

## LEAF EPIDERMAL CHANGES IN THREE COMMON CROP PLANTS FOUND IN A GAS-FLARED COMMUNITY IN OBEN VILLAGE, EDO STATE NIGERIA

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### Abstract

*The study investigated the impact of gas flaring on soil and air quality, as well as quantitative and qualitative anatomical characters of three selected plants *Musa paradisiaca*, *Carica papaya* and *Talinum triangulare* in and around Oben Flow Station. Most of these test crop plants located around the gas flare site showed signs of chlorosis, wilting and stunted growth. The interrelationship between the morphological characters of polluted and non-polluted plants was compared. The epidermal cell shape identified on adaxial and abaxial side of all the study plants varied from polygonal, elongated to wavy/sinous cell shape. Anticlinal wall varied from curved, undulated to straight in all test plants. Stomata number was more on the abaxial surface than adaxial surface in all the plants. In this study hypo-stomatic and amphi-stomatic were identified, among the species where anomocytic stomata is found in abaxial layers of all the three species Hemiparacytic in adaxial of *T. triangulare* and Anomocytic with cell inclusion distributed all around the cell layers of the Plants. The leaves of non-polluted plants grew normally while the polluted plants suffered anatomical aberration; Cell disruption in leaves was eminent.*

**Key Words:** *Flaring, pollution, plant anatomy, *Musa paradisiaca*, *Carica papaya* and *Talinum triangulare**

### Introduction

Gas flaring, as a common practice in the oil production process, is an incomplete combustion of surplus hydrocarbon congregated in an oil or gas production gas flare site (Atuma and Ojeh, 2013). Although the international community, in trying to combat the menace of global warming and other attendant effects of air pollution, proposes a complete cessation of flaring

activities, there are still areas around the world where significant gas flaring activities are still being observed.

Governments and various bodies all around the world have created tremendous awareness as to the need to protect the environment. This to a large extent has led to the enactment of regulations. In Nigeria, for example, the Harmful Wastes (Special Criminal Provision etc.) Act of 1988, provides

that it is an offence for anyone to “carry, deposit, dump, or be in possession, for the purpose of carrying, depositing or dumping, any harmful waste anywhere on Nigerian soil, inland waters or seas”. At the State levels there are laws put in place to check the menace of waste pollution; like the Pollution Prevention and Control, Miscellaneous Provision Edict 1985 of Imo State enacted to deal with the obligatory provisions by industries that produce toxic wastes to, as a matter of fact, adequately treat and dispose of such wastes.

In spite of all these laws, the problem of poor implementation has led to even more precarious situations. According to the NNPC (2010), about 65% of the gas produced in Nigeria is being flared. In the same report, gas flared was higher than 45.4% and 42.7% by 2002 and 2003 respectively, compared to the percentage of gas used (54.6% and 57.3%, respectively).

There are a number of scientific studies and technical reports on the level of impact of gas flaring in the environments, but most of these are conducted on a questionnaire basis rather than on pure scientific and empirical assessments of such environments; this is most likely because of restriction and/or lack of access to gas flare sites in this part of the world.

Apart from the threat of climatic change through the release of carbon dioxide, gas flaring results to acid rain, which contributes in the decay of building materials and paints. Acid rain resulting from flared gas also acidifies lakes and streams and destroys vegetation. In the present study, the impact of gas flaring on vegetation has been selected.

Effects on foliar anatomy of particular popular plant species found in Oben community (a very popular gas flaring site in Edo State) have been selected. Three species *Musa paradisiaca*, *Carica papaya* and *Talinum triangulare* have been chosen for the study. Measurable and qualitative variations at both cellular and histological levels characterize a very significant aspect of the total plant response to environmental fluctuations. Hence, a consideration of the harmful and often permanent changes that different categories of pollutants have on plant structure is essential to solving the major problems.

## Materials and Methods

### Study Area

The site, which is the Oben Flow Station located within OML 4 SPDC land location about 90km South of Benin City, lies between the points 50° 52' 3"E and 60° 0' 39"N; and is bordered by Oben, Iguelaba, Ikobi and Obozogbe-Nugu communities in Orhionmwon Local Government Area of Edo State, Nigeria. The facility shares a common boundary with Pan Ocean's OML-98. The occupational structure of the host communities is typically characterized by farming, trading, and transportation. The major food crops cultivated in the area are yams, cassava, *M. paradisiaca* and cocoyam. However, cash crops such as rubber and oil palm, as well as citrus fruits are also cultivated.

