils within species' ecological range to determine if the absence of a soil moisture content \times defoliation effect is sustained.

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