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RESEARCH ARTICLE

GROWTH AND ANATOMICAL RESPONSES OF ABELMOSCHUS ESCULENTUS (L.) MOENCH AS INFLUENCED BY DETERGENT EFFLUENT

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Abstract

Background: Considering the growing focus on sustainable agriculture and worries about the detrimental impact of widespread industrial (detergent) effluent use on plant growth. Abelmoschus esculentus (L.) Moench growth and anatomical response were studied.

Methodology: The plants were grown on soil that was not contaminated (0%) and soil that had been exposed to various effluent treatments (5, 10, 15, 20, and 25%). The data generated were subjected to analysis of variance (ANOVA).

Results: The growth of the plants was found to be negatively impacted by the effluent though at 5% concentration growth and anatomical characters were enhanced. The result of the study showed that at 5% effluent concentrations, the plant height are of same value with the control plants after 14 weeks of planting (33.41±2.67), while stem girth and number of leaves (6.47±0.39 and 6.66±0.54 respectively) were comparatively higher than on control plants (5.09±0.28 and 5.83±0.44 respectively), for fresh and dry weight of shoot and root, plants treated with effluent concentration of 15%, 20% and 25% were the most affected when compared with the control plants. This indicates that plants treated with effluent concentration above 5% were largely affected.

Conclusion: The effluent has an impact on the anatomical structures of the plants, according to the anatomical analysis of the plants treated with effluent concentration. On both the adaxial and abaxial surfaces, there were noticeably fewer epidermal cells as the concentration increased. This study found that detergent effluent, particularly at higher concentrations of 15% and 25%, had a substantial negative impact on the growth and anatomical structure of okra while at lowest (5%) concentration of the effluent displayed a positive impact on growth and anatomical responses of the okra plant. Thus, at low concentrations, industrial effluent can be non-toxic to plant growth.

Keywords: Abelmoschus esculentus, detergent effluents, stomata, fruits, epidermal cell

INTRODUCTION

The poor handling of the enormous amounts of trash produced by diverse anthropogenic activities is one of the most serious issues facing emerging nations. The hazardous disposal of these materials into the environment becomes more of a challenge. The most damaged aquatic bodies are freshwater reservoirs in particular. Due to this, these natural resources are frequently inappropriate for use in primary or secondary production (Fakayode, 2005).

The primary causes of pollution in all settings are industries. Various degrees of pollutants may be released into the environment either directly or indirectly through public sewer lines, depending on the type of industry. Employee sanitary waste, manufacturing process wastes, wash waters, and comparatively uncontaminated water from heating and cooling operations are only a few examples of wastewater generated by industries (Glyn *et al.*, 1996).

In Nigeria, almost all industrial effluent waters are discharged untreated onto land or into water bodies. Even in locations where some treatment facilities exist, their operation is subpar. (Arjun *et al.*, 2013) These refuse waters contaminate the water resources and, ultimately, the agricultural land. The growth and yield of crops, as well as the health of the soil, are reduced when producers use polluted water for irrigation of cultivated land (Nand and Kaul, 1994). Most urban farmers in Nigeria divert treated or untreated effluent-contaminated water to their farmlands for irrigation, particularly vegetable farms, to meet the rising demand for fresh vegetables in the country (Uaboi-Egbenni *et al.*, 2009; Fatoba *et al.*, 2010). However, such waters should be used with caution to irrigate vulnerable and herbaceous crops, such as vegetables (Ogunkunle *et al.*, 2013). Quantitatively and qualitatively, crops and vegetables grown in agricultural fields irrigated with textile, detergent, and other effluent-contaminated waters are negatively impacted.

Abelmoschus esculentus (L.) Moench is a member of the Malvaceae family and the Abelmoschus genus and it is one of the most widely known and utilised species of the Malvaceae family (Naveed, *et al.*, 2009) and it is an economically significant vegetable crop grown in tropical and sub-tropical regions of the world (Oyelade *et al.*, 2003; Saifullah, 2009).