The entire project site was radially demarcated into 5 portions, using the gas flare stack as reference point. Air quality monitoring as well as collection of soil samples for analyses was carried out at 50m, 100m, 150m, 200m, and 250m from the gas flare site. The control site was a farmland located exactly 1 km from the

flare site. Air quality parameters were measured with Aeroqual Series 200® at the different locations and between 10 and 11 am. The Aeroqual Air Quality Kit was placed at shoulder level to take reading. Soil samples collected from designated radial points from the station were analyzed for temperature, conductivity, total organic carbon, Zn, Fe and Cu using standard procedure (APHA, 1985 and AOAC, 2005). Following the methods of Cheesebrough (2001), these soils were also investigated for total heterotrophic as well as hydrocarbon-utilizing bacteria and fungi.

For determination of selected foliar epidermal characteristics, three crop species namely *M. paradisiaca*, *C. papaya* and *T. triangulare* were selected. These three crop plants were so selected because they were the closest (within 100m) to the gas flare site. Those plants collected from this demarcated region were labeled polluted, were as those collected beyond 1km from the gas flare site were labelled “unpolluted”.

Voucher specimens were deposited at the University of Lagos Plant Anatomy Laboratory, Lagos State, Nigeria; and were investigated by means of light microscopy following the methodologies modified from William (2000); Ahmad *et al.*; (2005); Ikhajiagbe *et al.* (2007). Slides were examined with top-view light microscopes at x100 and x400. All measurements in light microscope (LM) were made using a calibrated eyepiece micrometer with ×40 objective. From each species 5 cells and stomata were randomly selected for measurement. Stomata index was calculated using the formula:

$$\text{Stomata index} = \frac{\text{Stomata number} \times 100}{\text{Cell number per unit area} + \text{stomata number}}$$

Qualitative characterization of the foliar epidermal surfaces was also carried out, including investigations of stomata type, epidermal type and cell shape patterns. Quantitative characters measured were epidermal cell length and width, number of epidermal cells per field of view, epidermal cell wall thickness and stomata length. All measurements were made using the light microscope at magnification (x 400) using micrometer eyepiece.

## Results and Discussion

The physicochemical condition of the soil collected from radially demarcated distances from the gas flare stack showed minimal differences in soil temperature (29.2 – 40.5 °C) (Table 1). There were no significant changes ( $p > 0.05$ ) in soil moisture content (19.7 – 39.4%). Constant and continuous heating does not only decreases or reduces the soil moisture but indeed affects the photoperiodicity needed for flowering plants and fruiting. pH of the soil measures the acidity and alkalinity of the soil. Results showed pH increases at distances away from the flare site, implying lesser acidity along this direction; this compares with the findings of Nwaugo *et al.* (2006); Nwaogu and Onyeze (2010); Ezeigbo *et al.* (2013); Ubani and Onyejekwe (2013).

The acidity of the soil samples in Oben could be attributed to flaring which produces acidic oxides of sulfur, nitrogen, carbon, which dissolves in rain water to produce or form acid rain. Increased soil acidity can create biochemical situations that can be detrimental to plants and soil microorganisms (Ezeigbo, 2013; Nwaogu and Onyeze, 2010). One of such effects is the

inhibition in plants' capacity to take in cations.

Very high electric conductivity was recorded at 50m from the gas flare site (264.2µS/cm). However, soil conductivity beyond 150m from the gas flare site was comparable with that at the control site (p>0.05). Soil Fe content was

higher in the control (61.822 mg/kg) than around the gas-flared site.

Higher total heterotrophic as well as hydrocarbon-utilizing (Fig. 1) bacteria and fungi were obtained in the control site compared to soils obtained from gas flare site.

