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Influence of Rock Quarrying Activities on the Physiochemical Characteristics of Selected Edible Fruit Trees in Uturu, Abia State, Nigeria

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Abstract This study was conducted to investigate effects of quarry dust on physiological and biochemical properties of selected edible fruit trees in the vicinity of a rock quarry site at Ugwuele, Uturu. Five fruit trees growing in the quarry site were randomly selected for the study. Control samples were collected six kilometers away from the study area. Significant differences were found in ascorbic acid content and relative water content of plants at study area and control ($p<0.05$), with both parameters having lower values at the study area. No Significant difference ($p<0.05$) was established between plants at study site and control in pH. Analysis of foliar structure of the plants showed that plants from the quarry site had more damaged stomata pores and guard cells in comparison with control samples. In addition, distorted vernal arrangement was observed in plants at the quarry site. The study suggests that fruit trees within the quarry site showed signs of stress, including oxidative stress. In order to protect human health, plants growing in the vicinity of quarry sites should not be consumed, as they may have accumulated toxic contaminants. It is very important that plants tolerant to air pollution from quarry sites are identified and used in the bio-mitigation air pollution from such sources.

Keywords: *quarry site, fruit trees, oxidative stress, foliar structure, air pollution tolerance*

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1. Introduction

Quarrying is an important economic activity involving the extraction of non-fuel and non-metal minerals that are utilized mainly in the construction industry. However, it is an important source of air pollutants in the environment. [1] These pollutants are largely particulate in nature, resulting from mechanical processes such as crushing of rock into smaller sizes or through aggregation by condensation of gases and vapour. Suspended particulate matter from quarry sites is a major source of air pollution, the severity of which depends on factors that include local climate, particle load in the ambient air and the size and chemistry of the dust particles [2]. Particulate pollution from quarrying and other activities have been known to have adverse effects on human health, soil and air quality and on vegetation. [1,3,4]

Plants play many roles including serving as a source of medicine and food, and are essential in the stabilization natural systems. [5] They are stationary, and are therefore a reliable means of monitoring air pollution from various sources including quarry sites. [6,7,8] Their leaves and other parts also provide canopy that trap and accumulate particulate matter. [9,10] Plants with higher length such as trees are known to suffer more stress and damage [11].

Physiological alterations and biochemical changes are observed in plants in the vicinity of stressed environments. Physiological damage is usually observed before morphological changes manifest. [12] These include distorted foliar structure and abrasion of leaves and cuticles. [13,14,15] necrosis and stunted growth, inhibition of photosynthesis, stomata conductance and generally, oxidative stress. [16,17] Oxidative stress reflects an imbalance between the biological system’s ability to readily detoxify the reactive intermediates or to repair resulting damage. The impacts of quarrying activities on plants are significant, and the accumulation of particulate matter on plant may be toxic and leads to health problem in individuals who consume the plants. [14] The response of plants to air pollution from a given source can also be examined by determining the air pollution tolerance index of such plants through the assessment of pH, ascorbic acid, total chlorophyll, and relative water content of leaves. [12,18]

The aim of this study is to assess oxidative stress and air pollution tolerance of edible fruit trees growing in the Ugwuele quarry site. This was done through the examination of the foliar structure of the plants, and determination of the pH, ascorbic acid and relative water content of their leaves.

2. Methodology

2.1. Study Area

The study area is situated at Ugwuele in Uturu community Abia State. It is located at latitude $5^0$35’N and $5^0$55’ N and between longitudes 7 ^{0}22’E and 7^0 30’E while the quarry site is located at 5^0 87’ 15’’ N and 7^0 42’ 54’’ E (Figure 1). The relief is mainly undulating with elevation ranging from 100 meters to about 224metres. Soil type is false bedded sand stones of the Mastrichitan geologic era with igneous rock outcrops. The study area is in the humid tropics with annual rainfall between 1500mm and 2000mm with mean relative humidity and temperature of over 70% and 27°C respectively. The vegetation of the study area is the rain forest type but as a result of high incidence of anthropogenic interference including rock mining, the vegetation is tending to the derived savanna type. Quarrying activities have been carried out in the study area for more than twenty years with the attendant environmental problems.

2.2. Plant Selection

The study population consisted of all edible fruit trees growing within the quarry site. Out of the ten identified plants, five were randomly selected using sampling without replacement. The selected trees were; *Anacardium occidentale, Annona muricata, Citrus sinensis, Magnifera indica and Psidium guajava*. Control samples were collected six kilometers away from the study site in Achara community, Uturu. Leaf samples randomly collected from the top, middle and base of each of the selected trees. The collected samples from each tree were pooled together to form composite sample for laboratory analysis.

2.3. Laboratory Analysis

2.3.1. Foliar Analysis

Foliar epidermis of the adaxial (upper surface) and abaxial (lower surface) of the leaf samples were prepared by impression method as described in [14,15] Using a camel hair brush, nail vanish was applied on 22 x 22cm portion of both the adaxial and abaxial surface of the leaf to dry for 10 minutes. Subsequent coatings were applied for the second and third times and left to dry for 10 minutes respectively. The samples were then passed through air current for 1 hour to ensure maximum dryness.