Okra can be cultivated on a wide variety of soils, but high yields are achieved on well-drained, fertile soils with adequate organic matter (Akinyele, 2007). The crop is extensively grown throughout the year in tropical regions. Okra is a nutritious vegetable that plays an essential role in the human diet and is a good source of protein, carbohydrates, vitamins, calcium, potassium, enzymes, and total minerals, which are frequently deficient in the diets of developing nations (Ahmed *et al.*, 1995). In addition to curing ulcers and relieving haemorrhoids, it has been reported to have medicinal value. (Ahmed *et al.*, 1995) Okra has medical applications as a plasma replacement or blood volume expander, as well as in the treatment of genito-urinary disorders, spermatorrhea, and chronic dysentery. The purpose of this investigation was to examine the growth and morphological response of *Abelmoschus esculentus* to detergent effluent.

MATERIALS AND METHOD

Experimental Site

The research was conducted at the Greenhouse of the Botanical Garden at the Federal University of Kashere is located at Latitude 9^{0}52'N and Longitude 11^0 0'E in Akko, Akko Local Government, Gombe State (Kolawole *et al.*, 2021).

Source of seeds, detergent effluents, and soil

The okra seed variety (Clemson spineless) was purchased from a local market in Bauchi, while the detergent effluent was obtained from Nasco Household Limited in Jos. The loamy topsoil which is dark brown to brownish grey in colour, absorbs water moderately and usually maintain its porosity was used in this investigation and the soil was obtained from the Local farmers in Kashere, Gombe state.

Experimental Design and Soil preparation

The experiment was laid out in a completely randomised design (CRD) with six treatments and five replicates each. To obtain varied homogenous mixtures/concentrations, the topsoil was sieved with a 2 mm mesh and a 5 kg weighed, then mixed by hand with varying volumes of effluent. Thirty labelled, perforated nursery plastic pots were filled with 5kg of the soil mixture to provide five pots for each concentration. The concentration was determined in the following manner:

- 0 millilitres of effluent plus 5 kilogrammes of sediment equals the control
- 250 ml of effluent plus 5 kg of sediment equals a concentration of 5% concentration
- 500 ml of effluent plus 5 kilogrammes of sediment equals a 10% concentration
- 750 ml of effluent plus 5 kilogrammes of sediment equals a 15% concentration
- 1000 ml of effluent plus 5 kilogrammes of sediment equals a 20% concentration
- 1250 ml of effluent plus 5 kilogrammes of sediment equals a 25% concentration

Planting and watering

The okra seeds were planted in a randomly manner and spaced from each other on each container. Every three days, both the detergent effluent treated seeds and the control seeds were sprayed with water. During the course of 14 weeks, 200 ml of the effluent was applied to each container, excluding the control, at each watering.

Weeding

Frequent weeding by hand was performed to prevent competition between weeds and desired plants.

Measurement of Growth parameters

Measurements started four (4) weeks after planting and lasted for fourteen (14) weeks with an interval of two weeks. The morphological parameters measured include Plant Stem Height, Leaf Length and Breadth, Stem Girth and Leaf Area. The stem height, leaf length and breadth were measured with a standard meter rule while the stem girth was measured with an Electronic Digital Caliper (Titan 23175 model). The leaf Area was calculated according to Pearcy *et al.* (1989) and Kolawole *et al.* (2018).
Leaf Area = (L X B) K Where L= length of leaf, B=maximum width and K= 0.72

ANATOMICAL STUDIES

Materials and procedure

Concentrated nitric acid or trioxonitrate (v) acid, distilled water, aqueous safranin solution, and diluted glycerine, petri dishes, forceps, slide, cover slip, and microscope (Kolawole *et al.*, 2017).

Method

Okra leaves were collected for anatomical investigation. Leaf cuticles were macerated in nitric acid or trioxonitrate (v) acid, then rinsed in distilled water, stained in 1% aqueous safranin solution, and mounted in diluted glycerine (Dutta 2003; Kolawole *et al.*, 2017). Using a fine grade camel hairbrush, epidermal layers were meticulously removed from the surface of leaf samples. Both the adaxial and abaxial surfaces of the leaves were prepared, labelled, and examined at various magnifications using an Olympus light microscope. Observations were recorded from 20 fields of view for each epidermal surface (abaxial and adaxial), epidermal cell shape, anticlinal cell wall pattern and epidermal cell size of the investigated plant species (Khatijah and Zaharina 1998, Adedeji 2004, Chukwuma *et al.*, 2014).