Table 1: Physicochemical condition of soil obtained from selected distances from the gas flare site

	Distance from gas flare site					
	50m	100m	150m	200m	250m	Control
Soil Temperature (°C)	30.9 <sup>b</sup>	29.2 <sup>b</sup>	40.5 <sup>a</sup>	37.3 <sup>ab</sup>	35.6 <sup>a</sup>	27.3 <sup>b</sup>
Moisture (%)	21.4 <sup>a</sup>	20 <sup>a</sup>	18.3 <sup>a</sup>	19.8 <sup>a</sup>	19.7 <sup>a</sup>	39.4 <sup>a</sup>
Ph	4.97 <sup>b</sup>	5.83 <sup>ab</sup>	5.97 <sup>ab</sup>	6.65 <sup>a</sup>	6.39 <sup>a</sup>	6.89 <sup>a</sup>
Soil Temperature (°C)	30.9 <sup>b</sup>	29.2 <sup>b</sup>	40.5 <sup>a</sup>	37.3 <sup>ab</sup>	35.6 <sup>a</sup>	27.3 <sup>b</sup>
Moisture (%)	21.4 <sup>a</sup>	20 <sup>a</sup>	18.3 <sup>a</sup>	19.8 <sup>a</sup>	19.7 <sup>a</sup>	39.4 <sup>a</sup>
Elec. Cond. (µS/cm)	564.2 <sup>a</sup>	72.9 <sup>bc</sup>	110.4 <sup>b</sup>	31.2 <sup>d</sup>	55.1 <sup>cd</sup>	52.8 <sup>cd</sup>
Total Org. Matter (%)	0.47 <sup>a</sup>	0.74 <sup>a</sup>	0.99 <sup>a</sup>	1.09 <sup>a</sup>	0.78 <sup>a</sup>	0.92 <sup>a</sup>
Pb (mg/kg)	<0.001 <sup>a</sup>	<0.001 <sup>a</sup>	<0.001 <sup>a</sup>	<0.001 <sup>a</sup>	<0.001 <sup>a</sup>	<0.001 <sup>a</sup>
Cu (mg/kg)	0.218 <sup>a</sup>	0.034 <sup>a</sup>	0.072 <sup>a</sup>	0.196 <sup>a</sup>	0.823 <sup>a</sup>	1.142 <sup>a</sup>
Zn (mg/kg)	9.462 <sup>bc</sup>	12.843 <sup>ab</sup>	0.516 <sup>c</sup>	22.183 <sup>a</sup>	9.768 <sup>bc</sup>	20.771 <sup>a</sup>
Fe (mg/kg)	4.095 <sup>b</sup>	11.716 <sup>b</sup>	<0.001 <sup>b</sup>	46.21 <sup>b</sup>	39.125 <sup>b</sup>	651.822 <sup>a</sup>
Exch. Na (mg/kg)	14.378 <sup>a</sup>	13.296 <sup>a</sup>	7.885 <sup>a</sup>	12.957 <sup>a</sup>	<0.001 <sup>b</sup>	9.154 <sup>a</sup>
Exch. K (mg/kg)	2.125 <sup>b</sup>	3.183 <sup>b</sup>	35.571 <sup>a</sup>	4.982 <sup>b</sup>	29.621 <sup>a</sup>	<0.001 <sup>b</sup>
Exch. Mg (mg/kg)	1.784 <sup>a</sup>	1.927 <sup>a</sup>	3.363 <sup>a</sup>	2.003 <sup>a</sup>	2.268 <sup>a</sup>	2.217 <sup>a</sup>
Exch. Ca (mg/kg)	77.102 <sup>b</sup>	52.141 <sup>c</sup>	95.712 <sup>a</sup>	14.712 <sup>d</sup>	105.725 <sup>a</sup>	25.329 <sup>d</sup>

Means on the same row with similar alphabetic superscript do not differ significantly from the other (p>0.05).