Epidermal strips of the leaf samples were scrapped gently with the aid of forceps and placed on a clean slide, stained with safarin, washed three times with alcohol and covered with a cover slip before mounting for microscopic examination. Slides were viewed under a light microscope at x40 magnifications and photomicrographs were taken with 20iss light microscope with MC’35 camera for 53mm film at x400 magnification.

2.3.2. Biochemical Analysis

Biochemical analysis involved the determination of the, ascorbic acid, chlorophyll, pH and relative water content of leave extracts.

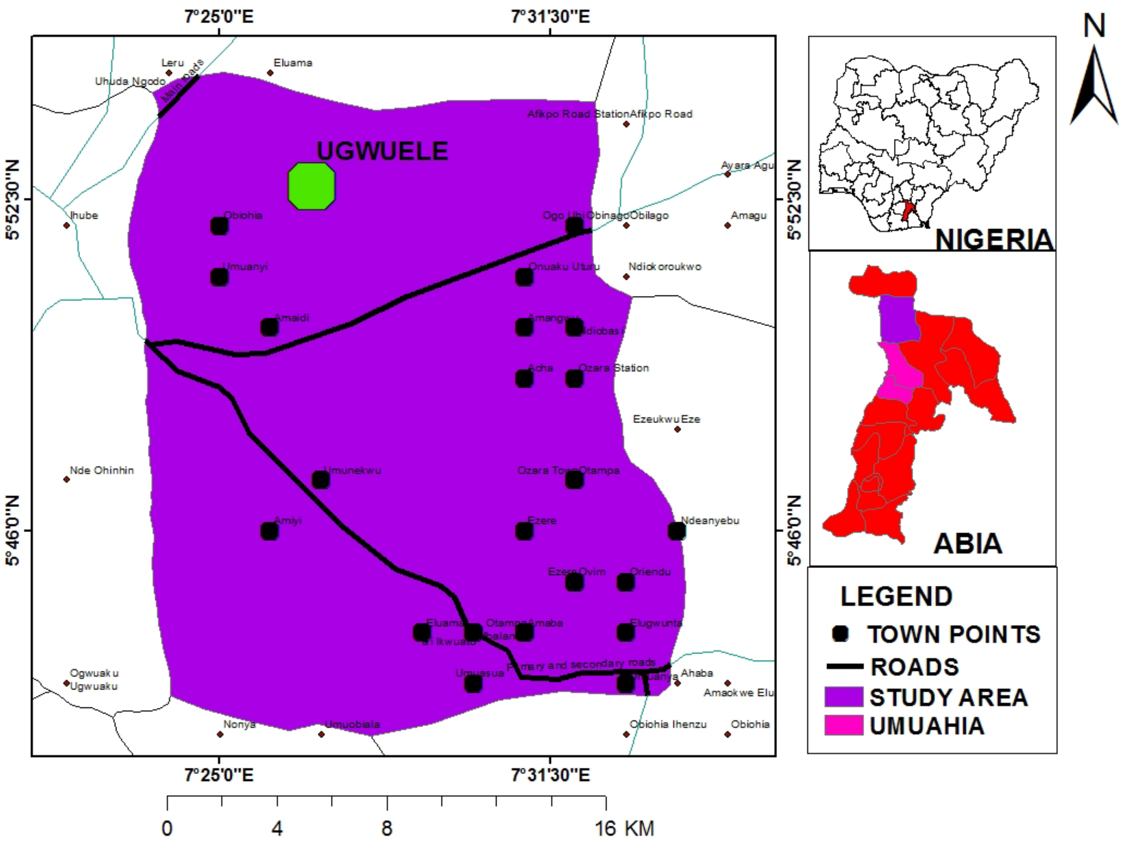

Figure 1. UGWUELE STUDY AREA MAP

2.3.3. Determination of Ascorbic Acid Content

Ascorbic acid was determined by the titremetric method as described in. [6] The ascorbic acid was extracted with 20ml of distilled H_2O in 30ml H_2SO_4 and 0.5mol, oxalic acid. Exactly 2g of the pulverized leaf sample was added with the mixture and stirred thoroughly for 10mins. The mixture was filtered and 10ml of the filtrate was titrated against 0.05mol iodine solution, using starch mucilage as an indicator to end point.

2.3.4. Determination of pH

The pH was determined by the Direct Reading Engineering Method using a digital pH meter [15]. The leaf extract was made by manual squeezing with distil water and squeezed directly into an already labeled test tube. The pH meter was pre-calibrated prior to its usage, using buffer solution of pH 7.4. The electrode was carefully dipped into the extract in a 10ml beaker. The value displayed on the Crystal Liquid Display Panel (CLD) of the pH meter, was recorded as the pH value.

2.3.5. Determination of Relative Water Content

To determine relative water content, fresh leaf sample was weighed and recorded as Fresh Mass (FM). It was floated in distilled H_2O inside a closed petri-dish, at a room temperature for 24hrs. At the end of the incubation period, leaf sample was wiped dry to obtain the Turgid Mass(TM). It was then placed in a pre-heated oven at 100°C for 5hrs after which the leaf was weighed to obtain the Dry Mass (DM). The assay was done in triplicates and the average of the readings was used. The relative water content was calculated as described by. [19]

2.3.6. Determination of Chlorophyll Content

Chlorophyll Content was determined by the method described by. [20] Exactly 3g of the leaf samples were blended and then extracted with 10 ml of 80% acetone, left for 15 minutes and the liquid portion decanted and centrifuged at 2,500 rpm for 3 minutes. The supernatant was collected and its absorbance measured at 663 nm using spectrophotometer.

