Samantha F. Vallesteros, et. al. International Journal of Engineering Research and Applications
www.ijera.com
ISSN: 2248-9622, Vol. 13, Issue 7, July 2023, pp 272-279

RESEARCH ARTICLE OPEN ACCESS

Relationship of ALIM (*Melanolepis multiglandulosa)* Tree Traits and its Chlorophyll Content to the Selected Vegetation Characteristics

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ABSTRACT
Alim (*Melanolepsis multiglandulosa*) is a small tree that is abundantly growing in a wide range of sites. To assess its potential as indicator species, leaf chlorophyll content was correlated with selected seedling and site variables. Chlorophyll content was taken in five fully expanded leaves of sample seedlings within 20 m x 20 m plots. Only a few significant correlations were observed between chlorophyll content and seedling traits. Height-Stem Diameter Ratio correlated strongly with chlorophyll content. A correlation between chlorophyll content and vegetation characteristics, and among vegetation characteristics was observed. Only a few relationships came out to be significant having coefficient values of -0.5016 to 0.8970. Among the site characteristics, canopy closure and crown depth were strongly and very strongly correlated with chlorophyll content. The study showed potential for chlorophyll content of alim seedlings as an alternative to the actual assessment of forest understory condition that is affected by canopy closure and canopy depth.
***Keywords* -** canopy closure, canopy depth, chlorophyll content, forest understory, vegetation

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Date of Submission: 13-07-2023 Date of acceptance: 28-07-2023

I. INTRODUCTION

The scientific interest in understanding the structure and function of tree-dominated vegetation, such as brushland, riparian vegetation, plantation, and forest, has been increasing due to its role in the sequestration and storage of carbon dioxide. The forest and vegetation of a specific ecosystem are generally influenced by external factors, particularly man-made development activities [1].

Scientists and researchers are interested in vegetation growth and health that can be assessed using some plant indicators. In Nueva Vizcaya, one potentially useful plant indicator is Alim (*Melanolepis multiglandulosa*) because it is widely distributed in the province through the heights and types of vegetation.

Alim is a tropical tree species that abundantly grows in a wide range of sites but prefers grassland edges, brushland, and open-canopy secondary forests. In secondary forests, alim is one of the most abundant small tree species occupying the bottom and middle canopy layers. Likewise, its seedlings and saplings are significant components of the understory vegetation.

Alim is a pioneer species, often found in open areas but can also tolerate low sunlight environments. Mother trees are prolifically seeding yearly. Likewise, seeds germinate well, and seedling survival is high. The plant is tolerant to a strenuous site, but its appearance somewhat reflects the site's condition. Plants in the natural community should optimize light absorption when considering the importance of chlorophyll content for photosynthesis. Such qualifications may be used to assess certain vegetation characteristics to which the plant corresponds [2].

If there is one easily measurable and observable part of the plant, which can be used to characterize the plant itself or its immediate surroundings, it must be the leaves. Leaves are sensitive to site conditions. They exhibit differences in size, shape, color, and vigor. Leaf chlorophyll content may also vary with leaf appearance, vigor, and the plant environment. Leaf color and chlorophyll content are essential factors in plant growth and contribute significantly to plant appearance.

With the advent of portable equipment to measure chlorophyll content in leaves, scientists and researchers are searching for the use of chlorophyll content as an indicator of plant health, vigor, and yield. In this research, the chlorophyll content of Alim chlorophyll will correlate with certain vegetation traits in the hope of finding beneficial relationships that can be used to characterize vegetation [3].

The study may discover that alim leaf chlorophyll content responds to the immediate environment, one important component of which is the vegetation to which it belongs. This response would allow a more accessible assessment of vegetation characteristics. This response will be established by exploring relationships between leaf chlorophyll content and several easily measurable vegetation traits.

The information produced by the study has potential value for research and land management. Researchers can do verification studies and replicate the methodology or part of it for the same species, related species, or different species. On the other hand, land managers interested in natural relationships of different land or vegetation components stand to benefit from the results of the study by way of avoiding repeated measurements of plot variables as indicator plants, potentially alim, in this case, can give information at an acceptable level of confidence. In management language, this translates to resource-saving.