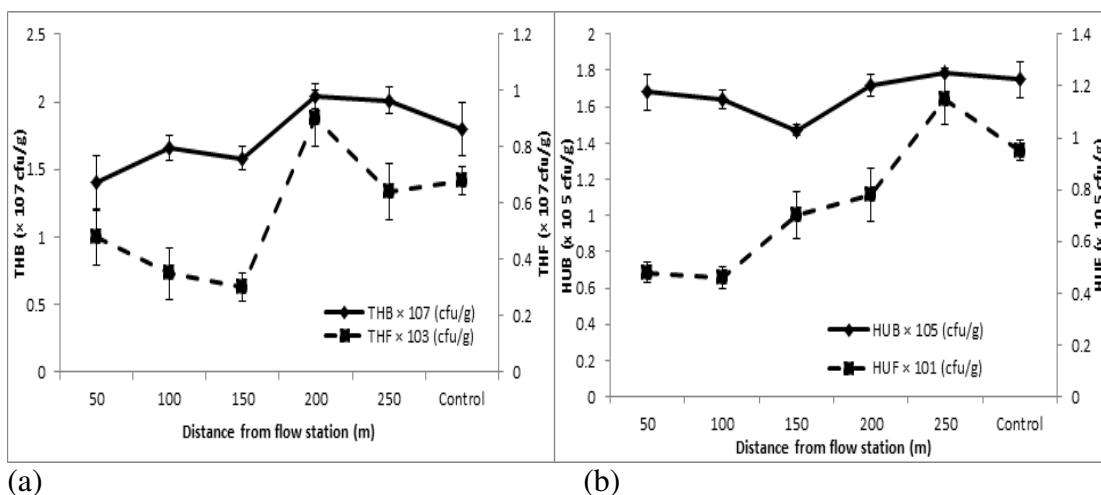


Fig. 1: (a) Total heterotrophic bacteria (THB) and fungi (THF) and (b) Hydrocarbon-utilizing bacteria (HUB) and fungi (HUF) of soils obtained from gas flare site.

Air quality parameters measured within the different locations is presented in Table 2.  $SO_x$  content in air around the gas flare site (0.58 – 3.80 ppm) were generally above DPR Limits (0.01 – 0.1 ppm).  $SO_x$  at the control site were 0.02 ppm. The values recorded for  $NO_x$ , CO, VOC,  $H_2S$  and  $O_3$  around the gas flare site and the control sites were generally below DPR statutory limits (Table 2).

Table 2. Air quality parameters measured with Aeroqual Series 200® at the difference locations and between 10 and 11 am

Parameters	DPR Limits	Radial dimensions within oil installation					Control
		50m	100m	150m	200m	250m	
$SO_x$ (ppm)	0.01-0.10	3.36 <sup>a</sup>	3.80 <sup>a</sup>	3.80 <sup>a</sup>	0.52 <sup>b</sup>	0.58 <sup>b</sup>	0.02 <sup>c</sup>
$NO_x$ (ppm)	0.04-0.06	0.024 <sup>a</sup>	0.016 <sup>a</sup>	0.016 <sup>a</sup>	0.021 <sup>a</sup>	0.021 <sup>a</sup>	0.012 <sup>a</sup>
CO (ppm)	10.00-20.00	3.72 <sup>b</sup>	19.52 <sup>a</sup>	19.52 <sup>a</sup>	6.72 <sup>b</sup>	6.72 <sup>b</sup>	1.86 <sup>b</sup>
VOC ( $\mu g/m^3$ )	160.00	21.82 <sup>bc</sup>	97.91 <sup>a</sup>	97.93 <sup>a</sup>	41.10 <sup>b</sup>	41.05 <sup>b</sup>	3.41 <sup>c</sup>
$H_2S$ ( $\mu g/m^3$ )	0.010	0.001 <sup>a</sup>	0.004 <sup>a</sup>	0.004 <sup>a</sup>	0.002 <sup>a</sup>	0.002 <sup>a</sup>	0.001 <sup>a</sup>
$O_3$ ( $\mu g/m^3$ )	0.010	<0.001 <sup>a</sup>	<0.001 <sup>a</sup>	<0.001 <sup>a</sup>	<0.001 <sup>a</sup>	<0.001 <sup>a</sup>	<0.001 <sup>a</sup>

DPR Dept. of Petroleum Resources, Nigeria

Table 3. Qualitative taxonomic character presentation of common economic crops around the gas-flared site compared to the control (AB abaxial, AD adaxial)