2.4. Statistical Analysis

All the assays were made in triplicate and the results expressed as arithmetic mean ± SD (standard deviation). The mean and SD were determined using EXCEL software. One-way ANOVA with a Turkey test post-hoc was used to identify statistical differences among groups. A *p*-value of ≤ 0.05 was considered statistically significant.

3. Results

3.1. Biochemical Properties

Figure 2 shows the effect of rock quarrying on ascorbic acid values of five edible fruit trees. The ascorbic acid value ranged from 0.88mg/g in *Annona muricata to* 1.76mg/g in *Citrus sinensis* with a mean value of 1.32mg/g while at the control site it ranged from 0.88mg/g in *Psidium guajava* to 3.52 mg/g in *Citrus sinensis* with a mean value of 1.58mg/g. Significant differences were found in ascorbic acid content of *Magnifera indica*, *Citrus sinenses* and *Annono muricata* between study area and control ($p<0.05$). *Citrus sinenses* had significantly higher ascorbic acid content than other plants ($p<0.05$).

Figure 3 shows the effect of rock quarrying on the pH values of five edible fruit trees. The pH at the study site ranged from 4.20 in *Annacadium occidentale to* 5.40 in *Psidium guajava* with a mean value of 4.84 while at the control site it ranged from 4.40 in *Annacardium occidentale* to 5.50 in *Citrus sinensis* with a mean value of 5.0. No Significant differences were found in pH between study area and control, and among the studied plants ($p<0.05$).

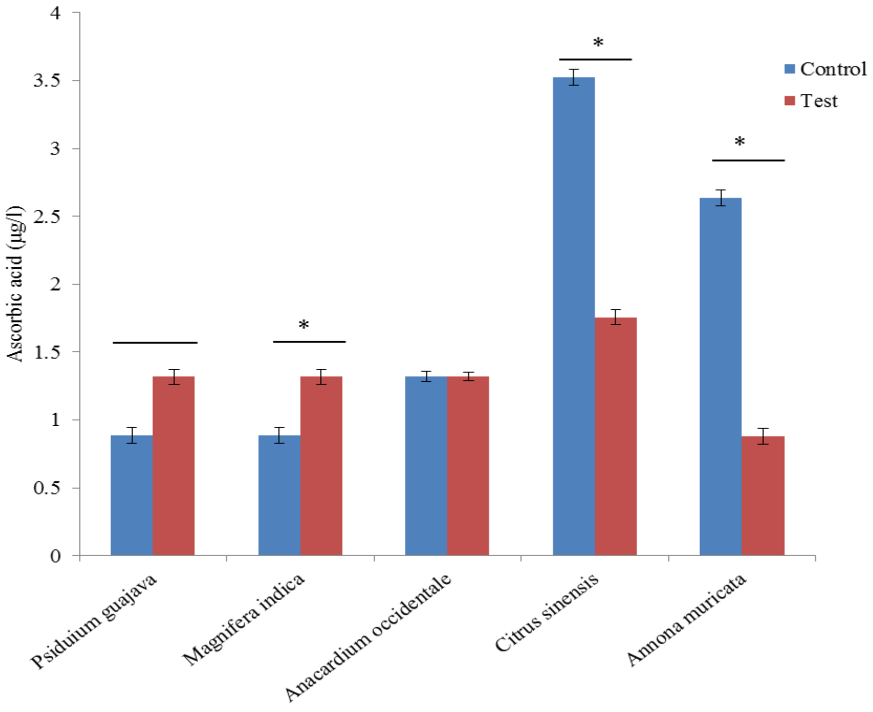

Figure 2. Effect of rock quarrying on the ascorbic acid content of five edible fruit trees

