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Physiochemical Properties and Anticipated Performance of Selected Plant Species in Lokpaukwu Quarry Site in Abia State, Nigeria

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Abstract The study assessed the physiochemical properties and anticipated performance index (API) of some plants in Lokpaukwu quarry site to determine the impact of quarrying activities on the plants as well as the most suitable plants for green belt development in the area. Three plants species; *Dialium guineense*, *Psidium guajava* and *Pterocarpus soyauxii* were randomly selected for the study. Fresh leaves of the species were collected for foliar epidermal studies through clearing method and biochemical parameters; air pollution tolerance index (APTI) and API studies were evaluated using standard procedures. Results of foliar analysis showed clear evidence of environmental stress at the polluted site such as stretched epidermal cells as well as plasmolysed stomata with deformed guard cells. There were variations in some stomata parameters (such as stomata density and pore size) of the plants collected from the polluted site when compared with those from unpolluted site (control). Biochemical properties had significantly higher values ($p < 0.05$) at the control relative to those at the polluted site except for relative water content. The variations also occurred among the plants species. The values for ascorbic acid and total chlorophyll were highest in *P. soyauxii*. The APTI and API followed the sequences: *P. soyauxii* > *D. guineense* > *P. guajava* and *P. soyauxii* > *D. guineense* = *P. guajava* respectively. Based on APTI and API, *P. soyauxii* emerged the most suitable plant for bio-mitigation of air pollution and green belt development amongst the three plants.

Keywords: *Lokpaukwu, quarry site, foliar parameters, APTI, API, green belt development*

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1. Introduction

Air pollution is one of the key problems facing the world today [1]. The fact that the human population, vehicular traffic, road transportation and industrialization have been on the increase in the past few years has contributed to the exacerbation of air pollution as a result of the release of pollutants in form of gases and particulate matter into the atmosphere [2]. An important source of air pollution is the quarry industry which generates gaseous and particulate pollutants [3]. These pollutants are known to impact adversely on plant health as manifested by physiological and biochemical alterations and changes in affected plants [4,5,6,7]. Plants exposed to pollutants experience physiological change before exhibiting visible damages to leaves [8]. Trees play a significant role in purifying the atmosphere by absorbing and utilizing gaseous pollutants and particulate matter [9,10]. They provide enormous leaf area upon which the atmospheric pollutants – dusts and gases are impinged, absorbed and accumulated, and as a result air pollution is reduced [11]. Plants are both scavengers and the initial interceptors of air pollutants making them useful in air pollution monitoring [12]. Plant tolerance to air pollution is important in understanding and identifying that have the capacity to indicate or mitigate air pollution [13,14]. This property of plants is determined by several factors such as pollutant characteristics and load [3], and plant characteristics [15]. In other to assess plant tolerance to air pollutants Singh and Rao (1983) developed the concept of air pollution tolerance index (APTI). This is a synthesis of critical plants biochemical properties namely pH, total chlorophyll, ascorbic acid and relative water content of leave extract [13,14,16].This index is useful in evaluating plant response to air pollutants, not only on biochemical parameter but also provides information that enables the investigator combat air pollution through green belt development [17]. More recently, the concept of anticipated performance index, an improvement of APTI have been used in comprehensively assessing the capacity of dominant species for air pollution mitigation and green belt development taking into consideration ethnobotanical characteristics [17,18,19]. In the study area, rock quarrying is the major source of air pollution which severely affects vegetation health and performance. However, in the study areas, no work has been published on the effects of quarry sourced air pollutants on the physiochemical and biological properties of plants although this has been done elsewhere within the region [14,18]. This study was therefore necessary to be embarked on given that it would help understand plant physiological response to air pollution from the study area as well as identify tolerant species which can be used for both mitigation of air pollution and green belt development.