Statistical Analysis

The data were subjected to analysis of variance (ANOVA) and where significant differences was observed, the means was separated using Duncan's multiples range test at $P \leq 0.05$. All statistical analysis was conducted using the SPSS statistical package, version 20.0 (Property of International Business Machine Corp IBM).

RESULTS

This investigation revealed that the responses of *Abelmoschus esculentus* grown in different concentrations of industrial effluent appear to be dose-dependent with regard to plant height. The results at 4, 6, 8, 10, 12 and 14 WAP (Weeks after Planting) revealed that plant height varied among the different concentration levels, with the 5% level of contamination producing the tallest plants and the 15% concentration producing the shortest. Statistically, the 5% concentration level produced substantially greater height at the 0.05 probability level than the other concentration levels. The heights of the control plants did not differ significantly from the remainder of the concentration over the course of the weeks (Table 1).

Table 1: Plant height (cm) of Okra treated with detergent effluent.

Effluent concentration (%)	4 WAP	6 WAP	8 WAP	10 WAP	12 WAP	14 WAP
Control	11.41 ± 1.82^{a}	14.75 ± 1.37^{a}	19.50 ± 2.25^{b}	24.91 ± 3.38^{b}	29.16 ± 4.45^{a}	33.41 ± 4.46^{a}
5	11.75 ± 0.38^{a}	14.08 ± 0.22^{a}	21.16 ± 0.44^{a}	27.25 ± 1.66^{a}	30.83 ± 2.12^{a}	33.41 ± 2.67^{a}
10	11.13 ± 1.35^{a}	13.58 ± 2.00^{ab}	17.21 ± 5.94^{bc}	22.33 ± 3.57^{bc}	26.00 ± 4.36^{b}	28.83 ± 5.02^{ab}
15	9.25 ± 1.42^{b}	11.41 ± 2.02^{b}	14.56 ± 2.52^{b}	18.00 ± 2.25^{c}	21.00 ± 2.12^{c}	23.41 ± 2.79^{c}
20	10.00 ± 0.80^{ab}	11.66 ± 1.10^{a}	14.98 ± 2.11^{b}	19.08 ± 2.31^{c}	22.75 ± 2.03^{c}	24.50 ± 1.60^{c}
25	10.38 ± 2.69^{ab}	13.08 ± 3.29^{ab}	16.75 ± 3.64^{bc}	21.66 ± 5.67^{bc}	23.66 ± 6.62^{c}	26.00 ± 7.05^{b}

Stem girth

This study found that the stem girth of *Abelmoschus esculentus* differed among the different levels of contamination, with the 5% level of contamination having the highest stem girth and the (15% concentration) has the lowest stem girth at 4, 6, 8, 10, 12 and 14 WAP (Weeks after Planting). According to statistics, 5% level of concentration resulted in significantly larger girth than the other levels of concentration at 0.05 level of probability. When compared to the previous weeks after planting, there are no significant differences between 0%, 10%, 15%, 20%, or 25% at P (0.05) level of significance at 4 WAP, but there is a significant difference at 5% level of concentration.

Number of leaves

As the concentration of effluent increased, a substantial variation in the number of leaves in *Abelmoschus esculentus* was observed. However, compared to plants cultivated at lower concentrations, there was a considerable decrease in the number of leaves at the 25% level of concentration. Statistical analysis revealed that, at a 0.05 level of probability, the number of leaves does not alter substantially over the weeks at any level of concentration (Table 3).

