

Influence of Phosphorus and Zinc Application on Leaf Area Ratio (LAR) of Cowpea (*Vigna unguiculata* (L.) Walp) Varieties in the Scrub Savanna of Nigeria

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Abstract

Currently there is increasing attention to evaluate the possible Relative Growth Rate (RGR) of plant species, and to evaluate whether the disparities are principally caused by structural or by physiological characters. Leaf area ratio (LAR) of plant is defined as leaf area per unit total plant biomass and net assimilation rate. Phosphorus (P) and Zinc (Zn) are the most important factors affecting LAR of cowpea (*Vigna unguiculata* L. Walp). Two field trials were conducted at the Faculty of Agriculture and Agricultural Engineering Research Farm of Abubakar Tafawa Balewa University, Bauchi, North Eastern, Nigeria, between 2006 and 2007. The experiments consisted of fifty-four (54) treatments consisting of six varieties by three SSP levels by three Zinc levels. The objective of this study was to examine whether there are disparities in the LAR of cowpea varieties at different P and Zn levels or none? Bauchi is located at 10° 22' N latitude and 90° 47' E longitude at about 109.45m above sea level. Results indicated that year, varieties, P and Zn were found to have significant (P<0.05) effect on LAR. Kanannado recorded the highest LAR than other five varieties. The differences in LAR observed in the varieties may be attributed to the differences in phenotypes and climatic factors such as rainfall, sunshine and availability of nutrients. The increase in LAR indicated an encouraging influence on crop growth rate, dry matter and yield. Application of SSP at 50kg ha^{-1} to Kanannado and IT89KD-288 was observed to be more valuable for enhancement of LAR and hence recommended for growers in the study area.

Keywords: Cowpea varieties, leaf area ratio, *Vigna unguiculata* L., P levels, Zn levels

INTRODUCTION

Cowpea (*Vigna unguiculata* L. Walp) is a major leguminous food crop cultivated all over the world (Musa *et al.*, 2017) where its grains are highly valued for food, and the fodder and haulm used for animal feed during the dry season (Langyintuo *et al.*, 2003). It is a moderately inexpensive and readily accessible source of protein and minerals and contains high concentrations of iron (Fe) and zinc (Zn) (Belarmino *et al.*, 2013). It is one of the most olden crops known to man. Its origin and successive cultivation are related with pearl millet and sorghum

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in Africa. It is an essential part of conventional cropping systems all over the continent, principally in the semi-dry area of West Africa (Steele, 1972). It is nowadays a generally customized and extremely variable crop, domesticated around the world mainly for seed, but also as a vegetable for leafy greens, green pods, fresh shelled green peas, and shelled dried peas, and a cover crop and fodder (Quinn, 2004).

An essential nutrient for the biogenesis of chlorophyll, phosphorus is a component of cell nucleus is indispensable for cell division and development of meristematic tissue. Phosphorus deficiencies result in a decrease in the rate of leaf expansion and photosynthesis per unit leaf area henceforth drop in fodder yield. Crop yields are frequently limited by low soil levels of mineral micronutrients such as zinc (Zn). Essentiality of zinc in plants was recognized as early as 1915 by a scientist called Maze in *Zea mays*. Zinc is an essential mineral nutrient and a cofactor of over 300 enzymes and proteins involved in cell division, nucleic acid metabolism and protein synthesis (Marschner, 1986). Cakmak (2000) has speculated that zinc deficiency stress may impede the activities of a number of antioxidant enzymes, leading to extensive oxidative damage to membrane lipids, proteins, chlorophyll and nucleic acids. Zinc can impact on carbohydrate metabolism at many levels. Moreover, Zn is essential in the biosynthesis of tryptophan, an originator of the auxin-indole-3-acetic acid (Oosterhuis *et al.*, 1996). Zinc deficiency symptoms comprise small leaves, shortened internodes giving the plant a stunted appearance. Availability of zinc in soils and its absorption and translocation in plants is influenced by all other plant nutrients. Zinc in general interacts negatively with phosphorus which depends upon a number of physicochemical properties of soils (Kumar *et al.*, 2016). Plant growth analysis is largely stated as indexes of growth such as crop growth rate, relative growth area, net assimilation rate, leaf area ratio, and leaf area index (Fageria *et al.*, 2006) that offer the first hint toward an understanding of discrepancy in growth rates between genotypes or species (Lambers, 1987).