2. Materials and Methods

2.1. Study Area

Lokpa-ukwu community is located between latitudes 05°55’00’’N and 06°03’00’’N, and longitudes 07°21’05’’E and 07°31’33’’E. The quarry site lies between latitude 5°50’00”N and 6°00’00”N and longitude 7°2’00”E and 7°30’00”E. Mean annual rainfall is about 2250 mm concentrated in the rainy season that lasts from March to October. A short dry period (August break) divides the periods of peak rainfall into two. The dry season lasts from November to February and it is characterized by dry dusty winds from the north. The area has a fairly constant average temperature of about 27°C. Lokpa-ukwu community lies in the southern tropical rain forest region of Nigeria. Most of the area, apart from areas converted to quarry and farmland is forest dominated by oil palm trees and fruit trees like mango, guava and breadfruits. The study area is characterized by high and low land. The relief of the area is generally undulating and no location exceeds 500 m above sea level.

2.2. Plant Selection and Sample Collection

Plants species growing within 100-meter perimeter of the boundary of the quarry site constituted the study population. A transect of 50 m by 50 m in size was used as sampling plots on each side of the perimeter. Frequency of occurrence of plants species in each transect was then used to determine the most dominant plant species. Out of the twelve dominant plant species identified, Lucky dip without replacement was used in picking the first three samples as described by Ogbonna *et al.* [22]. The selected plants were *Psidium guajava*, *Dialium guineense*, *Pterocarpus soyauxii*. Leaf samples were taken from the lowest branch of each selected plants facing the pollution source. Freshly collected samples were labeled, placed in a sealed poly bag and immediately sent to the laboratory for analysis. Control samples were collected from plants of equal girth within the premises of Abia State University, Uturu. The plants were collected during the dry season (February 2018) in the afternoon hours. The plants collected were sent to the laboratory for the analysis of foliar and biochemical parameters.

2.3. Micro-morphological Analysis of the Foliar Epidermis

Foliar epidermises of the adaxial (upper) and abaxial (lower) surfaces of the leaves were prepared by clearing method. The leaf samples were cleared by soaking in petri dishes containing commercial bleach (3.5% sodium hypochlorite) for 18 hours. Then, the epidermal strips of the samples were scraped gently with the aid of forceps and placed on clean slide, stained with safranin and covered with a cover slip. The light was viewed under Olympus Tokyo (Japan no. 271961) microscope at X400 magnification and photomicrographs were taken with Motic camera 2.0. The stomata parameters (size, pore size, frequency, density and index) were observed and assessed. The stomata length and width were measured with the aid of Motic software in five replicates for each sample [14].