Table 2: Stem girth (cm) of Okra treated with detergent effluent

Effluent concentration (%)	4 WAP	6 WAP	8 WAP	10 WAP	12 WAP	14 WAP
Control	3.17 ± 0.19^{ab}	3.39 ± 0.49^{a}	3.93 ± 0.42^{ab}	4.19 ± 0.42^{ab}	4.80 ± 0.30^{ab}	5.09 ± 0.28^{ab}
5	3.77 ± 0.33^{b}	4.29 ± 0.25^{a}	5.10 ± 0.25^{a}	5.64 ± 0.26^{a}	6.12 ± 0.26^{a}	6.47 ± 0.39^{a}
10	2.63 ± 0.37^{ab}	2.80 ± 0.36^{a}	3.46 ± 0.38^{b}	3.87 ± 0.44^{b}	4.55 ± 0.48^{b}	4.83 ± 0.51^{ab}
15	2.02 ± 0.32^{a}	3.08 ± 0.65^{a}	3.60 ± 0.87^{b}	4.09 ± 0.96^{b}	4.61 ± 1.05^{b}	5.23 ± 1.14^{a}
20	2.92 ± 0.53^{ab}	3.32 ± 0.42^{a}	3.85 ± 0.39^{ab}	4.31 ± 0.51^{ab}	4.79 ± 0.46^{ab}	5.18 ± 0.50^{a}
25	3.33 ± 0.33^{ab}	3.51 ± 0.52^{a}	4.03 ± 0.48^{ab}	4.30 ± 0.58^{ab}	4.97 ± 0.38^{ab}	5.18 ± 0.39^{a}

Table 3: Number of leaves of Okra treated with detergent effluent

Effluent concentration (%)	4 WAP	6 WAP	8 WAP	10 WAP	12 WAP	14 WAP
Control	4.20 ± 0.41^{a}	4.58 ± 0.58^{a}	4.43 ± 0.58^{ab}	4.85 ± 0.35^{ab}	5.28 ± 0.34^{ab}	5.83 ± 0.44^{ab}
5	4.50 ± 0.38^{a}	4.91 ± 0.08^{a}	5.70 ± 0.56^{a}	5.76 ± 0.50^{a}	6.16 ± 0.44^{a}	6.66 ± 0.54^{a}
10	4.50 ± 0.50^{a}	4.33 ± 0.46^{a}	4.16 ± 0.46^{b}	4.75 ± 0.25^{ab}	5.00 ± 0.38^{ab}	5.50 ± 0.28^{ab}
15	3.33 ± 0.33^{b}	3.91 ± 0.65^{b}	4.50 ± 0.90^{ab}	5.75 ± 0.80^{a}	6.00 ± 0.66^{a}	6.66 ± 0.65^{a}
20	4.33 ± 0.36^{a}	4.66 ± 0.16^{a}	4.66 ± 0.22^{ab}	4.75 ± 0.38^{ab}	5.50 ± 0.80^{ab}	6.25 ± 0.90^{a}
25	4.41 ± 1.50^{a}	4.48 ± 0.28^{a}	4.91 ± 0.08^{a}	4.75 ± 0.87^{ab}	4.16 ± 0.32^{b}	4.41 ± 1.70b

Weight of the shoot

The lowest mean fresh shoot weight for Okra was reported at a concentration of 15%, and the highest was recorded at a concentration of 5%. Similar to this, the 15% level of concentration showed the lowest mean dry shot weight, while the 5% level of concentration revealed the highest mean dry shot weight (Table 4). As the least weight of shoot for both fresh and dry weight is recorded at the 15% level of effluent concentration, this result demonstrates that the concentration of effluent at this level of concentration is more negatively impactful than other levels of concentration. Dry shoot weight does not significantly change across all levels of concentration, however fresh shoot weight does significantly differ across all levels of concentration

Weight of the root

For the weight of Okra root, the mean fresh root weight was lowest in plants cultivated at 15% and 25% effluent concentration levels and highest in plants grown at 5% effluent concentration levels. According to statistical analysis, plants grown at the 5% concentration level were statistically different from plants grown at other concentration levels. Additionally, at concentration levels of 5% and 10%, the dry weight of roots was at its maximum, but at concentration levels of 0% (control) and 25% effluent concentration, it was at its lowest (Table 5). Fresh and dry root weight was slightly significantly different across all levels of concentration.