In recent times there has been growing interest to assess the possible Relative Growth Rate (RGR) of plant species, and to evaluate whether the disparities are principally caused by structural or by physiological characters (Ruggiero *et al.*, 2012). For herbaceous plants and grasses, variation in LAR is the major determinant of interspecific variation in Relative Growth Rate (Poorter & Remkes, 1990; Atkin *et al.*, 1996; Medek *et al.*, 2007). Variation in LAR is usually considered the result of disparity in particular leaf area (SLA) (Poorter & Remkes, 1990; Galmes *et al.*, 2005; Ceriani *et al.*, 2008) and in a rare instance by leaf mass ratio (LMR) (Walter *et al.*, 1993; Quero *et al.*, 2008). LAR of plants at specific instants in time (t) is defined as ratio of assimilatory plant material per unit of plant material (Fageria *et al.* 2006). LAR expresses the valuable leaf area for photosynthesis (Benincasa 2003) or the ratio between the area accountable for capture of both light and carbon dioxide and the entire plant dry mass (Silva and Nogueira 2003). The LAR is the ratio of leaf area and entire plant weight and is the product of a morphological component (specific leaf area), the ratio of leaf area and leaf weight, and the leaf weight ratio, designating the portion of total plant weight apportioned to the leaves (Poorter and Remkes, 1990). Smeets and Garretsen (1986) and Dijkstra and Lambers (1986) reported interspecific variation in Relative Growth Rate (RGR) to be due to variances in LAR.

LAR effects the interception and use of solar radiation, and therefore, growth and yield (Amanullah *et al.*, 2016). LAR is an important yield-determining factor for field grown crops because LAR is a major determinant of light interception and transpiration (Fageria *et al.*, 2006). Rapid leaf area increase is a required attribute in the primary growth phases of cereal crops grown in low rainfall regions, for it gives rise to fast canopy shutting, thus decreasing the

evaporation from the soil surface, and consequently increasing crop water use efficiency (Richards et al., 2002). In more appropriate environments, fast canopy development will make the crop more competitive with weeds for light interception (Lemerle *et al.*, 2001). The aim of this study was to examine whether there is any disparity in the LAR of cowpea varieties under various P and Zn levels or non?

MATERIALS AND METHODS

Description of the Site of the Study

Two field experiments were conducted at the School of Agriculture Research Farm of the Abubakar Tafawa Balewa University, Bauchi, Nigeria, during the growing seasons of 2006 and 2007 to investigate the effects of phosphorus and zinc interactions on leaf area ratio (LAR) of cowpea (*Vigna unguiculata* (L.) Walp) varieties. Bauchi, is located at 10.3010° N latitude and 9.8237° E longitude at an altitude of 109.45m above sea level. It has a monsoonal climate characterized by well-defined rainy and dry seasons. Annual rainfall is mostly distributed between the months of May and October. Average rainfall for the 2006 and 2007 mean monthly temperature; and other meteorological data collected during the experimental periods are presented in Tables 1 and 2, respectively. The soils of the experimental site were found to be moderately well drained, deep, and tropically sandy loam. The physicochemical properties of the soil of the experimental sites for the two years were determined using the procedures described by Black (1965).

Experimental Design

A split-split plot design with a total size of the experimental area of 62m by 50m was used. There were three (3) replicates and each replicate consisted of three sub-plots; each measuring 18.9 m by 2.25 m. Each sub-plot was divided into six (6) sub-sub-plots with each measuring 6.30m by 2.25m. A space of 1m each was left between main plots, and replicates. Half a meter (0.5m), and 50cm were left between sub-plots, and sub-sub-plots respectively. Main plots were assigned to three different levels of single super phosphate (SSP) namely 0kg/ha-1, 25kg/ha-1, and 50kg/ha-1 at random. Sub-plots were assigned to three (3) different levels of Zinc (Zn) namely 0kg/ha-1, 2.5kg/ha-1, and 5kg/ha-1. A total of fifty-four (54) treatments consisting of six varieties by three SSP levels by three Zinc levels. The treatments were randomized using table of random numbers as described by Gomez and Gomez (1984). The experiment continued up to three and half (3½) months that is, from planting to harvest period.