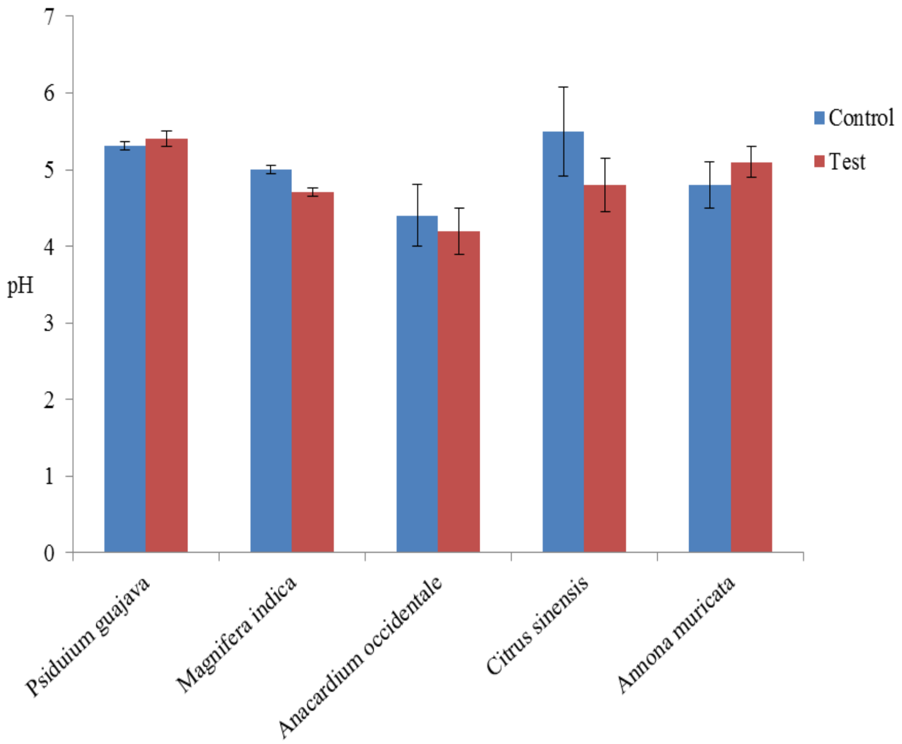

Figure 3. Effect of rock quarrying on the pH values of five edible fruit trees

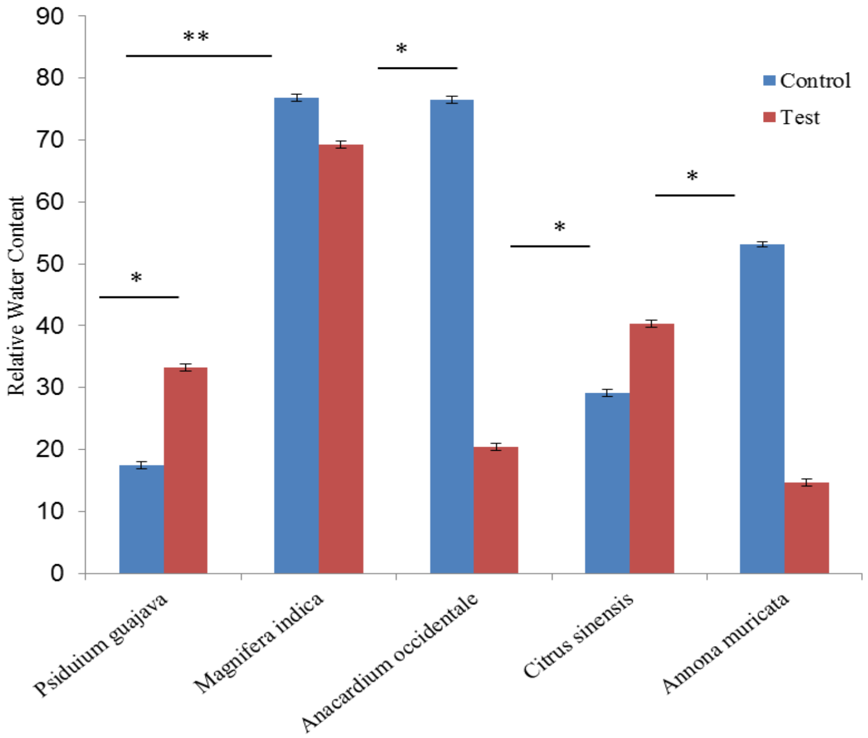

Figure 4. Effect of rock quarrying on the relative water content of five edible fruit trees

Figure 4 shows the effect of rock quarrying on the relative water content of five edible fruit trees. Relative water content ranged from 14.71% in *Annona muricata* to 69.23% in *Magnifera indica* with a mean value of 35.63% at the study site, while at the control it ranged from 17.44% in *Psidium guajava* to 76.92% in *Magnifera indica* with a mean value of 50.64%. There was significant difference in relative water content of plants in study area with the exception of *Magnifera indica*, whose relative water content was also significantly higher than those of other plants ($p<0.05$).

Figure 5 shows the effect of rock quarrying on the chlorophyll content of five edible fruit trees. There were observable significant increase ($P<0.05$) on *Psidium guajava*, *Magnifera indica* and *Citrus sinensis* when compared to their respective control. *Anacardium occidentale* and *Annona muricata* had no significant difference ($p>0.05$) when compared to their respectively control but interestingly *Anacardium occidentale* had the highest chlorophyll content in its control and study site when compared with the other studied plants.

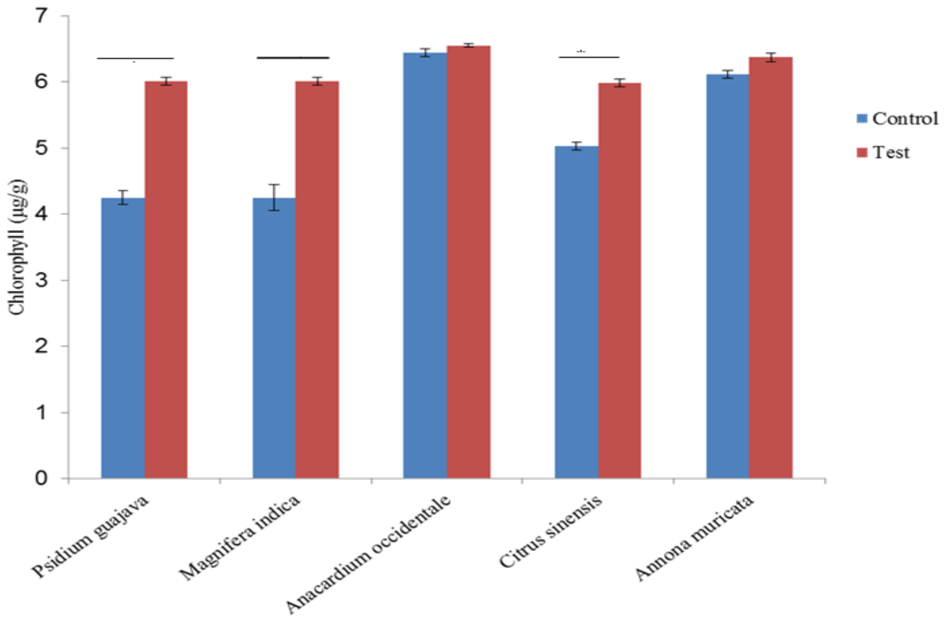

Figure 5. Effect of rock quarrying on the chlorophyll content of five edible fruit trees