Characters	Non-Polluted <i>Musa paradisiaca</i>	Polluted <i>Musa paradisiaca</i>	Non-Polluted <i>Carica papaya</i>	Polluted <i>Carica papaya</i>	Non-Polluted <i>Talinumtriangulare</i>	Polluted <i>Talinumtriangulare</i>
Epidermal cell shape(AB)	Rectangular	Rectangular	Sinuus/ Wavy	Sinuous/Wavy	Sinuous/Wavy	Sinuous/Wavy
Epidermal cell shape (AD)	Polygonal/ Rectangular	Polygonal/ Rectangular	Sinuus/ Wavy	Sinuous	Sinuous/Wavy	Sinuous/Wavy
Epidermal cell inclusion	Absent	Present	Present	Present	Absent	Absent
Anticlinal wall pattern AB	Straight	Straight	Undulate	Slightly curved	Undulate	Undulate
Anticlinal wall pattern AD	Straight	Straight/Curved	slightly curved	Undulate	Undulate	Undulate
Stomatal type AB	Anomocytics	Anomocytic	Anomocytics	Anomocytic	Hemiparacytic	Hemiparacytic
Stomatal type AD	Absent	Anomocytic	Absent	Absent	Hemiparacytic	Hemiparacytic

Table 4. Quantitative foliar anatomic character presentation of common economic crops around the gas-flared site compared to the control (AB abaxial, AD adaxial, mean±SE)

Characters	Non-Polluted <i>Musa paradisiaca</i>	Polluted <i>Musa paradisiaca</i>	Non-Polluted <i>Carica papaya</i>	Polluted <i>Carica papaya</i>	Non-Polluted <i>Talinumtriangulare</i>	Polluted <i>Talinumtriangulare</i>
Epidermal cell number/mm <sup>2</sup> – AB	106.4±1.52	181±4.19	186.8±2.84	34.2±1.56	29.2±0.55	186.4±1.75
Epidermal cell number/mm <sup>2</sup> – AD	40.6±1.72	147.2±2.1	165.2±4.55	14.8±0.42	19.8±1.95	194.6±10.52
Epidermal cell length (µm) AB	8.1±0.76	7.05±0.27	3.95±0.17	14.3±0.99	14.25±0.46	3.45±0.22
Epidermal cell length (µm) AD	11.3±0.0.18	7.05±0.08	3.65±0.1	16.08±0.83	14.2±0.51	4.1±0.97
Epidermal cell width (µm) AB	1.7±0.07	2.6±0.09	2.3±0.19	5.45±0.31	6.6±0.28	2.4±0.05
Epidermal cell width (µm) AD	3.35±0.06	2.7±0.03	2.45±0.11	7.85±0.13	6.05±0.14	4±0.14
Epidermal thickness (µm) AB	0.25±0	0.25±0	0.25±0	0.13±0	0.25±0	0.63±0
Epidermal thickness (µm) AD	0.38±0	0.25±0	0.125±0	0.25±0	0.5±0	0.25±0
Stomata number/mm <sup>2</sup> AB	6.02±0.61	4.45±0.27	67.21±2.47	37.45±0.97	7.21±0.76	26.21±1.19
Stomata number/mm <sup>2</sup> AD	Absent	Absent	Absent	8.5±0.57	4±0.5	8.5±0.57
Stomata length (µm) AB	1.9±0.05	2.55±0.04	1.6±0.02	2.1±0.03	2.35±0.03	3.6±0.02
Stomata length (µm) AD	Absent	2.56±0.05	Absent	2.56±0.57	2.55±0.07	2.5±0.57
Stomata width (µm) AB	0.75±0	1.1±0.03	1.33±0.41	0.45±0.02	0.75±0	0.55±0.01
Stomata width (µm)AD	Absent	0.46±0.03	Absent	0.7±0.04	0.75±0	0.7±0.04
Stomata Index AB	5.36%	2.37%	26.46%	52.24%	20.46%	12.35%
Stomata Index AD	Absent	Absent	Absent	18.52%	20.00%	18.52%

