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Original Article

Light intensity effect on seed emergence and seedling growth of African walnut (*Plukenetia conophora* Mull Arg.)

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ABSTRACT

Plukenetia conophora is a multipurpose climbing vine of immense benefits but its light requirements for seed germination and seedling growth remain unknown. Hence, the need to determine the optimum light requirement for seed germination and growth of *P. conophora.* Based on this, 50 seeds were sown in light and dark conditions. Furthermore, 100 uniform seedlings were subjected to 25%, 50%, 75%, and 100% light intensities in a completely randomized design in five replicates. Data on emergence percentage were collected. Number of leaves, collar diameter, length of vine, and leaf area were assessed fortnightly for 12 weeks. Relative growth rate (RGR), absolute growth rate (AGR), and net assimilation rate (NAR) were also determined. The result showed that seeds kept under light condition had a percentage emergence of 87.65% at a mean day of 22 while seeds kept under dark condition gave a percentage emergence of 65.25% at a mean day of emergence of 29 days. The effect of light intensity on seedlings growth of *P. conophora* was significantly different ($P < 0.05$). The 25% light intensity gave the highest number of 11.699, collar diameter of 3.166 mm, vine length 135.332 cm, leaf area of 70.960 cm^2, leaf dry weight of 2.319 g, stem dry weight of 3.978 g, and root weight of 20.393 g. The 25% light intensity also gave the highest RGR of 4.14×10^{-2} g/week between the 2nd and 4th week, AGR of 7.87×10^{-1} g/week 2nd–4th weeks, and the overall highest NAR of 8.95×10^{-3} g/week/cm^2 between 10th and 12th weeks. Therefore, light conditions are recommended for optimum germination of *P. conophora* seeds whereas 25% light intensity is recommended seedlings growth and development.

Keywords: *Plukenetia conophora*, light intensities, dark condition, growth rate, emergence

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INTRODUCTION

Plukenetia conophora is a climbing vine belonging to the family *Euphorbiaceae.* It is a wild woody perennial climber whose length ranges between 12 and 30 m.[1] The plant is noted to have originated from Sierra Leone from where it spread to other parts of West Africa. They produce edible seeds which are traded at premium prices. The seeds of *P. conophora* are recalcitrant and they disappear almost immediately after the fruiting season due to scarcity as the seeds are highly competed for by man and animal leaving behind little or nothing for planting.

Germination and establishment of *P. conophora* under natural condition is often hampered by human activities, animal and environmental factors which make its large-scale production difficult.[2] On the other hand, plantations of *P. conophora* are not easily established due to inadequate knowledge about its basic requirements for seedling growth and development.

One of such basic requirements for plant growth and survival is light. Light is a primary source of energy and is one of the most important environmental factors that determine plant growth and development.[3,4] Light in terms of intensity and quality is essential for every aspect of plant growth and development. Light is essential for seed germination, plant growth, flowering, and fruit initiation.[4-7] Light is noted to play a major role in photosynthetic process in making natural food for the trees[8] as well as morphogenesis and physiological responses.[9,10]

Light also plays critical role in seed germination as seeds are mostly sensitive to light to initiate germination. Seed germination is often the most sensitive stage in the life cycle

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of a plant. Seed germination can be defined as the growth of the embryo of the mature seed.[11] Seed germination is an irreversible process that results in the emergence of radical and plumule. According to Fenner *et al.*,[12] the effect of light can result in either the death or survival of the embryo. Seeds are categorized based on their sensitivity to light in terms of germination, those that require light to germinate, and those that germinate in the dark. Each category provides the emerged seedlings with a definitive survival strategy.[6] For example, seeds that require light to germinate are termed positive photoblastic while the negative photoblastic is those that require darkness to germinate and neutral photoblastic is those neutral to light.[13]

To adjust to different light conditions in the environment, plants have to developed many mechanisms at various levels during the development of plant.[14] When light is not sufficient, plants usually enhance their light interception, allocate more biomass to leaves, and have a higher leaf area per unit leaf mass. This will help to maximize the limited light absorption for the purpose of food production.[15] In the same vein, when there is high intensity, there will be reduction in specific leaf are and increase leaf thickness to protect the plant from the effect of such exposure. This adjustment in response of plant to high radiation will help the plant to modulate the damage effect of the light.[16]

Tolerance of plant to low or high light varies among species.[17-19] The capacity of trees to grow and survive in shade or to respond to high light levels is based on different strategies of allocating biomass.[20-22] This can be assessed by the various morphological changes the plants expressed. By assessing these morphological responses to different light levels, information on the shade or light tolerance status can be generated and be used in conservation strategy of the plant. According to Kobe *et al.*,[23] the quantitative understanding of seedling response to light is the key to characterize the plant as light or shade tolerance species.