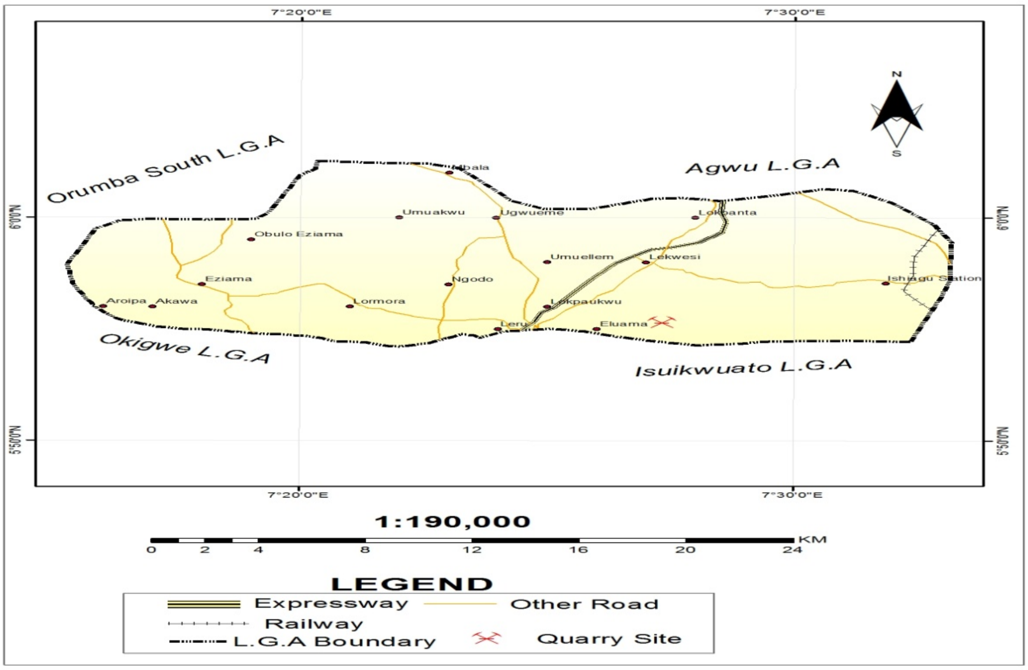

Figure 1. Map of Lokpaukwu, Study Area (Source: Onyeonoro *et al.* [21])

2.4. Determination of Biochemical Parameters

2.4.1. Ascorbic Acid Content

The method of Ogbonna *et al.* [14] was followed in the determination of ascorbic acid content. One gram of the leaf sample was extracted with 4 ml of oxalic acid – EDTA solution. To a test tube containing the extract, 1 ml of 5% H_2SO_4, 2 ml of ammonium molybdate and 3 ml of water were added successively and allowed to settle for 15 minutes. The absorbance was measured at 760 nm and the concentration of ascorbic acid was calculated from a standard curve.

2.4.2. Determination of Total Chlorophyll Content (TCH)

Three (3) grams of the leaf sample was macerated with 10 ml of 80 % acetone and the liquid portion was decanted after allowing to settle for 15 minutes, then centrifuged at 2,500 rpm for 3 minutes. The absorbance of the supernatant was measured at 663 nm using UV-Vis spectrophotometer [14].

2.4.3. Determination of Leaf pH

Few quantity of the leaf sample was macerated with de-ionized water and filter through an ash less filter paper. The filtrate was read for pH with the aid of a digital pH meter [14].

2.4.4. Determination of Percentage Relative Water Content (RWC)

The fresh leaf sample was weighed and the fresh mass (FM) was recorded. The same sample was floated in distilled water inside a closed petri dish at room temperature for 24 hours. At the end of the incubation period, the leaf sample was wiped dry gently with blotted paper and re weighed to obtain the turgid mass (TM). It was then placed in a pre-heated oven at 80 ºC for 48 hours. Thereafter the leaf was weighed to obtain the Dry Mass (DM). Relative water content was calculated using the formula:

$$RWC = \frac{FM - DM}{TM - DM} x100$$

Where; FM = Fresh mass; DM = Dry mass and TM = Turgid mass [14].

Calculation of air pollution tolerance index (APTI): The APTI was calculated as described by Ogbonna *et al.* [14]. The formula is given as;

The formula is given as;

$$APTI = \frac{A(T+P)+R}{10}$$

Where; A = Ascorbic acid content (mg/g); T = Total chlorophyll (mg/g); P = pH of the leaf extract and R = Relative water content of leaf (%).

On the basis of APTI , the selected plants were rated as follows: APTI 30 – 100 is considered tolerant plant species; APTI 17 – 29 is considered intermediate plant species. APTI 1-16 is considered sensitive plant species. APTI < 1 is considered very sensitive plant species [23].

2.5. Calculation of Anticipated Performance Index (API)

Anticipated performance index was calculated according to the method described by Dhankhar *et al.* [23] with slight modification. This was determined by combining the values of APTI with the biological characters (plant type, habit, size, canopy structure, and leaf size and texture) and economic values (e.g. use as ethnomedicine, food, fuel wood, fodder and construction material).

2.6. Statistical Analyses

Analysis of variance was performed to check for significance (at $p < 0.05$) among the three samples while Duncan multiple range test was used for mean separation. Students' independent t-test was used to check for significance (at $p < 0.05$) between samples from polluted and control sites. The statistical software, SPSS version 20 was used for all statistical analysis.