Experimental Plant Materials

Six cowpea varieties collected from International Institute for Tropical Agriculture (IITA) were used in the study. The varieties are IT90K-277, IT93-455-1, IT89KD-288, IT97K-568-18, IT90K-82-2, and Kanannado.

Soil Sampling and Analysis

In each experimental year, soil samples were collected randomly from selected spots in the experimental field before land preparation. The samples were taken at two depths (0-15 and 15-30cm), using a tubular auger. The physicochemical properties of the soil were determined using procedure described by Black (1965). The following soil properties were studied: Nitrogen, phosphorus, potassium, power of hydrogen (pH), cation exchange capacity (CEC) and particle size.

Land Preparation

The land was cleared, ploughed and harrowed. It was then marked into 162 sub-sub-plots. The sub-plot size was 14.2cm². There were 18 sub-sub-plots in a main plot, and 3 main plots in a replicate, and 3 replicates in the whole field experiment.

Sowing of Cowpea Varieties

Sowing was done 3rd and 5th August for the years 2006 and 2007, respectively. Sowing was 75cm row to row and 25cm plant to plant, and three seeds per hill. Seedlings were thinned to one per hill two weeks later. The planting dates were considered in such a way that the varieties mature after end of the rainy season as recommended by IITA (2000).

Fertilizer Application (SSP and Zinc)

Single Super Phosphate (SSP) was incorporated into the soil before sowing as top dressing is not recommended by IITA (2000). Soil application of phosphorus is more effective in increasing phosphorus content (of the soil) than foliar application (IITA, 1973). Zinc sulphate was used as the sources of Zinc and was incorporated in to soil.

Weeds and Pest Control

The first weeding (hoe weeding) was done about three weeks after sowing (21 DAS). Second weeding was at 42 DAS. For the control of insect pests, three sprays of insecticides at 30, 50 and 60 days were used, using an insecticide called *dimethyl cyclopropanecarboxalate*

Determination of Leaf Area Ratio (LAR)

Leaf area ratio was determined as:

$$LAR = \left(\frac{A}{W} \right)$$

where A is leaf area and W is dry weight of the plant (Fageria *et al.*, 2006).

Data Analysis

The results obtained were analyzed using analysis of variance (ANOVA). F test was used for a split-split-plot design using SAS software to test for significant effects of treatments as described by Snedecor and Cochran (1967), Gomez and Gomez (1984), where the observed variance ratios were compared with the table values at either 1 or 5%. Differences between means were separated by the use of Duncan multiple range test (DMRT). Correlation and path co-efficient analyses were carried out to ascertain the causes and effects of the parameters on the seed yield using the procedure described by Little and Hills (1978) in order to assess the type and magnitude of the cause-and-effect relationships among the variables.

Results

Leaf Area Ratio

Year had significant ($P < 0.05$) effect on LAR at most sampling periods. The year 2006 recorded higher LAR than 2007, except at 5 WAS. Varieties had significant effect on LAR in 2006, 2007, and at combined effects of two years, throughout sampling periods, excepts at 9 WAS in 2006. In 2006, the highest and least LAR values were recorded by Kanannado and IT93-455-1 respectively (Table 1). The highest and least varietal effects on LAR in 2007 were recorded by IT89KD-288 and IT93-455-1 respectively (Table 2). Result of the combined effect of varieties on LAR indicated that Kanannado and IT89KD-288, and IT90K-277-2 and IT93-455-1 recorded the highest and least effects respectively (Table 3).

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In the year 2006, both SSP and Zinc levels were observed to have significant ($P < 0.05$) effects on LAR throughout sampling periods. 50 SSP kg ha^{-1} recorded higher LAR than 25 and 0 SSP kg ha^{-1} . For Zinc levels, the control (0 kg ha^{-1}) induced higher effect than 2.5 and 5 kg ha^{-1} (Table 1). In 2007, SSP levels induced significant effects at most sampling periods, with 50 kg ha^{-1} recording higher effect on LAR than 25 and 0 kg ha^{-1} . 2.5 and 5 Zn kg ha^{-1} recorded higher LAR than the control (Table 7). Results of the combined effect of two years on LAR have shown that, 50 SSP kg ha^{-1} recorded higher LAR than 25 and 0 kg ha^{-1} . However, for Zinc levels the control (0 kg ha^{-1}) induced higher effect than 2.5 and 5 kg ha^{-1} (Table 3).