This study aimed to answer the following questions:

1. Will alim condition as an understory plant be described in height, stem diameter, leaf size, and density?
2. Will the seedling vigor score be derived based on selected seedling characteristics?
3. What is the correlation between chlorophyll content and seedling traits?
4. How does chlorophyll content correlate with vegetation characteristics?

II. METHODOLOGY

This study required forestry equipment and devices such as a chlorophyll content meter, Global Positioning System (GPS) receiver, light meter, densitometer, diameter tape, measuring tape, range finder, digital vernier caliper, height pole, and camera. Other materials are a meter stick, laptop, USB flash drive, clipboard, datasheets, bolo, plastic envelope, pencil, marker, and ballpen

The study was performed in a section of Bangan Hill, specifically the southern section, which faces the NVSU Campus (Figure 1). The said portion of Bangan Hill has relatively thick vegetation, but many small canopy gaps are nurturing alim growth. The study area is around 10 hectares (ha). The site is experiencing an annual rainfall of around 2,100 mm based on 10-year data collected by the NVSU-PAGASA Agromet Station [3], located 450 m straight distance from the study area. Based on the long-term 1950 to 2000 data published by the World Climate Data collected [4], the average temperature in the locality is 24.63°.

Elevation was observed in Google Earth Pro and found to range from 285 to 370 m above mean sea level (Figure 2). The site is inclined, and the slope ranges from 20 to 35% (Figure 3).

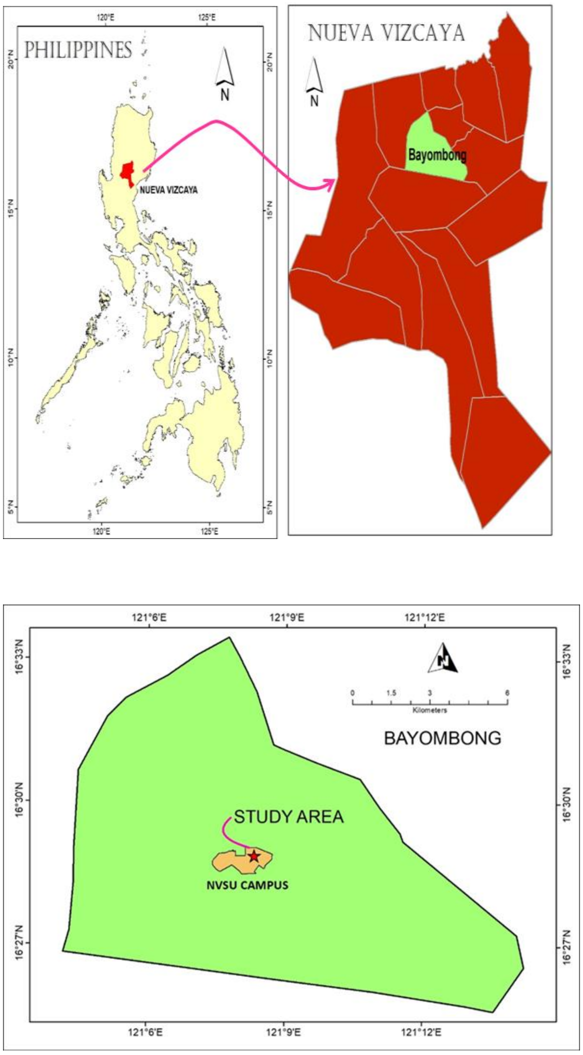

Figure 1. Location of the study area

Figure 2. Google Earth clip showing a planimetric view of the study area and two elevation points, lowest and highest.

Figure 3. Tilted view of Bangan Hill depicting the study area's slope. Elevation exaggeration is 1.5. The illustration highlights the eastern slopes.

Procedures

Sample plots were established within an area of approximately 10 ha of land. Plot locations were determined randomly using ArcMap's "Create Random Points tool" (Figure 4). ArcMap is a geographic information system (GIS) software part of ArcGIS. The number of plots shall depend on the available budget and resources of the study. The study established 15 plots. This number is deemed sufficient and attainable given the limitation on equipment availability.