Since plant species vary in their response to light, it is important to determine the optimum conditions of light or dark necessary for seed germination and seedling establishment under varying light conditions. The study was, therefore, conducted to determine the effect of light on seed emergence and seedling growth of *P. conophora* with the aim of producing vigorous seedlings for regeneration purpose and also categorizing the plant based on its light tolerance.

MATERIALS AND METHODS

Sources of Seed Collection and Extraction

The fresh and mature seeds of *P. conophora* were collected from high yielding mother vines during the fruiting period of 2018 at Ibadan in Oyo State of Nigeria. Seeds were collected from all the four quarters of each tree in their natural population.

The collected fruits were extracted manually with the use of kitchen knife to obtain the seeds. Damaged and deformed seeds which are mostly the floated seeds when immersed in water were discarded while the good seeds that sink in water are retained for the experiment.

Experimental Site

The experiment was conducted at Tree Physiology and breeding nursery of sustainable forest management department of Forestry Research Institute of Nigeria, Ibadan (FRIN). FRIN is located on the longitude 070 23'18''N to 070 23'43''N and latitude 03051'. The rainfall distribution pattern is bimodal with peak in June and July. The dry season occurs between November and March while the wet season is usually between April and September. The mean minimum and maximum temperatures are 19.5°C and 34.9°C while relative humidity was between 50% and 86.7%.

Effect of Light on Seed Germination

This study was conducted at the seed store of forestry Research Institute of Nigeria. The extracted seeds were subjected to two different light regimes of light and dark. The light regime was made up of daylight supplemented by four 60 w fluorescent tubes, while the dark was made up of glass funnels wrapped with a double layer of black polythene sheet. Fifty seeds were placed in each of the light regime. The seeds were placed on Whatman No. 9 filter papers and placed on transparent glass sheets inside two Copenhagen germination tanks at the seed store laboratory of FRIN. The filter paper was moistened with 10 ml of distilled water to keep it moist. These were observed daily for seed emergence. Seed emergence is said to have taken place when radicle and plumule emerged. Seed emergence was recorded daily until no further emergence occurs.

Percentage seed emergence was determined according to the formula below:

$$\text{Germination Emergence } (\%) = \frac{\text{Number of seeds emeerged}}{\text{Total number os seeds sown}} \times 100$$

Effect of Light Intensity on Seedling Growth and Development

A total of 200 seeds were sown directly at one seed per pot into 2 kg capacity polythene pots filled forest top soil obtained at the FRIN arboretum.

Light screening chambers with one layer, two layers, and three layers of 1 mm green plastic mesh net were constructed. The

one, two, or three layers were used to achieve varying levels of light reduction according to treatments used (Figure 1).

- T1 = Seedlings placed in the open which receives 100% sunlight
- T2 = Seedlings in frames covered with one layer of mesh net received 75%
- T3 = Seedlings in frames covered with two layers of mesh net received 50%
- T4 = Seedlings in frames covered with three layer of mesh net received 25% as used by various researchers to achieve varying light intensities.[24,25]

The light intensities within and outside the cages were monitored using light meter (Digital Lux Meter Model LX-101, Taiwan) (Figure 2).

A total of 100 seedlings (100) at four leaf stage was selected from the germinated seedlings for this experiment out of which 10 seedlings each were placed under each of the four varying light intensity. Additional 15 seedlings were added to each of the varying light intensities to cater for biomass assessment as well as mortality. The parameter assessed includes: Seedling (vine) height (cm) by the use of meter rule, collar diameter (mm) by the use of digital Vernier caliper, number of leaves by physical counting, and leaf areas (cm^2) were measured using portable leaf area meter model YMJ-B 1120578 fortnightly for 12 weeks. Leaf (g), stem (g), and root biomass (g) were determined destructive fortnightly for 12 weeks after transplanting. Data collected on dry weights and leaf areas were used to calculate relative growth rate (RGR), absolute growth rate (AGR), and net assimilation rate (NAR).

Biomass of seedlings was estimated destructively by separating the seedlings into root, stem, and leaves. The fresh weights were taken before drying. The seedlings are oven dried for 60°C for 72 h until a constant weight was achieved, this served as the dry weight.