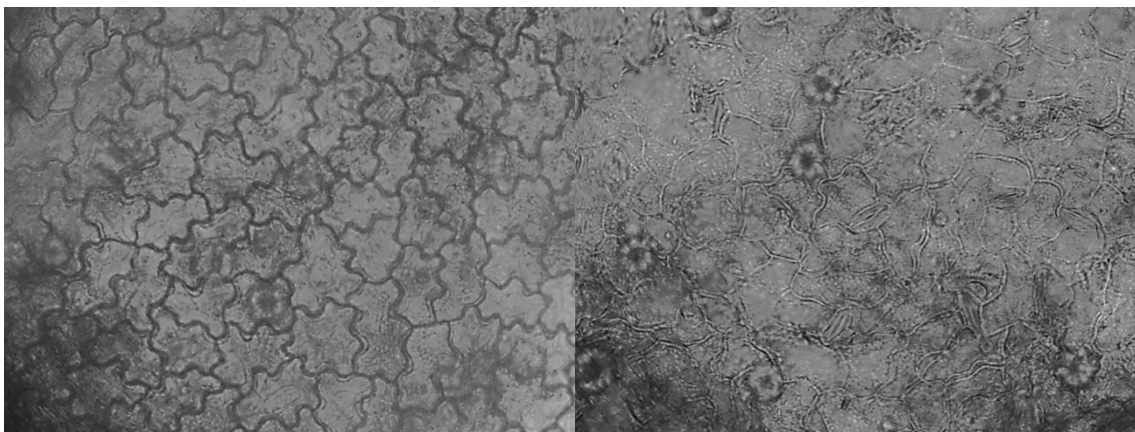


Plate 1: Polluted Upper x400 and lower x400 leaf epidermis of *Carica papaya*

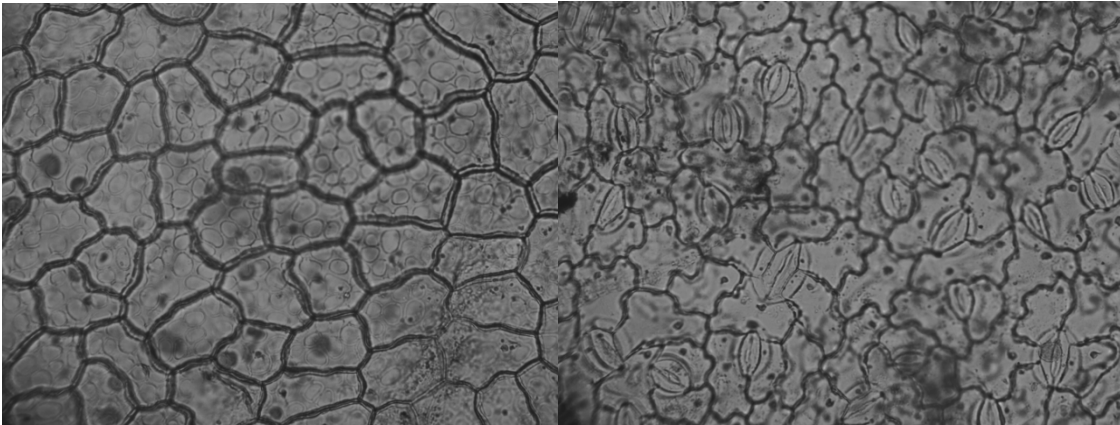


Plate 2: Non-Polluted Upper x400 and lower x400 leaf epidermis of *Carica papaya*