4. Discussion

Ascorbic acid content varied among plants and site. Ascorbic acid content of control samples was higher than those of the study area, suggesting that concentration in plants at the study area have been altered due to the effect of pollution from quarry activities. Ascorbic acid is of key importance in plant physiology mainly due to its possession of antioxidant and cellular reductant properties and its diverse role in plant growth and development and the regulation of broad spectrum of plant cellular mechanisms against environmental stress. [7] Ascorbic acid content of plant is an important indicator of oxidative stress and therefore a crucial tolerance factor that activates defense mechanisms. [7,16,21,22] The significance of this parameter is evident in this study, as *Magnifera indica* had the highest ascorbic acid content and APTI value at both study area and control.

Another important factor in APTI is relative water content. In this study, APTI was observed to be the highest in *Magnifera indica*, which also had the highest percentage relative water content in the control and study area. This has also been observed by other researchers including. [7] High water content in plants is essential for physiological balance in plants when they are exposed to pollution. Water content in plants may however be increased by stomata occlusion in areas where ambient particulate load is high. [14,23] observed that high pH in plants was suggestive tolerance to stress. In this study, the absence of a tolerant plant may be explained by the low pH values observed. Reduction in foliar chlorophyll as observed in this study has been reported elsewhere. [1] Although. [6] had observed that chlorophyll content while being an important stress indicator may not crucially influence APTI. *Annona muricata* with the highest chlorophyll value had a lower APTI value compared with *Magniferia indica.*

APTI of the study followed the order *Magnifera indica> Citrus sinensis> Psidium guajava> Anacardium occidentale> Annona muricata.* APTI values was highest in *Magnifera indica* but was lower than values reported in other studies, including. [24,25] Temporal, spatial and species differences as well as the character and source of pollution may account for observed variations in APTI. None of the fruit trees assessed can be considered tolerant of air pollution from the study site, since no plant has APTI of up to 30. All plants were sensitive to air pollution in this study possibly as a result of close proximity to the source of pollution, and the observed high particulate matter load on the leaves. These plants may only serve as bioindicators of air pollution at the study area.

Table 1. Stomata Type and Pore Size of Leaves (Stomata Type and Pore Size**)**

SAMPLE	LEAF SURFACE	STOMATAL TYPE		PORE SIZE	
		Study Site	Control	Study Site	Control
Psidium guajava	Adaxial Abaxial	Anisocystic Anisocystic	Actinocystic Actinocystic	0.1µm 0.2 µm	0.1 µm 0.2 µm
Anarcadium Occidentale	Abaxial Adaxial	Anisocystic Anisocystic	Diacytic Diacytic	0.3µm 0.4µm	0.2µm 0.1µm
Magnefera indica	Abaxial Adaxial	Cyclocytic Cyclocytic	Anisocystic Anisocystic	0.3 µm 0.2 µm	0.2 µm 0.3 µm
Citrus sinesis	Adaxial Abaxial	Diacytic Diacytic	Cyclocytic Cyclocytic	0.1-0.2 0.4-0.6	0.1 µm 0.2 µm
Anona muricata	Adaxial Abaxial	Anisocystic Anisocystic	Cyclocytic Cyclocytic	0.2 µm 0.1 µm	0.1 µm 0.2 µm