Data were collected within 20 m x 20 m sample plots. Plots were oriented where the side follows cardinal compass directions, i.e., north-south and east-west. Plots were numbered, and their XY coordinates were in the bottom-left corner of the plot. Plot locations were found using a GPS receiver.

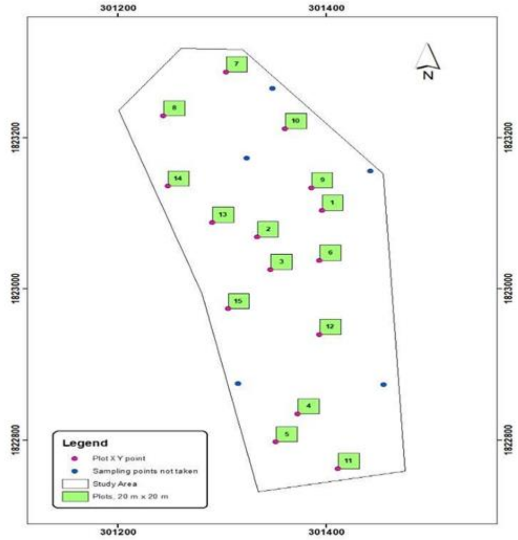

Figure 4. Distribution of sampling plots within the 10-ha study area. There were five plots that were not included.

The following data were collected:

1. Plot coordinates – latitude and longitude in decimal degrees at five decimal places precision. Plot coordinates were used for identifying the plots and organizing data, and not a variable of the study.
2. Leaf chlorophyll content – leaf chlorophyll content was obtained using CCM-200 Opti-Sciences chlorophyll content meter. CCM-200 provides relative chlorophyll concentration.
3. Number of alim seedlings and saplings – count of alim seedlings and saplings within the plot. Sizes were designated for the purpose of the study: seedlings are those young plants not less than 30 cm tall and not more than 2 cm stem diameter; saplings were bigger than seedlings with diameter between 2 cm and 10 cm.
4. Stem diameter – diameter of the stem of seedlings taken at 5 cm point from the ground. The stem diameter unit of measure was centimeters.
5. Number of first-order branches – number of first-order branches of sapling alim trees. The first-order branch is a branch that emanates from the main stem.
6. Basal area – the sum of basal areas of all trees 10 cm in diameter and up from the 20 m x 20 m plot. Basal area was calculated by multiplying 0.7854 and DBH squared and expressed in square meters.
7. Tree total height – the distance between root collar and the tip of the bud of the main stem for seedlings and saplings where said tip can be accessed. For large and tall trees, the upper limit of total height shall be the topmost portion of the

crown.

8. Number of trees per plot – total number of trees with a minimum diameter of 10 cm, including the alim trees (see No. 3 data).
9. Canopy closure – The proportion of the sky hemisphere obscured by vegetation when viewed from a single point (Jennings et. al., 1999) The canopy closure data was taken from 10 randomly selected points within the plot using a densitometer.
10. Light intensity – the amount of sunlight at a particular time of day, or in this study, between 9 AM and 10 AM. It was obtained using a light meter, which made light intensity in lux.
11. Ground cover – Obtained from a 4 m x 4 m subplot that was located at the center of the plot. Ground cover refers to the percentage of the ground covered by a plant not more than 1 meter in height.
12. Time of observation – Light intensity and chlorophyll content observation were recorded and expressed in hour-minute format.

Data Collection

Before data collection, datasheets were prepared (Appendix 1). After each plot observation, data were backed up by taking photographs of the filled-out datasheets.

All alim individuals under 30 cm tall to 5 cm stem diameter were observed and counted. Height was determined using a meter stick, stem diameter by vernier caliper or diameter tape, and density by counting the number of individuals per plot.

Within the 20 m x 20 m plot, tree seedlings or saplings were randomly selected for leaf chlorophyll content observation. Five fully expanded leaves were selected per plant as there were many leaves. Leaves must be those positioned 3 to 4 leaves from the youngest one in the main stem or branches. Leaves must not be yellowing.