3. Results

3.1. Stomata Parameters

Table 1 below show the result of quantitative stomata studies of the plants across the two study locations. There were significant differences ($p < 0.05$) in the quantitative stomata parameters among the plants. For instance, *P. guajava* had the highest value in stomata length and pore size at both study area and control while *D. guineense* had the lowest value in stomata length at the study area. *P. guajava* had the highest value in stomata size at both study area, and for stomata frequency at both study area and control, while *P. soyauxii* had the lowest value in stomata frequency (Table 1). Comparing the stomata data from the polluted sites with those from control sites, significant differences were also found at $p < 0.05$. It was observed that the samples collected from the polluted sites had higher stomatal frequency but smaller stomata and narrower stomata pores. This was particularly evident in *D. guineense* and *P. soyauxii.* However, *P. guajava* did not show any variation in terms of stomata parameters from the two locations (Table 1).

3.3. Biochemical Properties and Air Pollution Tolerance of Plants

Variations were observed in all parameters among plants collected from the polluted area and control sites (Table 2). Significant differences ($p < 0.05$) were found in all parameters in plants at the study area with *P. soyauxii* having the highest value for total chlorophyll content. The highest values for ascorbic acid content and relative water content were recorded in *D. guineense* while pH was highest in *P. guajava.* All parameters, except relative water content, were significantly higher in samples from the control sites than those from polluted sites (Table 2).