Table 4: Fresh and dry weight of shoot of Okra treated with effluent at 14 WAP

Effluent conc. (%)	Fresh shoot weight(g)	Dry shoot weight(g)
Control	11.66 ± 0.88^{bc}	9.50 ± 0.66^{a}
5	16.33 ± 0.88^{d}	10.74 ± 0.31^{a}
10	12.66 ± 1.76^{c}	9.93 ± 0.32^{a}
15	5.33 ± 0.33^{a}	2.72 ± 0.21^{a}
20	8.33 ± 0.33^{ab}	6.12 ± 0.23^{a}
25	11.66 ± 1.20^{bc}	8.83 ± 1.08^{a}

Table 5: Fresh and dry weight of root of Okra treated with effluent at 14 WAP

Effluent conc. (%)	Fresh root weight(g)	Dry root weight(g)
Control	1.66 ± 0.33^{a}	0.66 ± 0.16^{a}
5	4.33 ± 0.33^{b}	2.00 ± 0.00^{a}
10	2.00 ± 0.57^{a}	1.66 ± 0.33^{ab}
15	1.33 ± 0.33^{a}	0.83 ± 0.16^{a}
20	1.66 ± 0.33^{a}	0.66 ± 0.28^{a}
25	1.33 ± 0.33^{a}	1.03 ± 0.11^{a}

Variations in the epidermal characteristics of *Abelmoschus esculentus* leaves.

With stomata on both the abaxial and adaxial surfaces in all treatments (control, 5%, 10%, 15%, 20%, and 25% effluent concentrations), the leaves of *Abelmoschus esculentus* plants were amphistomatic. Paracytic types of stomata were found in all the treatments. This demonstrated that the presence of stomata on both leaf surfaces was unaffected by the effluent. *Abelmoschus esculentus* stomata were affected by the effluent, and reductions in stomata size were seen after the concentration reached 5%. All of the treatments (control, 5%, 10%, 15%, 20%, and 25% effluent concentrations) resulted in irregularly shaped epidermal cells on the abaxial leaf surface (Table 6). Adaxial (upper) leaf surfaces in (5%, 10%, 20%, and 25%) showed polygonal cell form, whereas (control and 15%) showed irregular cell shape.

This demonstrates that at greater effluent concentrations, the effluent had an impact on epidermal cell shape. In every treatment, there are different numbers of epidermal cells on the adaxial and abaxial surfaces. The control had the most cells while declines were seen at 5% effluent concentration levels. This demonstrates that the effluent had an impact on the number of epidermal cells on both leaf surfaces (the fewer the number of epidermal cells present, the greater the effluent concentration). On the surface of the abaxial leaf, the anticlinal wall pattern was wavelike in all treatments, however on the surface of the adaxial leaf, a curved pattern was visible in (5%, 20%, and 25%), while a straight anticlinal pattern was found in (control, 10%, and 15%).

Table 6: Epidermal cell shape and Anticlinal wall pattern of Okra treated with effluent