Foliar analysis

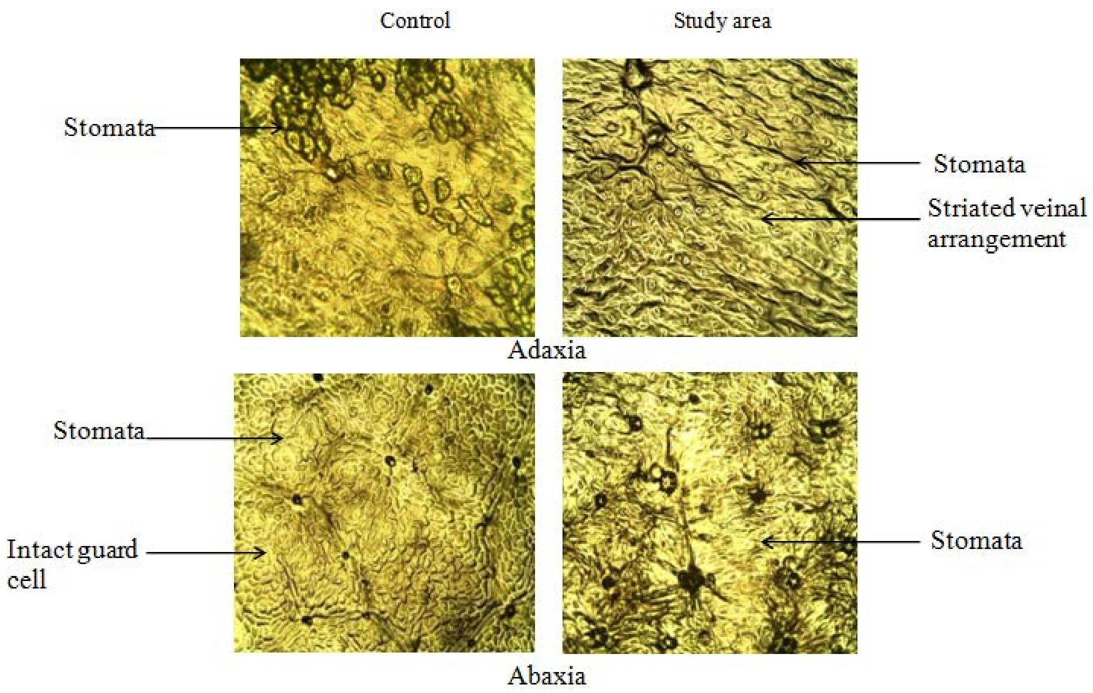

Plate 1. *Psidium guajava* (abaxial and adaxial)

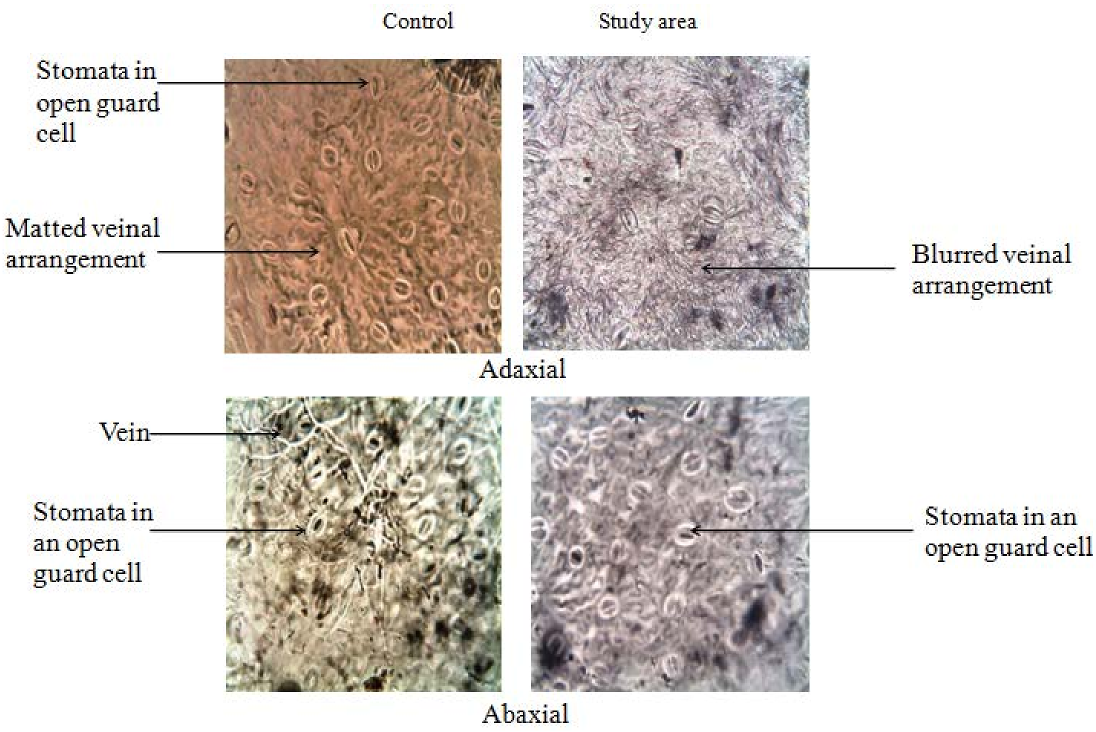

Plate 2. *Anarcadium occidentale* (abaxial and adaxial)

The adaxial (upper surface) photomicrograph of *Psidium guajava* in the control showed distinctive actinacytic stomata of size 0.1µm, with normal guard cells. The abaxial (lower surface) has a stomata pore of 0.2µm of the same stomata type. The study area sample showed evidence of stomata of anisocytic type in both the adaxial and abaxial, with the guard cells completely destroyed, with a stretched venial arrangement at the adaxial surface. These alterations were indicative of foliar destruction as a result of quarry activities. [14] (Plate 1)

In *Anacardium occidentale*, the adaxial features showed relaxed venial arrangements in the control with diacytic stomata type with pore size of 0.3 – 0.4µm. The abaxial surface showed blurred venial arrangement, while the stomata type was anisocytic at both surfaces. The adaxial surface showed evidence of distortion as most of the features seen in the control were bleached in the study area sample. These deformities were all suggestive of epidermal damage in the plants exposed to polluted environment, as has earlier been reported by [1,14]. Alteration and destruction of foliar structure affect the growth and yield of both the nutrients and phyto constituents. (Plate 2)