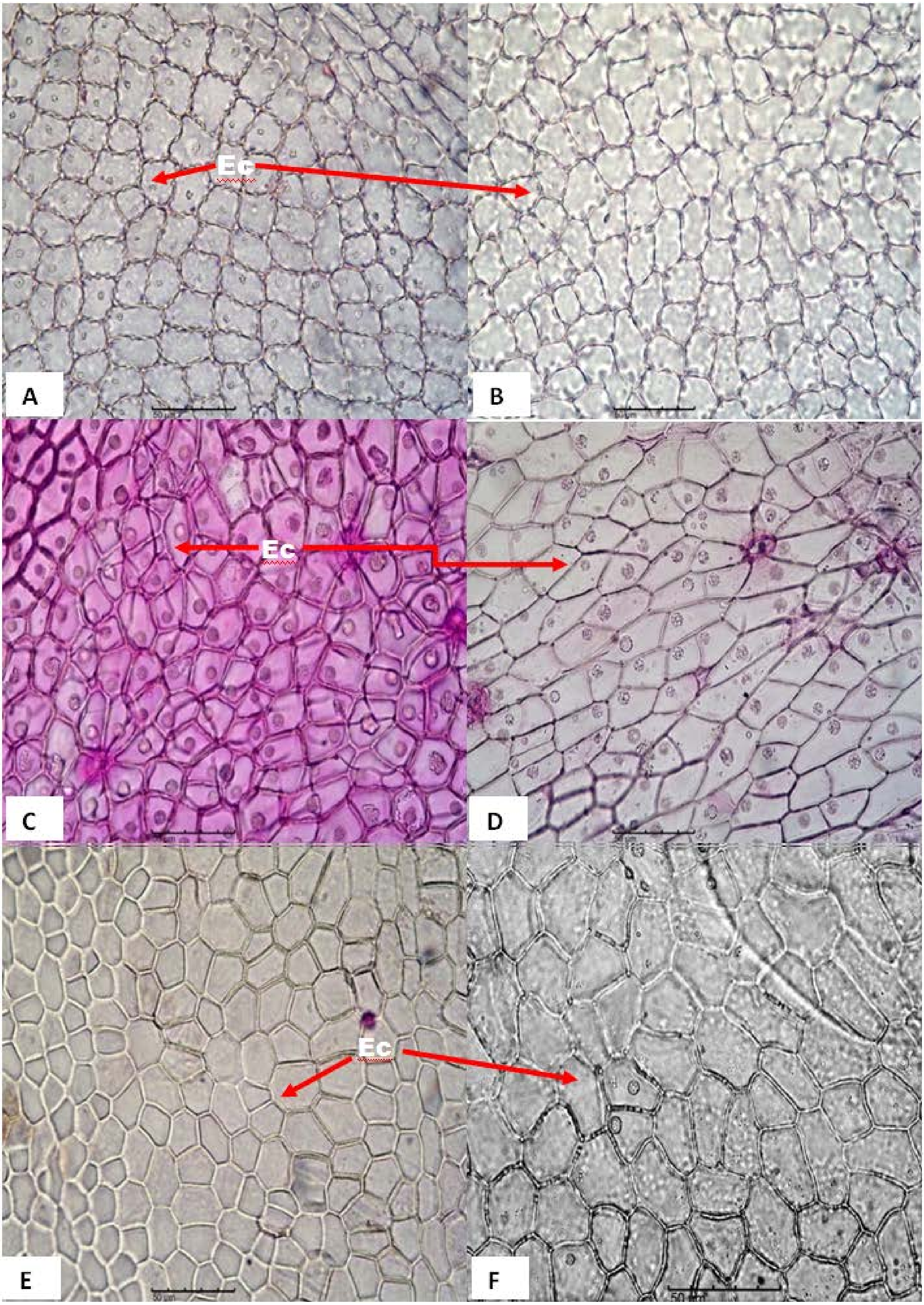

Plate 1. Adaxial surfaces of the leaf epidermises from the control and polluted sites respectively (A-B) = *D. guineense*; (C-D) = *P. guavaja*; (E-F) = *P. soyauxii.* Pictures show stretched and necrotic epidermal cells (Ec) in samples collected from the polluted sites