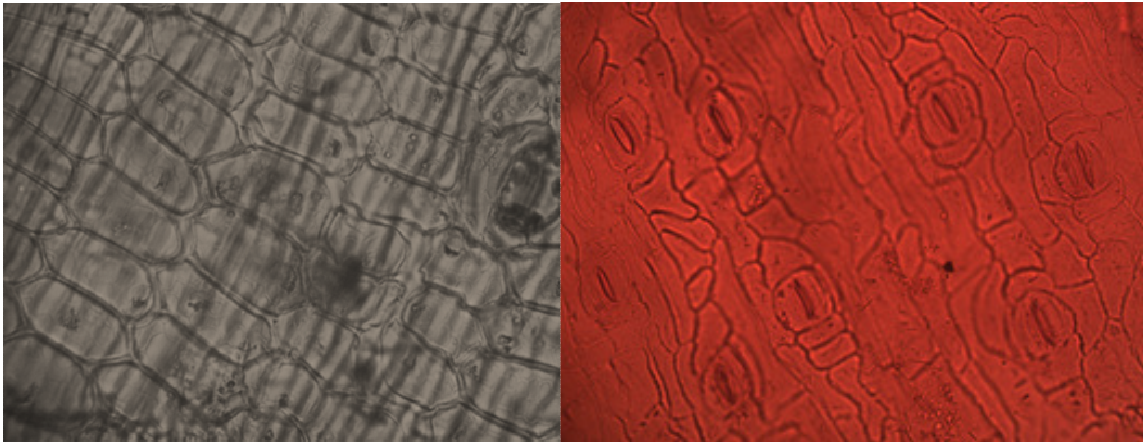


Plate 3: Polluted Upper x400 and lower x400 leaf epidermis of *Musa paradisiaca*

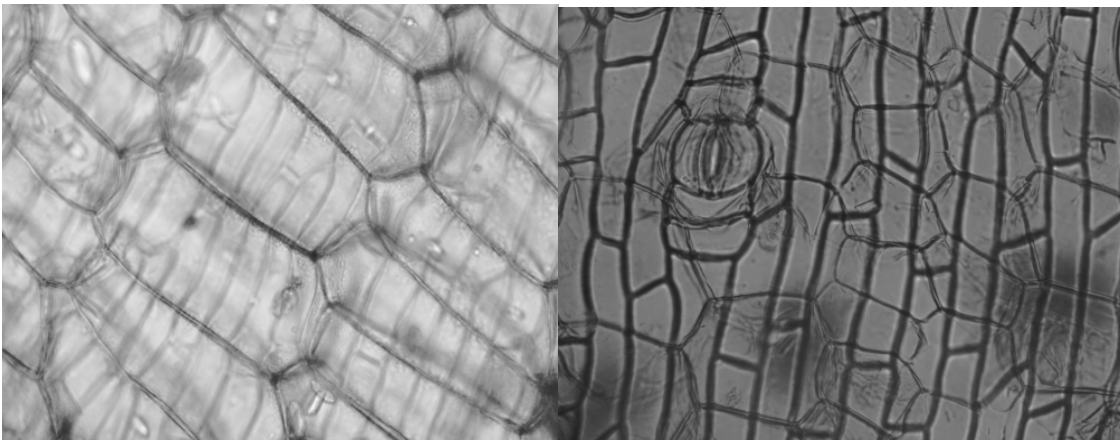


Plate 4: Non-Polluted Upper x400 and lower x400 leaf epidermis of *Musa paradisiaca*

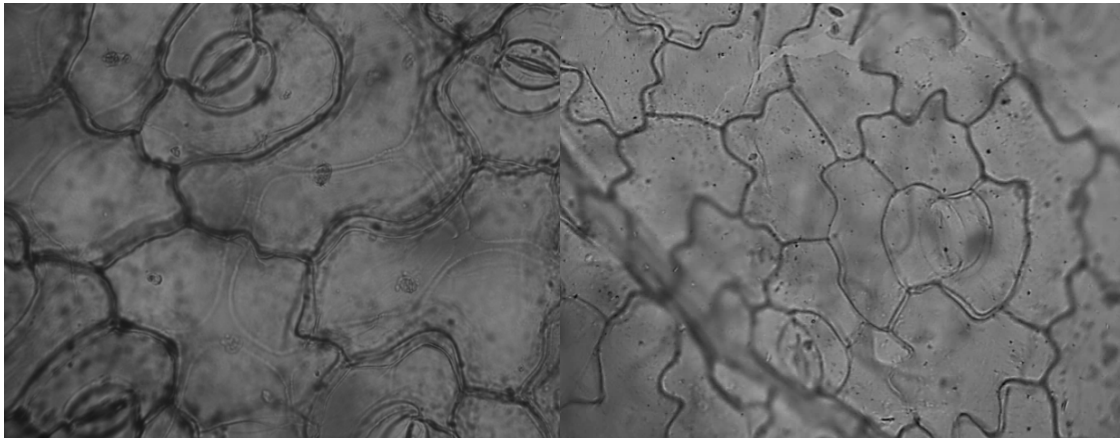


Plate 5: Polluted Upper x400 and lower x400 leaf epidermis of *Talinum triangulare*