RGR of seedlings was calculated using equation:

$$\text{RGR}\ \left(\text{g / week}\right) = \frac{(\text{lnTB2} - \text{TB1})}{(\text{t2} - \text{t1})}$$

Where, lnTB1 and lnTB2 are natural log of initial and final total biomass and t1 and t2 are initial and final time period, respectively

$$\text{AGR}\ \left(\text{g / week}\right) = \frac{(\text{W2} - \text{W1})}{(\text{t2} - \text{t1})}$$

Where, W2 and W1 are initial and final dry weight and t2 and t1 are initial and final time

Figure 1: The constructed cages made of aluminum covered on all sides with one (right), two (left), and three layers (middle)

Figure 2: Light meter (Digital Lux Meter Model LX-101, Taiwan) used in monitoring the light intensity within and outside the cage

$$\text{NAR}\ \left(\text{g / week / cm} = \frac{\text{AGR}}{(\text{LA2} - \text{LA1})}\right)$$

Where, LA2 and LA1 are final and initial leaf, respectively.

Experiment Design and Analysis

The experiment was laid out in completely randomized design with five replicates. The data collected were subjected to descriptive statistics as well as analysis of variance (ANOVA) and significant means were separated using least significant difference (LSD) at 5% level of probability. Results were presented in tables.

RESULTS

Effect of Light and Dark Condition on Seed Emergence of *P. conophora*

The result from the effect of light on seed emergence of *P. conophora* seeds shows that seeds kept under light condition had a percentage emergence of 87.65% at a mean day of 22 while seeds kept under dark condition gave a percentage emergence of 65.25% at a mean day of emergence of 29 days.

Effect of Varying Light Intensities on Growth of *P. conophora*

The effect of light intensities on the growth of *P. conophora* was significant ($P < 0.05$) for all the growth traits observed (Table 1). Variations were observed from all the various light

intensities used. The highest mean value of 11.699 leaves was obtained from the seedlings of walnut exposed to minimum sunlight of 25% (Table 2) which was followed by the seedlings exposed to 50% sunlight with mean value of 11.087 leaves. The least number of leaves was observed when the young seedlings of walnut were exposed to total (100%) sunlight.

In terms of the collar diameter growth of walnut, it was observed that exposing the young plant to 25% of the total sunlight resulted in highest collar diameter of 3.166 mm while exposing it to total sunlight (100%) resulted in smallest girth with mean value of 0.911 mm. Furthermore, exposing the young seedling to 50% and 75% sunlight produced the same effect of collar diameter which was not significantly different from each other.

The vine length decreased with increased in light intensities as observed. The least exposed vine produced the tallest vine of 135.332 cm while the highest exposed (100% sunlight) produced the shortest vine of 118.420 cm.

The 25% light intensity exposure had the highest leaf area with mean value of 70.960 cm^2. Next to this is exposing the young plant to 50% and 100% light intensities produced the same effect which was not significantly different from each other while the 75% exposure resulted in the least leaf area.

The effect of light intensities on the leaf dry matter of *P. conophora* was significant as the least light intensity of 25% took the lead by producing mean leaf dry weight of 2.319 g next is the 50% exposure which produced a mean leaf dry weight of 2.022 g. The least mean dry weight of 1.836 g was observed among seedlings of *P. conophora* exposed to 75% light intensities.

Table 1: Percentage emergence (%) of *Plukenetia conophora* seeds are affected by light and dark conditions

Emergence condition	Percentage emergence (%)	Mean days of emergence (days)
Light	87.65	22
Dark	65.25	29

The stem dry matter weight of *P. conophora* as affected by different light intensities followed a different pattern when compared to other traits. Total light intensity exposure resulted in the highest stem dry weight of 3.978g and 25% light exposure resulted in the second best stem dry weight. The least stem dry weight was observed in seedlings exposed to 50% light intensity.

In terms of root dry weight of *P conophora*, the 25% light exposure produced the highest root weight of 20.393 g which was not significantly different from 50% with mean weight of 19.915 g and 100% exposure which had a mean weight of 19.558 g.

Means with the same letters along a column are not significantly different from each other according to LSD at 5% level of probability

The RGR estimated in *P. conophora* as affected by varying light intensities shows that variations occur among the various light intensities used (Table 3). The highest RGR of 4.14 × 10^{-2} g/week was observed in seedling grown under 25% light intensities between the 2nd and 4th week of the experiment while the lowest of 5.96 × 10^{-3} g/week was observed in seedlings grown under 75% light intensities between the last phase of the experiment (10–12th weeks).