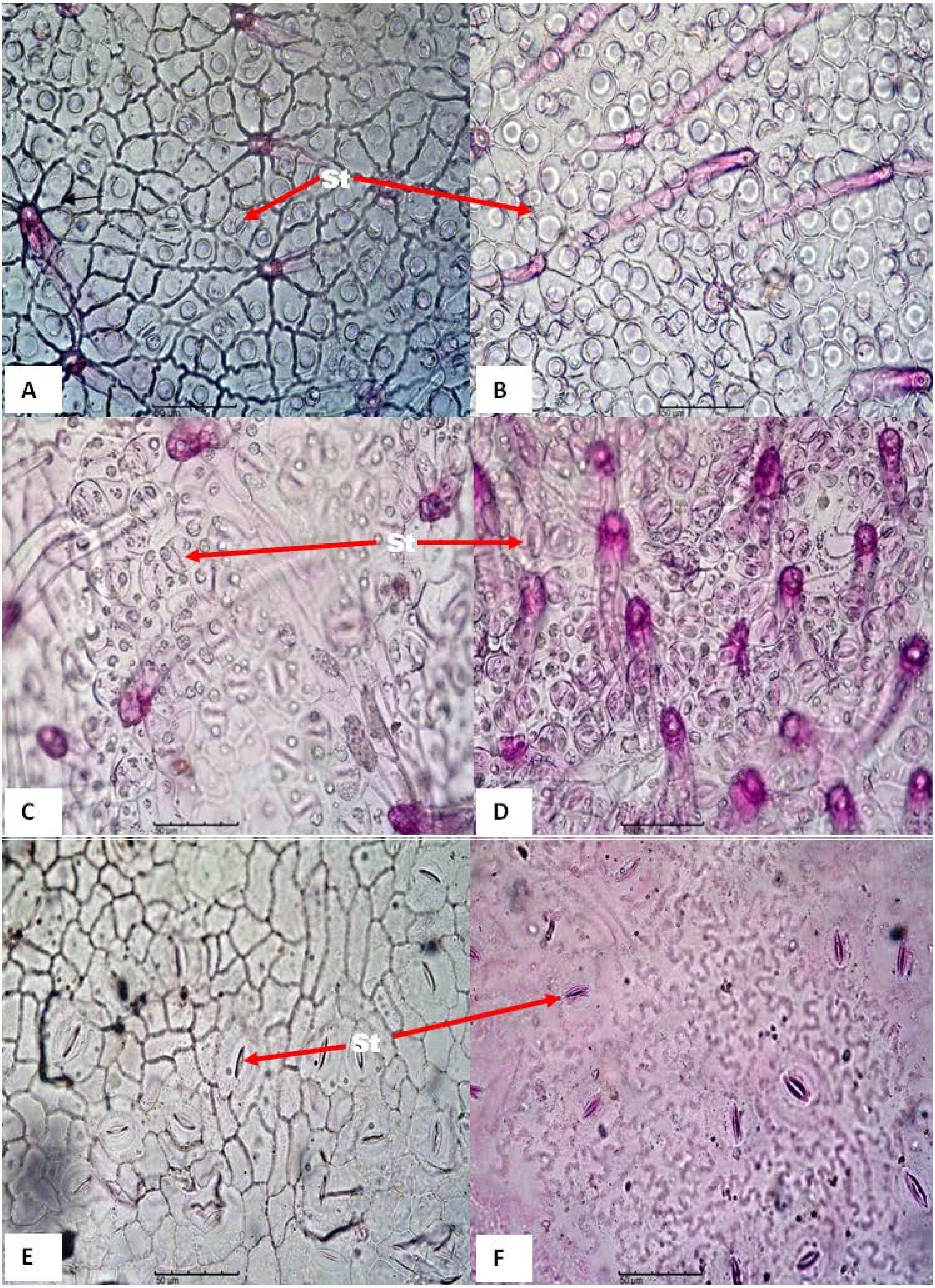

Plate 2. Abaxial surfaces of the leaf epidermises from the control and polluted sites respectively (A-B) = *D. guineense*; (C-D) = *P. guavaja*; (E-F) = *P. soyauxii.* Pictures show shrunk stomata with deformed guard cells (St) in samples collected from the polluted sites

Table 1. Stomata parameters of the plants at both the polluted and control sites

Parameter	Site	*D. guineense*	*P. guajava*	*P. soyauxii*
Stomata length (μm)	Polluted	16.77 ± 1.27^{b}	21.11 ± 1.03^{a}	21.07 ± 0.75^{a}
	Control	15.65 ± 1.31^{c}	17.90 ± 0.65^{b}	$24.21 \pm 0.29^{a*}$
Stomata width (μm)	Polluted	$11.94 \pm 0.96^{b*}$	15.03 ± 1.41^{a}	14.70 ± 1.41^{a}
	Control	7.29 ± 0.59^{c}	14.97 ± 0.66^{b}	$15.41 \pm 0.17^{a*}$
Stomata size (μm^2)	Polluted	202.72 ± 32.01^{b}	319.37 ± 41.78^{a}	311.12 ± 37.83^{a}
	Control	115.68 ± 19.54^{c}	268.74 ± 20.25^{b}	372.99 ± 7.70^{a}
Stomata frequency (mm^{-2})	Polluted	$135.29 \pm 3.40^{b*}$	392.16 ± 8.55^{a}	$121.57 \pm 1.96^{c*}$
	Control	111.76 ± 3.40^{b}	384.31 ± 10.38^{a}	109.80 ± 1.96^{c}
Pore size (μm^2)	Polluted	46.07 ± 7.27^{c}	72.58 ± 9.49^{b}	79.77 ± 9.70^{a}
	Control	$68.04 \pm 11.50^{b*}$	63.99 ± 4.82^{c}	96.65 ± 2.25^{a}

Values expressed as mean ±SE of 5-replicate data
Means with different letters as superscripts across a row are significantly different at $p \leq 0.05$
Means with same letters as superscripts across a row are not significantly different at $p \leq 0.05$
Significantly higher at $p \leq 0.05$ along a column for each parameter between the study sites.