All interactions (V x P, V x Zn, P x Zn, V x P x Zn) were not significant on LAR in 2006 except at 3, and P x Zn at 5 WAS. Interactions were also not significant on LAR in 2007, except of V x P at 3 and 5, and P x Zn at 5 WAS, respectively. Similarly, interactions were also not significant on LAR at combined effect of two years, except of V x Zn, P x Zn, V x P x Zn, Y x V x P, Y x V x Zn, Y x V x P x Zn and Y x P x Zn at 3 WAS and V x Zn at 5 WAS (Table 3).

Table 1. Influence of Single Super Phosphate and Zinc levels on leaf area ratio of cowpea varieties grown at Bauchi, 2006

Treatment	Sampling dates (WAS)			
	3	5	7	9
<u>Varieties</u>				
IT90K-277-2	3.31e	2.58d	1.59b	1.14
IT93-455-1	3.68d	2.46e	1.49b	1.09
IT89KD-288	5.59a	3.32b	3.14a	1.47
IT97K-568-18	4.30b	2.63c	1.63b	1.23
IT90K-82-2	4.31b	3.37a	1.94b	1.34
Kanannado	4.08c	2.23f	1.59b	1.59
SE \pm	0.091	0.032	0.454	0.166
<u>SSP (kg ha^{-1})</u>				
0	4.43a	2.74b	1.72b	1.27b
25	4.15b	2.77a	2.26a	1.24b
50	4.34a	2.79a	1.71b	1.44a
SE \pm	0.064	0.023	0.321	0.117
<u>Zinc (kg ha^{-1})</u>				
0	4.21c	2.81a	1.74b	1.48a
2.5	4.29b	2.74b	2.25a	1.23b
5.0	4.38a	2.74b	1.70b	1.23b
SE \pm	0.064	0.023	0.321	0.117
<u>Interactions</u>				
V x P	*	n.s	n.s	n.s
V x Zn	**	n.s	n.s	n.s
P x Zn	**	*	n.s	n.s
V x P x Zn	**	n.s	n.s	n.s

Means in a column followed by the letter(s) within treatments are not significant different at 5% level of probability using DMRT

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Table 2. Influence of Single Super Phosphate and Zinc levels on leaf area ratio of cowpea varieties grown at Bauchi, 2007

Treatment	Sampling dates (WAS)			
	3	5	7	9
<u>Varieties</u>				
IT90K-277-2	3.19f	2.60d	1.59d	1.15e
IT93-455-1	3.62e	2.47e	1.50e	1.12f
IT89KD-288	5.48a	3.31b	2.00a	1.38a
IT97K-568-18	4.51c	2.66c	1.60d	1.24c
IT90K-82-2	5.04b	3.34a	1.94b	1.34b
Kanannado	4.31d	2.21d	1.64c	1.19d
SE ±	0.069	0.039	0.022	0.011
<u>SSP (kg ha⁻¹)</u>				
0	4.34b	2.74b	1.70	1.23b
25	4.32b	2.75b	1.72	1.23b
50	4.42a	2.81a	1.71	1.25a
SE ±	0.049	0.028	0.016	0.008
<u>Zinc (kg ha⁻¹)</u>				
0	4.34	2.79a	1.70b	1.23b
2.5	4.37	2.76b	1.73a	1.24a
5.0	4.36	2.74b	1.70b	1.24a
SE ±	0.049	0.028	0.016	0.008
<u>Interactions</u>				
V x P	**	*	n.s	n.s
V x Zn	n.s	n.s	n.s	n.s
P x Zn	n.s	*	n.s	n.s
V x P x Zn	n.s	n.s	n.s	n.s

Means in a column followed by the letter(s) within treatments are not significant different at 5% level of probability using DMRT

Table 3. Combined Influence of Single Super Phosphate and Zinc levels on leaf area ratio of cowpea varieties grown at Bauchi, 2006 and 2007