For each leaf, only one chlorophyll content reading was observed, excluding the midrib, and the sensor is entirely on the blade (no space such as on the edge or a hole in the blade). The mean chlorophyll content per plant and the mean chlorophyll content per plot were computed.

Seedling vigor scores were derived from the following variables: height-stem diameter ratio, internode length, leaf width, leaf length, petiole length, leaf color, and leaf shape.

On the other hand, site characteristics were observed with equipment such as diameter tape for basal area, range finder for all height-related variables, densitometer for canopy closure, light meter for light intensity, and measuring tape for ground cover percentage.

Ground cover percentages were observed in a 4 m x 4 m subplot. Observation of light intensity in all plots should be within just one hour, between 11 AM and 12 NN. If the observation was not completed within the said time, the observation was done on other days, provided that the weather was more or less the same. Data collection occurred within one week only. Weather information was obtained from a particular website. Light measurement was not done when the sky was too cloudy, gloomy, or raining.

Data was encoded in Microsoft Excel, and various computations were performed there, including correlation analysis. Another software, the Statistical Tool for Agricultural Research (STAR), was used to get the p-values of the Pearson correlation and to plot scatter diagrams.

Experimental Design

The exact locations of the plots were determined. A minimum distance of 30 meters between plots was imposed in drawing samples. Twenty samples were drawn so that replacement plots that fall in non-vegetation surfaces such as buildings, pavement, and roads can be replaced readily.

As mentioned earlier, the plot dimension is 20 m x 20 m with a 4 m x 4 m subplot nested in this bigger plot for the ground cover variable. The basis of tree inclusion was the position of the stem in the plot. Trees at the border having more than 50% of their basal part within the plot were counted. However, in terms of crown; crown or crown portion within the plot was included.

Detailed procedures for measuring or observing each variable were to maximize the efficiency and accuracy of data.

Statistical Treatment of Data

The research used three datasheets (Appendix 1) to facilitate data recording in the field. Correlation analysis was used to analyze relationships between variables. There were 27 variables whose relationships between one and the other were analyzed using the Pearson correlation tool.

Microsoft Excel's Data Analysis Tool was used to run the correlation. The level of confidence was set to 95%. A scoring tool was provided to quantify the seedling vigor score. (Table 1).

Table 1. Scoring Tool for Vigor Score

Internode Length		Leaf Width		Leaf Length	
Less than 1	2	16 or less	2	14 or less	2
1 - 1.5	4	16.1 - 19	4	14.1 - 17	4
1.6 - 2	6	19.1 - 23	6	17.1 - 21	6
2 - 2.5	8	23.1 - 26	8	21.1 - 25	8
2.6 to 3	10	More than 26	10	More than 25	10

Petiole Length		Leaf color		Leaf Shape	
10 or less	2	Yellowish green	2.5	All shallow-lobed	2.5
10.1 - 14	4	Pale Green	5	Two shallow-lobed	5
14.1 - 18	6	Dark Green	7.5	Two deep-lobed	7.5
18.1 - 22	8	Green	10	All deep-lobed	10
More than 22	10				

III. RESULTS AND DISCUSSION

Seedling Traits

Alim is one of the most widely distributed plants in the Philippines and other tropical countries [4]. It can be seen in open areas, building surroundings, roadsides, brushlands, and openings of closed-canopy forests. However, its presence under a heavily shaded understory of thick forest vegetation is low or may be absent. This study looked into the condition of alim seedlings in understory conditions. As counting newly germinated seedlings within the 20 m x 20 m plot is complex, the analysis defined “seedlings” as those with sizes not less than 30 cm tall and not more than 2 cm stem diameter.

Seedling stem diameter ranged from 0.59 cm to 1.35 cm within the study area. In terms of height, the minimum was 35.76 cm, while the maximum was 122.33 cm. The researchers noted the presence of many smaller seedlings within the plot. The means were 0.9 cm and 88.35 cm for stem diameter and height.

Other easily measurable characteristics were also recorded: the number of first-order branches, internode length, leaf width, leaf length, and petiole length. Small seedlings tend to have no branches most of the time. Seedlings with first-order branches were observed in 6 out of 15 plots. As expected, taller seedlings are likely to possess first-order branches.