Table 2. Biochemical parameters of the plants at both polluted and control sites

Parameter	Site	*D. guineense*	*P. guajava*	*P. soyauxii*
Total chlorophyll (mg/g)	Polluted	9.87 ± 0.00^{b}	8.74 ± 0.00^{c}	29.21 ± 0.00^{a}
	Control	$12.56 \pm 0.001^{b*}$	$11.71 \pm 0.01^{c*}$	$44.65 \pm 0.03^{a*}$
Beta carotene (mg/g)	Polluted	3.49 ± 0.00^{c}	4.43 ± 0.00^{b}	9.76 ± 0.00^{a}
	Control	$6.50 \pm 0.01^{b*}$	$5.48 \pm 0.001^{c*}$	$15.42 \pm 0.001^{a*}$
Ascorbic acid (mg/g)	Polluted	8.17 ± 0.01^{a}	5.44 ± 0.01^{c}	7.41 ± 0.02^{b}
	Control	$10.24 \pm 0.01^{b*}$	$6.66 \pm 0.02^{c*}$	$11.63 \pm 0.04^{a*}$
pH	Polluted	5.30 ± 0.00^{c}	5.80 ± 0.00^{a}	5.70 ± 0.00^{b}
	Control	$6.55 \pm 0.05^{b*}$	$6.40 \pm 0.00^{c*}$	$6.75 \pm 0.05^{a*}$
RWC (%)	Polluted	$46.23 \pm 0.03^{b*}$	$51.22 \pm 0.02^{a*}$	$37.97 \pm 0.02^{c*}$
	Control	28.97 ± 0.02^{b}	31.58 ± 0.43^{a}	28.58 ± 0.04^{c}

Values expressed as mean ±SE of 5-replicate data
Means with different letters as superscripts across a row are significantly different at $p \leq 0.05$
Means with same letters as superscripts across a row are not significantly different at $p \leq 0.05$
Significantly higher at $p \leq 0.05$ along a column for each parameter between the study sites.

Table 3. APTI and API of the plants at both polluted and control sites

Parameter	APTI	APTI Grade	% API	Grade
		Polluted Site		
D. guineense	17.01 ± 0.02	Intermediate	50.00	Good
P. guajava	13.02 ± 0.00	Sensitive	50.00	Good
P. soyauxii	29.65 ± 0.06	Tolerant	56.00	Very Good
		Control Site		
D. guineense	22.46 ± 0.06	Intermediate	56.00	Very Good
P. guajava	15.22 ± 0.01	Sensitive	50.00	Good
P. soyauxii	62.65 ± 0.25	Tolerant	69.00	Excellent

4. Discussion

The significance of foliar epidermal parameters, biochemical parameters, APTI and API as bio-indicators in environmental monitoring has been reported by several authors [13,14,17,20,23]. In this study, we investigated the potentials of these variables in selected tree species towards ascertaining the best species for green belt designing around the Lokpaukwu quarry site, in Southeast Nigeria.

4.1. Foliar Parameters

The foliar epidermal study showed that the leaf samples, especially those of *D. guineense* and *P. soyauxii* collected from the polluted (quarry) site had some alterations in their stomata arrangement when compared with those collected from the control site. The control samples had well defined stomata patterns, which was not obtainable at the polluted site where the stomata showed signs of physiological stress. Similarly, the stomata from samples of control site had turgid and healthy guard cells unlike in polluted site where the stomata showed deformed and plasmolysed guard cells. All this represent that the plants were affected by dust pollution as a result of the quarrying activities going on at the polluted site [13,14,20].

The epidermal cells were intact and well outlined in the samples collected from the control site whereas they were stretched, and in some cases, deformed in the samples from the polluted area. Such arrangement could only be caused by contractile force induced by environmental assaults resulting from the quarry activities [17]. The epidermal layer of the plants from the polluted area revealed evidences of necrosis and white patches. This feature is attributable to wrinkles resulting from environmental stress. This agrees with the reports of

[14,17,20] who noted similar changes and proposed that they all represent environmental stress posed to the plants as a result of pollution.