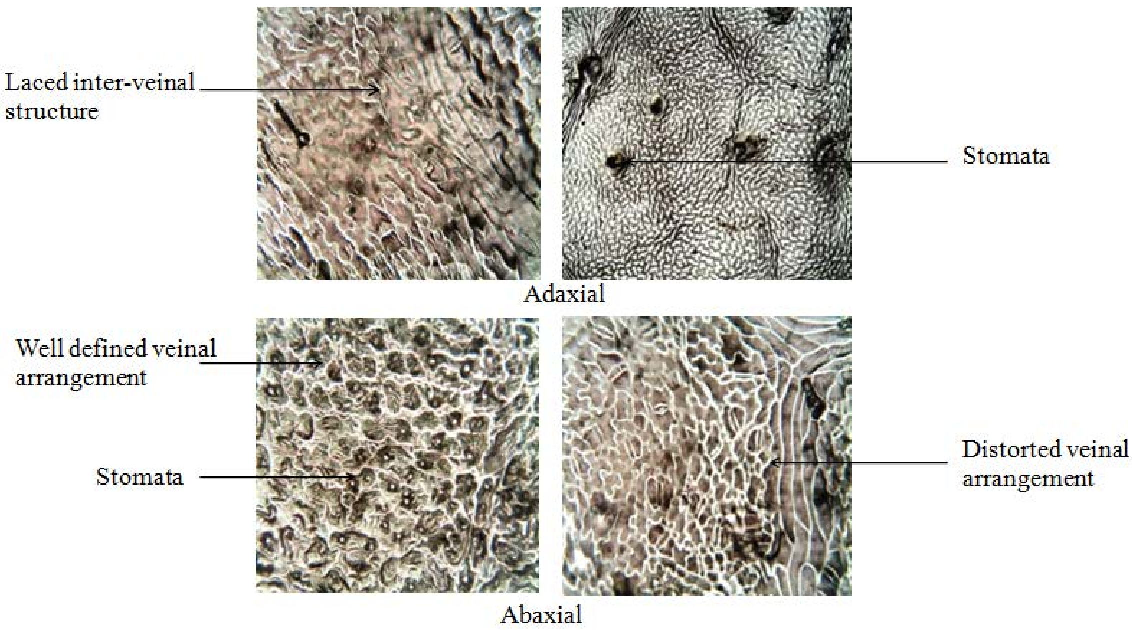

Plate 3. *Magnifera indica* (abaxial and adaxial)

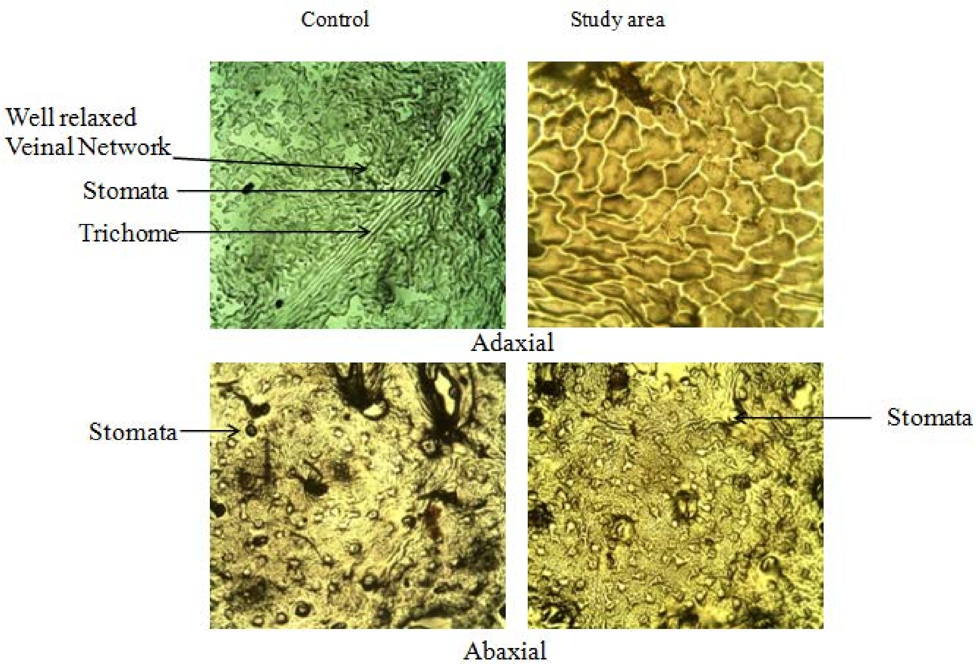

Plate 4. *Citrus sinesis* (abaxial and adaxial)

Magnifera indica revealed anisocytic stomata type in the control sample with pore sizes of 0.2µm and 0.3µm for adaxial and abaxial surfaces respectively. Inter-stomata space was very clear in both the adaxial and abaxial surfaces, with normal guard cells and progressive venial arrangement. This was not so in study area samples.

Inter-venial spaces were closed and the abaxial surface appeared squeezed as if it were under the influence of contractile force. Stomata were available but in scanty portion, unlike in the control where the stomata were densely distributed as have been reported earlier by. [15] Deformation features of abrasive nature, chlorosis and necrosis were very evident in the abaxial features of samples.

The photo-micrographic revelation was in agreement with the field physical observations, where blotches, growth retardation and defoliation were evident. These observations could only be attributed to environmental pollution resulting from quarry activities.