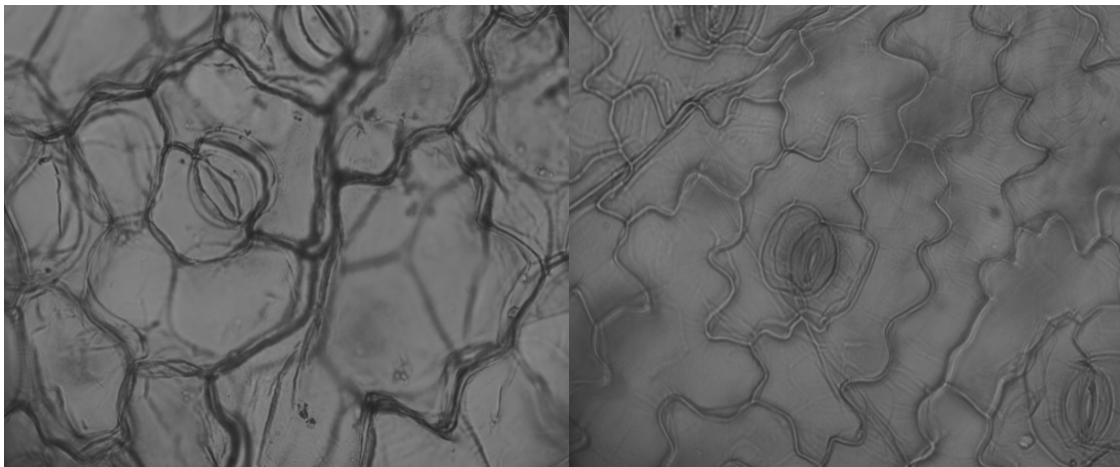


Plate 6: Non-Polluted Upper x400 and lower x400 leaf epidermis of *Talinum triangulare*

Table 3 and 4 shows the interrelationship between the morphological characters of polluted and non-polluted test plants. The epidermal cell shapes were identified on adaxial and abaxial sides of leaves. Among the test plants, these vary from polygonal, elongate to wavy/sinous cell shape. Anticlinal wall varied from curved, undulated and straight in all the three (3) studied plants. The degree of curvature in abaxial and adaxial layers of *T. triangulare* describes differences that exist between them and ultimately the effect of gas flaring on the anatomy of the plant. Stomata number was more on

the abaxial surface than adaxial surface in all the plants. In this study hypo-stomatic and amphi-stomatic were identified among the species where anomocytic stomata is found in abaxial layers of all the three species Hemiparacytic in adaxial of *T. triangulare* and Anomocytic with cell inclusion distributed all around the cell layers of the Plants. There were more epidermal cell numbers per squared millimeter on both adaxial and abaxial surfaces in the polluted *M. paradisiaca* and *T. triangulare* plants than in the unpolluted ones. However, for the *C. papaya* plant, more epidermal cells



existed in the non-polluted plant than in the polluted (Table 4).

Plants detect and react to environmental stresses in a number of ways, including anatomical adaptations. In the present study, the extent of curvature in anticlinal wall of test plants and closeness of plants to gas flaring site has showed plants negative growth response to gas flares. These findings are in accordance with previous reports of Ghose *et al.* (1980). Plant leaves were grossly affected by the flare. The effect was seen in the cell almost fading; this can be as a result of pollution. A comparison of the cells in the plates is visible (Plates 1 – 6). There were distortions or reduction in the number of stomata per unit area of the leaf. The waviness of anticlinal wall shows a strong degree of curvature in *C. papaya* as shown in Plates 1 and 2; this suggests the effects of the pollution in the soil, perhaps by gas flare deposits or other petroleum-related contaminants. This further confirms earlier report of Sharma *et al.*, (1980). *M. paradisiaca* anticlinal wall shows differences straight in the non-polluted plant, while it was straight-to-curve in the polluted.

The anticlinal wall of the *C. papaya* at the abaxial was rather distinct. It was undulated in non-polluted, but slightly curved polluted. The leaves of non-polluted plants grew normally while the polluted plants suffered anatomical disfigure. Cell disruption in leaves was eminent. Plant and animal species differ in their sensitivity to gas flaring. These differential responses have been further elucidated in this study.

### Conclusion

The effects of gas flaring on plant growth and development is far-reaching.

In the study, emphasis was on leaf epidermal changes. This is even more predicated on the fact that plant productivity relies, to a very large extent, on the activities of leaves around the dermal region, including stomatal opening and closure, implicated during plant-water relations and for CO<sub>2</sub> intake for photosynthesis. Understanding these effects has proven difficult, due in part to many complex interactions. Nevertheless, quantitative and qualitative histological and cellular changes often represent one aspect of the total plant response.

### Acknowledgments

The authors are grateful to the indigenes of Oben Village for allowing us access to gas-flared areas of the community.

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