4.2. Biochemical Properties

Ascorbic acid serves as an indicator of oxidative in plants and therefore is a critical parameter for the activation of defense mechanisms in plants [13,24]. Ascorbic acid is Ph dependent as high Ascorbic acid if linked to high ph in plants [25]. This trend was not seen in the polluted area but was observed in the control samples. This study also indicated that a lower ascorbic acid values was associated with lower APTI as was the case in *P. guajava*. This has been noted by Randhi and Reddy [26] among other researchers and explains the importance of ascorbic acid as a multiplication factor in the APTI equation [27,28,29]. The chlorophyll content of plant is an indication of its photosynthetic vigour as well as its tolerance capacity [30]. According to Katiyar and Dubey [31], chlorophyll content in plants varies from species to species, and also depends on age of leaf, and pollution level alongside with other biotic and abiotic factors. *P. soyauxii* had the highest total chlorophyll at the polluted area as well as the highest APTI and API, indicating the importance of the parameter in pollution tolerance by plants. Studies such as those of Misanjo *et al.* [32], Jyothi and Jaya [33] and Mir *et al.* [34] have reported reduction of chlorophyll content in plants in stressed environments. This was observed in our findings given that chlorophyll content was significantly higher in plants at the control area. Relative water content of plants is associated with protoplasmic permeability in cells which causes loss of water and dissolved nutrients [35] The maintenance of relative water content in plants is crucial for pollution tolerance [36]. Relative water content was found to be highest in *P. guajava* and lowest in *P. soyauxii*. The high water content of some plants in the study may be attributed to stomata occlusion by particulate deposits on leaves [14]. Relative water content was generally lower in this study compared to other studies within the mining region. This can be attributed to the fact that the study was carried out during the dry season. High pH may increase the conversion efficiency from hexose sugar to ascorbic acid [24]. This explains its relationship with ascorbic acid content of plants. Pandit and Paudel [37] noted that leaf pH of 7 in plants was suggestive of higher pollution tolerance. Based on the findings of the study, *P. guajava* had the lowest APTI even though it also had the highest Ph. This may be explained by the greater roles other biochemical parameters play in pollution tolerance of plants as reported by Enete and Ogbonna [13].

4.3. Air Pollution Tolerance and Anticipated Performance of the Plants

Air pollution tolerance index is an index that indicates the capacity of plants to combat air pollution. Plants that have higher index values are considered tolerant to air pollution and can be used in air pollution mitigation, while those with low index values are considered sensitive and are only used to indicate air pollution [36,38,39]. Our findings indicate that APTI is plant dependent as suggested by Tiwari and Tiwari [40] as seen from the variations in APTI values among the plants. The results of this study also show that APTI was significantly higher for plants collected at the control sites than at the polluted area. This is accounted for by similar trend in biochemical parameters of the plants. The sequence of air pollution tolerance of plants at the polluted area from the results was *P. soyauxii* > *D. guineense* > *P. guajava*. Thus *P. soyauxii* had the highest APTI of 29.65, suggesting that the plant has intermediate tolerance for air pollution from the polluted site. The results of this study, further indicates that at the polluted site *P. guajava* and *D. guineense* are sensitive to air pollution. A similar finding has been reported for *P. guajava* by Ogbonna *et al.* [20]. Our findings suggest that *P. soyauxii* can be used in bio-mitigation of air pollution at the polluted site while *P. guajava* and *D. guineense* can be used as bio-indicators of air pollution. API is used to indicate capacity of dominant plant species to improve air quality and for green belt development [37]. It combines morphological and socio-economic attributes of the plants to provide a better predictive method for selecting plants suitable for environmental monitoring. API of plants at the polluted site followed the order *P. soyauxii* > *D. guineense* = *P. guajava*. Therefore, based on API, *P. soyauxii* is the most suitable for green belt development among the three plants. This might have been influenced by its numerous economic uses. From our field study experience, *P. soyauxii* is used for timber in construction, fuel wood, and as food and medicine by the local people of Lokpaukwu community.

5. Conclusion

The study assessed the effects of air pollution from quarrying activities on foliar and biochemical properties of some plants in Lokpaukwu with a view of profiling plant tolerance and performance. Variations were observed in plant foliar character and biochemical properties as well as in APTI and API. Foliar analysis indicated that plants at the polluted site showed evidence of pollution stress such as destroyed stomata, plasmolysed cells and altered venial arrangements in comparison with plants at the control. Based on APTI and API Pterocarpus soyauxii was the most tolerant and suitable plant species or green belt development. It is, therefore, recommended that this plant should be considered by environmental researchers and managers to be planted at quarry sites because of its tolerance to stress and potentials in mitigating environmental pollution while the other plants may be used as bio-indicator of environmental pollution.

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