Plate 4 indicates that *Citrus aurantum* has cyclocytic stomata of pore size 0.1µm in the control with well arranged venial structure of progressive inter-wining at the adaxial surface. The abaxial surface had compact internal structure with prominent trichomes. On the other hand, this conspicuous feature was lacking in the study area sample, with pronounced inter-venial void guard cells. Also evident was destroyed, discontinued venial structure, all which are indications of pollution stress. [15] The destruction of the trichomes would enhance the destructive effects of small herbivores and wind on the leaves, thereby impacting negatively in foliar physiology, biochemistry and pharmacology.

The relatively wide disparity in the stomata pore size in the adaxial and abaxial of the study area and control sample could be explained by the destruction of the trichome, guard cells and inter-venial arrangement, exposing the finer internal organelles to the destructive effects of the quarrying constituents.

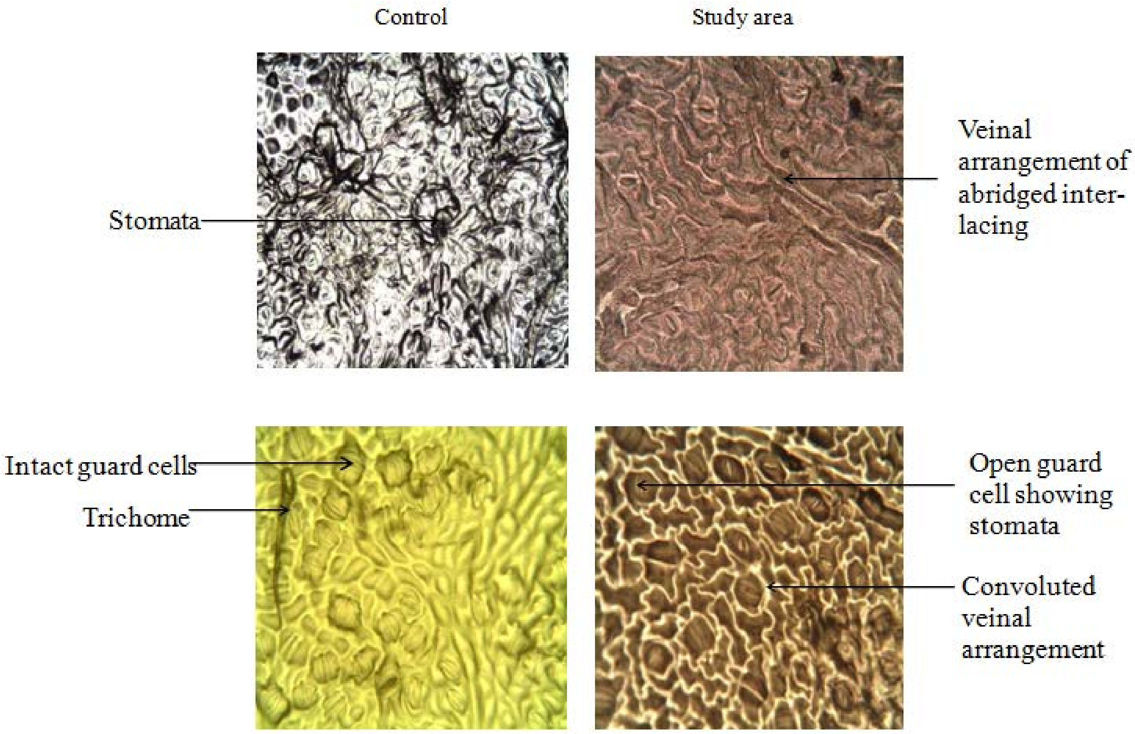

Plate 5. *Annona muricata* (abaxial and adaxial)

The foliar photomicrograph of *Anonna muricata* at the control area revealed cyclocytic stomata type of pore size 0.1µm, the adaxial and 0.2µm on the abaxial, clearly defined guard cells and a cobweb of venial arrangement. Numerous stomata are seen scattered all over the abaxial surface, giving the control sample a healthy feature needed for normal plant life. In the study area sample, these features were altered though the adaxial stomata could still be identified. In particular, the absence of guard cells was indicative of the destructive effects of pollution from quarry activities. Most gases associated with quarry activities react with water vapour and in the leaf to produce acidic solution. The distorted features as observed could be attributed to acid rain. In addition, most particulate matter in quarry area consists mostly of metals that are capable of forming hydroxides with humified air. When deposited on the leaves, increase the foliar pH, intensifying foliar pollution stress.

5. Conclusion

This study assessed APTI of five edible fruit trees growing in Ugwuele rock quarrying in Isuikwato L.G.A. The results of the study showed observable Variations in plant biochemical properties as well as the APTI. No plant was found to be tolerant of air pollution in this study. *Magnifera indica* had the highest APTI value but was sensitive to air pollution from the study site. The plants examined in this study can only be considered as bioindicators of air pollution. Clear distinctions were observed in the foliar structure of plants in study area in comparison to those at the control area. Distorted veinal arrangement and occluded among other observation offer strong evidence of stress from quarry related pollution. If these plants are consumed they can pose serious health risks since they are likely to have accumulated toxic pollutants. Moreover, it can be concluded that the nutritional value of these plants may have been compromised.

We therefore suggest that quarry operators should apply techniques that will reduce or suppress particulate load in the ambient atmosphere. Also, blasting and crushing activities should be carried out in the evening or early morning hours, when humidity is higher and air temperature lower, the residence time of particulates will be reduced. It is also very important that environmental managers consider plant tolerance and stress potentials in a quarry site.

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