Treatment	Sampling dates (WAS)			
	3	5	7	9
<u>Year (Y)</u>				
2006	4.29a	2.77	1.90a	1.31a
2007	4.36b	2.77	1.71b	1.24b
SE ±	0.011	0.003	0.124	0.062
<u>Varieties</u>				
IT90K-277-2	3.25f	2.59d	1.59c	1.15d
IT93-455-1	3.65e	2.46e	1.49c	1.11d
IT89KD-288	5.53a	3.32a	2.57a	1.43a
IT97K-568-18	4.40c	2.64b	1.61b	1.24c
IT90K-82-2	4.92b	3.36a	1.94c	1.34b
Kanannado	4.20d	2.22f	1.61b	1.39a
SE ±	0.096	0.039	0.227	0.051

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<u>SSP (kgha^{-1})</u>				
0	4.34b	2.74c	1.71b	1.25b
25	4.23c	2.76b	1.99a	1.23b
50	4.40a	2.80a	1.71b	1.34a
SE \pm	0.028	0.018	0.163	0.059
<u>Zinc (kgha^{-1})</u>				
0	4.28c	2.80a	1.70b	1.25a
2.5	4.23b	2.76b	1.99a	1.23b
5.0	4.40a	2.80b	1.71b	1.34b
SE \pm	0.028	0.018	0.163	0.059
<u>Interactions</u>				
V x P	n.s	n.s	n.s	n.s
V x Zn	*	*	n.s	n.s
P x Zn	**	n.s	n.s	n.s
V x P x Zn	*	n.s	n.s	n.s
Y x V	n.s	n.s	n.s	n.s
Y x P	n.s	n.s	n.s	n.s
Y x Zn	n.s	n.s	n.s	n.s
Y x V x P	*	n.s	n.s	n.s
Y x V Zn	**	n.s	n.s	n.s
Y x V x P x Zn	*	n.s	n.s	n.s
Y x P x Zn	**	n.s	n.s	n.s

Means in a column followed by the letter(s) within treatments are not significant different at 5% level of probability using DMRT

DISCUSSION

Effects of Year, Phosphorus, Zinc and Interactions on LAI of different Varieties of Cowpea

The finding that year had significant ($P < 0.05$) effect on LAR at most sampling periods was in conformity to that of Marrison *et al.* (1991) and Aduloju *et al.* (2009), who reported that year had significant effects on LAR of six newly developed soybean genotypes. The difference in the effect of year on LAR may be attributed to difference between the two years (2006 and 2007) in terms of meteorological factors such as sunshine, temperature, etc that were observed to be different. LAR, defined as leaf area (LA) divided by plant dry weight (w), is a growth determinant (Olowolaju *et al.*, 2020). According to Malagi (2003), growth is determined by genotype and environmental factors. Varieties had significant effect on LAR. This result agreed with that of Ali (2004) and (Putul *et al.*, 2021), who reported while working on the interactive effects of seed inoculation and phosphorus application on growth and yield of chick pea (*Cicer arietinum* L.), that LAR was significantly ($P < 0.05$) different among the varieties studied. Similarly, Alam (2014) while studying the physiological traits of wheat as affected by nitrogen fertilization and pattern of planting, reported that LAR differed considerably due to varieties in all the growth stages. Furthermore, Ayalew *et al.* (2022) reported that varieties are significantly different in the cowpea varieties they studied. The LAR and other growth parameters are largely depended on environmental factors. Higher level of SSP at 50 kg ha^{-1} recorded higher LAR than 25 and 0kg ha^{-1} (control). Kang and van Iersel (2004) reported Leaf area ratio increased with increasing fertilizer concentration. Similarly, Weldua *et al.* (2012) and Namakka *et al.* (2017) while working on the effect of Zn and phosphorus application on yield at yield component of Faba bean (*Vicia faba* L.), and effect of phosphorus levels on cowpea reported that higher levels of P at 40 and 60 kg ha^{-1} , and 29 and 39 kg Pha^{-1} had higher ($P < 0.05$) LAR than lower levels of 0 and 20, and 0 and 10 kg ha^{-1} respectively. Increment of LAR due phosphorus application may be attributed to the

activation of metabolic processes, where P is important nutrient that increases the leaf area ratio and improve the rate of photosynthetic assimilate (Yemane and Skjeivg, 2003) and (Mohammed *et al.*, 2022).