The absolute growth rate (AGR) estimated in gram per weeks in *P. conophora* seedlings as affected by the four different light intensities used showed that highest AGR of 7. 87 × 10^{-1} g/week was recorded among the seedlings exposed to 25% light intensities at the beginning of the experiment (2nd–4th weeks) followed closely by 50% at the same duration of the experiment (Table 4). The least AGR was observed in 100% light intensities at the 6th–8th week of the experiment.

The NAR observed in seedling growth of *P. conophora* recorded a steady increase across the experimental period in all the various light intensities. Among the various light intensities, the least NAR was observed in the 2nd–4th week while the highest was observed in the 10th–12th week of the experimental period. However, it is noteworthy to state that

Table 2: Mean value of the effects of varying light intensities on seedling growth in *Plukenetia conophora*

Light intensity	Number of leaves	Collar diameter (mm)	Vine length (cm)	Leaf area (cm^2)	Leaf dry weight (g)	Stem dry weight (g)	Root dry weight (g)
25% soil light	11.699[a]	3.166a	135.332[a]	70.960[a]	2.319[a]	3.532[b]	20.393[a]
50% soil light	11.087[ab]	0.954[b]	125.293[b]	61.418[b]	2.022[b]	3.014[d]	19.915[a]
75% soil light	10.547[c]	0.984[b]	122.140[c]	58.062[c]	1.836[d]	3.106c[c]	15.832[b]
100% soil light	9.699[d]	0.911[b]	118.420[d]	61.802[b]	1.925[c]	3.978[a]	19.558[a]
LSD	0.5	0.1	2.68	3.22	0.01	0.25	3.38

LSD: Least significant difference, The same alphabet are not significant different from each other according to LSD at 5% level of probability

Table 3: Variations in RGR (g/week) for the seedlings of *Plukenetia conophora* as affected by varying light intensities

Light intensities	RGR1	RGR2	RGR3	RGR4	RGR5
25% soil light	4.14×10^{-2}	2.24×10^{-2}	2.12×10^{-2}	2.62×10^{-2}	1.103×10^{-2}
50% soil light	3.62×10^{-2}	2.71×10^{-2}	1.55×10^{-2}	6.5×10^{-3}	7.57×10^{-3}
75% soil light	1.33×10^{-2}	1.78×10^{-2}	1.802×10^{-2}	1.51×10^{-2}	5.96×10^{-3}
100% soil light	2.79×10^{-2}	1.55×10^{-2}	7.5×10^{-3}	1.97×10^{-2}	9.57×10^{-3}

RGR: Relative growth rate, RGR1: RGR between the 2nd and 4th week, RGR2: RGR between the 4th and 6th week, RGR3: RGR between the 6th and 8th week, RGR4: RGR between the 8th and 10th week, RGR5: RGR between the 10th and 12th week

Table 4: AGR (g/week) for the seedlings of *Plukenetia conophora* as affected by varying light intensities

Light intensities	AGR 1	AGR2	AGR3	AGR4	AGR5
25% soil light	$7.\ 87\times10^{-1}$	7.4×10^{-1}	1.07×10^{-1}	1.39×10^{-1}	1.89×10^{-1}
50% soil light	$7.\ 75\times10^{-1}$	3.57×10^{-1}	1.22×10^{-1}	1.44×10^{-1}	1.513×10^{-1}
75% soil light	3.15×10^{-1}	4.9×10^{-1}	1.75×10^{-1}	1.605×10^{-1}	1.175×10^{-1}
100% soil light	4.065×10^{-1}	3.35×10^{-1}	1.045×10^{-1}	1.135×10^{-1}	1.46×10^{-1}

AGR: Absolute growth rate, AGR1: AGR between the 2nd and 4th week, AGR2: AGR between the 4th and 6th week, AGR3: AGR between the 6th and 8th week, AGR4: AGR between the 8th and 10th week, AGR5: AGR between the 10th and 12th week

Table 5: NAR (g/week/cm^2) for the seedlings of *Plukenetia conophora* as affected by varying light intensities