Leaf size variables such as petiole length, leaf width (or precisely the longest width of the leaf blade), and leaf length are easily measured using a ruler; hence, they were included in this study. The mean, minimum, and maximum values are shown in Table 2. The variabilities in petiole length and leaf size were relatively low, while the internode tended to be high.

Table 2. Statistics of the Leaf Variables

Variable	Mean	SD	CV	Minimum	Maximum
Internode length (cm)	1.94	0.56	28.88	0.50	2.67
Petiole length (cm)	20.43	2.66	13.03	14.67	24.67
Leaf width (cm)	19.34	2.91	15.06	15.00	24.23
Leaf length (cm)	18.60	2.16	11.60	14.83	22.45

Based on the count of seedlings and saplings per 400 m2 plot, the study area contains around 661.67 seedlings per hectare and 351.67 saplings per hectare. The highest counts observed were 74 and 56, translating to 1,850 seedlings and 1,400 saplings per hectare. The number of regenerations can be much more significant if the smaller individuals were included, i.e., seedlings less than 30 cm tall were counted. It is a general situation in naturally growing tropical forest vegetation that is not in the climax stage that the number of smaller trees per unit area is higher than that of the larger trees [6].

Seedling Vigor Score

As the study intended to explore whether alim can be used as an indicator of understory vegetation characteristics, several seedling traits were combined to derive a vigor score. The scoring basis was a dataset containing observations from 100 seedlings within the study area but outside the sampling plots. Scores were based on height-stem diameter ratio, internode length, leaf width, leaf length, petiole length, leaf color, and leaf shape. The healthiest individuals get high scores with a maximum value of 100.

Scores per plot range from 52.23 to 83.23, while the mean was 70.78. The standard deviation was 8.96, while the coefficient of variation was 12.66.

It was observed that the plots represent sites that support the tremendous growth of understory vegetation consisting of various species of herbaceous plants and grasses. The amount of sunlight that reaches the forest floor is moderate between 11 AM and 12 NN at an average of 860 lux.

Seedling vigor score and chlorophyll content exhibited weak to moderate negative correlations (-0.2238 to -0.5898). The result contradicted the assumption that seedling vigor score was positively correlated with chlorophyll content. However, seedling height and leaf size might have significantly influenced the vigor score. Shaded seedlings tend to be taller than those exposed to sunlight and tend to have larger to compensate for a smaller amount of sunlight received per unit area of the leaf [7].

Correlation between Chlorophyll Content and Seedling Traits

Chlorophyll content, expressed as chlorophyll index as measured by the chlorophyll content meter, is a good measure of seedling health [8] [9]. The relative chlorophyll units of the CL-01 chlorophyll Content Meter linearly and positively correlate to actual leaf chlorophyll content in mg/g/FM-1 in many species. The measurement of chlorophyll content can yield important information relating to biotic stress factors and abiotic issues such as light and moisture limitation [10].

The chlorophyll index was read from five leaves starting from the 3rd or 4th leaf from the top, which is fully expanded already. Average values were taken per plot. Across plots, the highest mean chlorophyll index was recorded for Leaf 3 and the lowest in Leaf 5 (Figure 15).

Results of the correlation analysis show a very high positive correlation among leaves within a plant. Pearson correlation coefficients ranged from 0.5200 to 0.9328 (Appendix 2). The lowest correlation was between Leaf 1 and Leaf 5, while the highest was between Leaf 3 and Leaf 4.

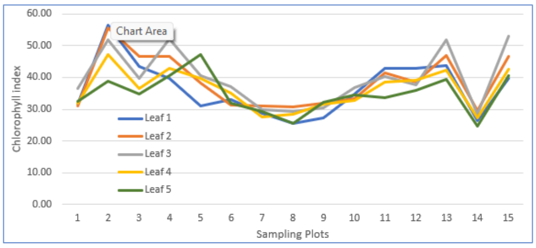

Figure 15. Variability of leaf chlorophyll content in five leaves across sampling plots

Overall, only a few significant correlations were observed between chlorophyll content and seedling traits (Table 3).