Application of Zn at 0 kg ha^{-1} recorded higher LAR than 2.5 and 5 kg ha^{-1} with an exception of the combined effect of the two years. This finding correlated with the work of Akay (2011) and Bozoglu *et al.* (2007), who reported during their different studies on effect of zinc fertilization on chickpea varieties, another legume, that 0 kg ha^{-1} induced higher LAR than higher doses (Yadav *et al.*, 2022). The finding that the control (Zn 0 kg ha^{-1}) had higher LAR than the higher levels of 2.5 and 5 kg ha^{-1} application in this work, is not in consistent with what zinc had been known to induce on growth parameters. According to Fageria (2009), zinc improves root development, fruiting, growth and development of crop plants and zinc is required in small amount but critical concentration was found to allow several key plant physiological pathways to function normally (Liu *et al.*, 2019). These pathways have important roles in photosynthesis and sugar formation, protein synthesis, fertility and seed production and growth regulation (Alloway, 2002). Correspondingly, Hasani *et al.* (2012) and (Hassan *et al.*, 2020), reported that zinc is an essential trace element for plants, being involved in many enzymatic reactions and increase for their good growth and development.

Interactions between Zn and P, Zn and V, and Zn and year were found to be significant on LAR. Khorgamy and Farnia (2009) and Weldua *et al.* (2012) reported that interaction of P and Zn had significant effect on LAR of chick pea, a similar legume. Interaction of P and Zn were reported to stimulate cell division and multiplication thereby increasing growth. Zinc is also vital for oxidation processes in plant cells, involved in transformation of carbohydrates, regulates sugars in plants and promotes photosynthesis and Nitrogen metabolism (Gupta, 1995). Phosphorus is required for synthesis of phosphorus, nucleotides, ATP. Phosphorus deficiency decreases unit leaf area photosynthetic activity for several plant species (Isreal and Rufty, 1988; Sudharani *et al.*, 2020). Interaction between Zn and variety and that between zinc and year were also reported by Khorgamy and Farnia (2009) to have significant effect on LAR of the chick pea cultivars they have worked with. Likewise, Shittu and Ogunwale (2013) reported that interaction between year and zinc and that between Zn and variety have significantly increased LAR. The significant effects of these interactions on LAR may be attributed to the fact that to the ability of Zn under a favourable environmental condition such as when there is sufficient water to induce or stimulate cell expansion (Kiran *et al.*, 2018). This is in agreement with the finding of Shaban and his colleagues. Shaban *et al.* (2012), while studying the response of Chickpea cultivars to integrated application of zinc nutrient with water stress, observed that zinc interact with a favourable climatic condition such as when there is sufficient or adequate water to significantly increased leaf area ratio (LAR).

CONCLUSION

The results indicated that varieties, P, and Zn levels were found to be significant on LAR throughout sampling periods in 2006 and 2007. In 2006, Kanannado and IT93-455-1 induced the highest and least effects on LAR, respectively, whereas in 2007 data collected revealed that, highest and least LAR were recorded by IT89KD-288 and IT93-455-1 respectively. Data on the combined influence of the 2 years, revealed that the highest and least LAR were obtained by IT89KD-288 and Kanannado, and IT93-455-1, respectively. Phosphorus at 50kg ha^{-1} recorded higher LAR than phosphorus levels at 0 and 2.5kg ha^{-1} in 2006, 2007, and at the combined influence of two years. The highest and least zinc influence on LAR in 2006 and 2007 were recorded by 0 and 2.5kg ha^{-1} levels, while on the combined effects of the two years, zinc at 5kg ha^{-1} obtained higher LAR than lower zinc

levels of 0 and 2.5kg ha^{-1} . Of all the six cowpea varieties, Kanannado recorded the highest LAR than the other five varieties at all growth stages except in 2007, where IT89KD-288 recorded higher LAR than the remaining five varieties. Besides, the remaining five varieties produced statistically comparable LAR at different growth periods. The higher LAR of IT93-455-1 might be ascribed to its stretched and wider leaves that give rise to advanced mean single leaf area, leaf area per hill, and per square metre. Leaf size and shape impact on a variety of significant physiological processes, comprising photosynthesis, transpiration and thermoregulation, and differ with a number of environmental factors.

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