Light intensities	NAR1	NA2	NAR3	NAR4	NAR5
25% soil light	4.48×10^{-3}	6.16×10^{-3}	7.34×10^{-3}	8.45×10^{-3}	8.95×10^{-3}
50% soil light	3.29×10^{-3}	4.65×10^{-3}	6.903×10^{-3}	$7.\ 37\times10^{-3}$	8.461×10^{-3}
75% soil light	3.25×10^{-3}	4.71×10^{-3}	5.695×10^{-3}	$7.\ 77\times10^{-3}$	$8.\ 13\times10^{-3}$
100% soil light	3.44×10^{-3}	5.15×10^{-3}	6.160×10^{-3}	7.66×10^{-3}	8.27×10^{-3}

NAR: Net assimilation rate, NAR1: NAR between the 2nd and 4th weeks, NAR2: NAR between the 4th and 6th weeks, NAR3: NAR between the 6th and 8th weeks, NAR4: NAR between the 8th and 10th weeks, NAR5: NAR between the 10th and 12th weeks

25% light at the 10th–12th week had the overall highest NAR of 8.95×10^{-3} g/week/cm^2 while the least of 3.25×10^{-3} g/week/cm^2 is observed in Table 5.

DISCUSSION

The aim of this research was to find out the effect of light on seed emergence and seedling growth of *P. conophora* with the aim of producing vigorous seedlings for regeneration purpose and also categorize the plant based on its light tolerance.

The result obtained from percentage seed emergence of *P. conophora* kept in both dark and light conditions clearly indicated that the seed of *P. conophora* can be grown on both dark and light condition. However, the higher percentage of seed emergence of *P. conophora* seeds kept under light condition over those kept in low condition as well as reduction in the mean number of days it took the seeds kept in light conditions to emerge over those kept in dark condition is a pointer that light favors the emergence of *P. conophora* and can be said that the seeds requires light for maximum germination. This is also an indication that light speed up the germination process as it brought about increase in seed emergence in lesser days (Table 1). This is in line with the findings of Oyedeji *et al*.[26] who reported that higher percentage germination in *Dialium guineense* was recorded for the seeds exposed to light than those exposed to dark condition, although there was no significant difference between the germination percentages of those planted under both light and dark media.

Effect of Varying Light Intensities on Growth of *P. conophora*

The result indicated that the effect of light intensities on the seedling growth of *P. conophora* was significant ($P < 0.05$) for all the growth traits observed (Table 1). Many scholars had shown that plant growth is affected by light intensity (Poorter and Nagel[5], Williams[6], Naoya *et al*.,[3] Sevillano *et al.*,[27] Nguyen *et. al.,*[18] and Amir *et al.*,[4]).

In this study, number of leaves, collar diameter, vine length, leaf, as well as the dry matter (leave dry weight and root dry weight) were enhanced with 25% light intensities (Table 2). Lower light intensity of 25% also resulted in increased in leaf area as well as dry matter content of the plant as well as increase in RGR, AGR, and NAR. This is an indication that the young seedling of *P. conophora* requires minimum light for growth as exposing them to total or open sunlight resulted in retardation

of growth. This is the line of thought of Onyekwelu *et al*.[28] who investigated the effect of light intensities on *Chrysophyllum albidum* and reported that the overall best growth and most stable seedlings of were obtained under minimum light intensity of 40%. He further noted that the young seedling of *C. albidum* needs shading during their early growth because if such seedlings are exposed to 100% light intensity and open sky; they may die shortly after emergence.

The result from this study also supports the general idea that plants which grow in limited light condition respond to the light environment by investing more biomass in the leaves by producing larger and more leaves to enlarge the leaf surface for light interception (Ardalan *et al.*).[29] This, although, is contrary to the observations made by Poorter[7] who stated that shaded species allocate lower biomass to leaves for some of rain forests tree species. Lusk and Piper[30] stated that the shaded understory plants with different life strategies cope differently with light conditions. These findings clearly support the facts that tolerance of plant to low or high light varies among plants (Wayne and Bazzaz[17], Poorter[7], and Niinemets and Valladare[19]).

CONCLUSION

The study which was conducted to assess the effect of light on seed emergence as well as the effect of different light intensities on the seedlings of *P. conophora* showed that that seeds of *P. conophora* can germinated on both light and dark conditions. However for maximum emergence capacity, the seeds should be planted in light conditions. The growth of *P. conophora* seedlings was enhanced at 25% light intensity in comparison to others as the 25% light intensity produces the best of all the traits examined. Therefore, light conditions recommended for optimum germination of *P. conophora* seeds whereas 25% light intensity is recommended seedlings growth and development.

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