Measurement of height and stem diameter is the most common morphological assessment of seedling quality [11]. Height-Stem Diameter Ratio appeared to be a better measure of the seedling trait. This variable correlated strongly with all chlorophyll content observations (all five leaves and their average), with correlation coefficients ranging from -0.6710 to -0.8173. A negative correlation means that the height-stem diameter ratio decreases as chlorophyll content increases. The lower value of height-stem diameter indicates stouter seedlings which may exhibit higher photosynthetic activity of the seedlings, influenced by the amount of sunlight received [11]. Under shade conditions, in general, seedlings have increased stem height, petiole length, and decreased stem diameter, as more carbon is allocated to elongate the stem and petiole than stem diameter growth [12].

Using the mean chlorophyll content (CC-All), height and chlorophyll content exhibited a moderate negative correlation. On the other hand, stem diameter showed a strong positive correlation with chlorophyll content (Table 3).

Table 3. Correlation Coefficients Between Chlorophyll Content and Seedling Trait

Seedling Traits	*Correlation coefficients with chlorophyll content*					
	CC-All	CC-L1	CC-L2	CC-L3	CC-L4	CC-L5
Stem Diameter (SD)	0.6092	0.4441	0.4684	0.5257	0.7046	0.7616
Height (HT)	-0.4594	-0.5828	-0.5650	-0.4000	-0.3909	-0.0691
Height-Stem Diam. Ratio (HSD)	-0.7914	-0.7185	-0.7582	-0.6908	-0.8173	-0.6710
No. of First Order Branches (FOB)	-0.1577	-0.2047	-0.2787	-0.1827	-0.0909	0.1205
Internode length (IL)	-0.0908	-0.0910	-0.0757	-0.0776	-0.0803	-0.0965
Leaf width (LW)	0.1170	0.0643	0.0534	0.0230	0.1763	0.2980
Leaf length (LL)	0.1206	0.1034	0.0711	-0.0123	0.1684	0.2983
Petiole length (PL)	0.1792	0.2281	0.1868	0.0975	0.1978	0.1102
Leaf color (LC)	-0.4361	-0.4357	-0.4985	-0.4669	-0.4256	-0.1018
Leaf shape (LS)	-0.1008	-0.1166	-0.1964	-0.0645	-0.1458	0.0965

The number of first-order branches correlated very weakly with all chlorophyll content variables. The majority of the seedlings have not formed branches. The same case is true for leaf width, length, and shape. Leaf color is negatively and moderately correlated with the chlorophyll content of the leaves except for the oldest one. Leaves that are yellowish-green and pale green have lower chlorophyll content.

Correlation between Chlorophyll Content and Vegetation Characteristics

The selected vegetation characteristics were recorded in sampling plots. These are total basal area, canopy closure, tree height, crown depth, light intensity under the canopy, and ground cover percentage. The result of the correlation analysis between these variables and the chlorophyll contents variables is presented in Table 4.

Table 4. Correlation Coefficients Between Chlorophyll Content and Vegetation Characteristics

Vegetation characteristics	Correlation coefficients with chlorophyll content					
	CC-All	CC-L1	CC-L2	CC-L3	CC-L4	CC-L5
No. of Alim Seedlings (NAS)	0.4445	0.2308	0.3160	0.5835	0.5196	0.4265
No. of Alim Saplings (NAP)	-0.4568	-0.3828	-0.4638	-0.4179	-0.5016	-0.3381
Diameter at Breast Height (DBH)	-0.1105	-0.1338	-0.0389	-0.1166	-0.0776	-0.1460
Basal Area (BA)	-0.1335	-0.2364	-0.0892	-0.1415	-0.0693	-0.0361
Tree Height (TH)	0.2986	0.3630	0.3726	0.2485	0.2793	0.0509
Canopy Closure (CCL)	-0.7693	-0.6820	-0.7154	-0.7690	-0.7551	-0.6042
Ground Cover (GC)	-0.0603	0.0167	-0.0450	-0.1780	-0.0666	0.0167
Crown Depth (CD)	0.8970	0.8266	0.8687	0.8425	0.8395	0.7333

Only a few relationships came out to be significant, i.e., starting from coefficient values of -0.5016 (p = 0.05) to 0.8970 (p = 0.0001). Among the site characteristics, the canopy closure and crown depth are strongly correlated with chlorophyll content, the highest being the CC-All, followed by CC-L2. It is implied that the chlorophyll content of sample alim leaves, preferably Leaf 2, can be used as a proxy to crown depth measurement. High crown depth tends to block sunlight or reduce light intensity. There is consistency with the canopy closure-chlorophyll content relationship. The correlation coefficients are negatively signed as a higher value of canopy closure means lower sunlight, the same effect with a large value of crown depth. Lower sunlight induces higher chlorophyll content in leaves as seedlings tend to increase chlorophyll content as an adaptation to low light intensity.

High canopy closure appeared to decrease the number of alim seedlings as the correlation coefficients are positively signed (-0.3818, p = 0.1602), although the coefficient is insignificant. In contrast, the number of saplings increases with increasing crown closure (0.4101, p =0.1290).

Surprisingly, there was a very weak correlation coefficient between canopy closure and the two light intensity observations, one over the alim seedlings and another combination of alim seedlings and randomly selected points. There may be a need to improve the randomization of the light-intensity sampling points.

The number of alim seedlings was moderately correlated (-0.5002, p =0.050) with ground cover percentage but very weekly to weakly correlated with other vegetation characteristics variables.

Other Important Relationships

Table 5. Significant Correlation Coefficients between two variables

Variables	Correlation Coefficients	P-value
Stem Diameter and Leaf Width	0.6949	0.004
Stem Diameter and Leaf Length	0.7130	0.0020
Petiole Length and Leaf Width	0.6246	0.0100
Petiole Length and Leaf Length	0.5284	0.0430
Canopy Closure and Internode Length	0.5248	0.040
Ground Cover Percentage and No. of Alim Seedlings	-0.5002	0.050

Presented in Table 5 are other significant correlation coefficients between variables. Stem diameter needs to be proportional to leaf size, as shown by the significant positive correlation between stem diameter and leaf width and between stem diameter and leaf length. The same relationship goes for petiole length and leaf shape.

Canopy closure tends to lengthen the internode length as the seedling needs to elongate its stem to position the leaves better to capture more sunlight. Higher ground cover was associated with a smaller number of seedlings. Higher ground cover means larger competition among the undergrowth plants.

All 15 plots selected randomly contained alim seedlings suggesting the species' wide distribution within the study area. Based on the count of seedlings and saplings per 400 m2 plot, the study area contains around 661.67 seedlings per hectare and 351.67 saplings per hectare. The variabilities in petiole length and leaf size were relatively low, while the internode tended to be high.

Seedling vigor scores were derived following the assumption that a score or index was to correlate significantly with leaf chlorophyll content. However, seedling vigor score and chlorophyll content exhibited weak to moderate negative correlations.

Overall, only a few significant correlations were observed between chlorophyll content and seedling traits. Height-Stem Diameter Ratio appeared to be a better measure of a seedling trait. This variable correlated strongly with chlorophyll content.

A correlation between chlorophyll content and vegetation characteristics and among vegetation characteristics was observed. Only a few relationships were significant, having coefficient values of -0.5016 to 0.8970.

IV. CONCLUSION

Among the site characteristics, it is the canopy closure and crown depth that were strongly and very strongly correlated with chlorophyll content.

The study showed the potential for chlorophyll content of alim seedlings as a proxy to the actual assessment of forest understory conditions dependent on canopy closure and crown depth.

V. RECOMMENDATIONS

1. Consider performing a similar study but instead of plotting data, use hundreds of point data. Correlate chlorophyll content with many site variables.
2. A survey of the chlorophyll content of alim across ecosystems such as urban, rural, agroecosystem, grassland, brushland, and forest is interesting.
3. Develop inversion regression models where

chlorophyll content and other seedling characteristics are the independent variables and canopy closure, or crown depth is the dependent variable.

Acknowledgments

The researchers would like to acknowledge their utmost gratitude and appreciation to the people who instructed, upheld, and directed them throughout the length of this project. Their contributions and assistance are heartily acknowledged and truly appreciated.

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