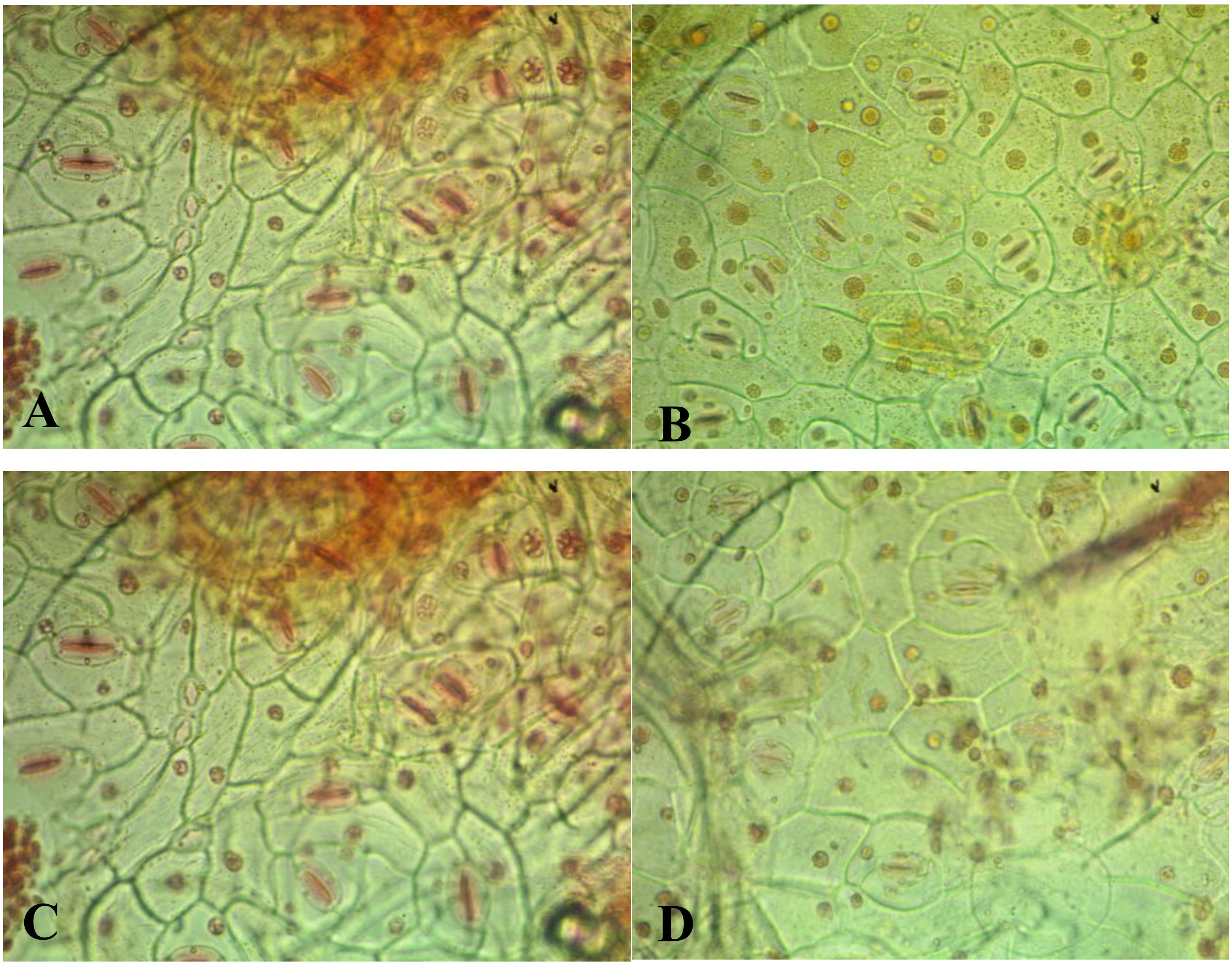
Effluent Concentration (%)	Epidermal cell shape		Anticlinal cell wall pattern	
	Abaxial	Adaxial	Abaxial	Adaxial
Control	Irregular	Irregular	Wavy	Straight/slightly curved
5	Irregular	Polygonal	Wavy	Curved
10	Irregular	Polygonal	Wavy	Straight/slightly curved
15	Irregular	Irregular	Wavy	Straight/slightly curved
20	Irregular	Polygonal	Wavy	Curved
25	Irregular	Polygonal	Wavy	Curved

Variation in the epidermal cell size of *Abelmoschus esculentus* leaves.

The highest epidermal size was found in the 5% level of concentration and the least was found in the control (0%) level of concentration for the epidermal measurement. However, the adaxial showed significant differences across all the level of concentration, with the highest epidermal size found in the 10% level of concentration and the least found in the 15% level of concentration (Table 7).

Table 7: Epidermal measurement of Okra treated with effluent

Effluent concentration (%)	Epidermal measurement (mean±S.E)	
	Adaxial	Abaxial
Control	2.31 ± 0.30^{ab}	2.04 ± 0.16^{a}
5	2.66 ± 0.45^{ab}	2.84 ± 0.41^{a}
10	3.32 ± 0.33^{a}	2.26 ± 0.21^{a}
15	1.99 ± 0.22^{a}	2.83 ± 0.23^{a}
20	2.50 ± 0.31^{ab}	2.13 ± 0.24^{a}
25	3.03 ± 0.40^{ab}	2.22 ± 0.26^{a}



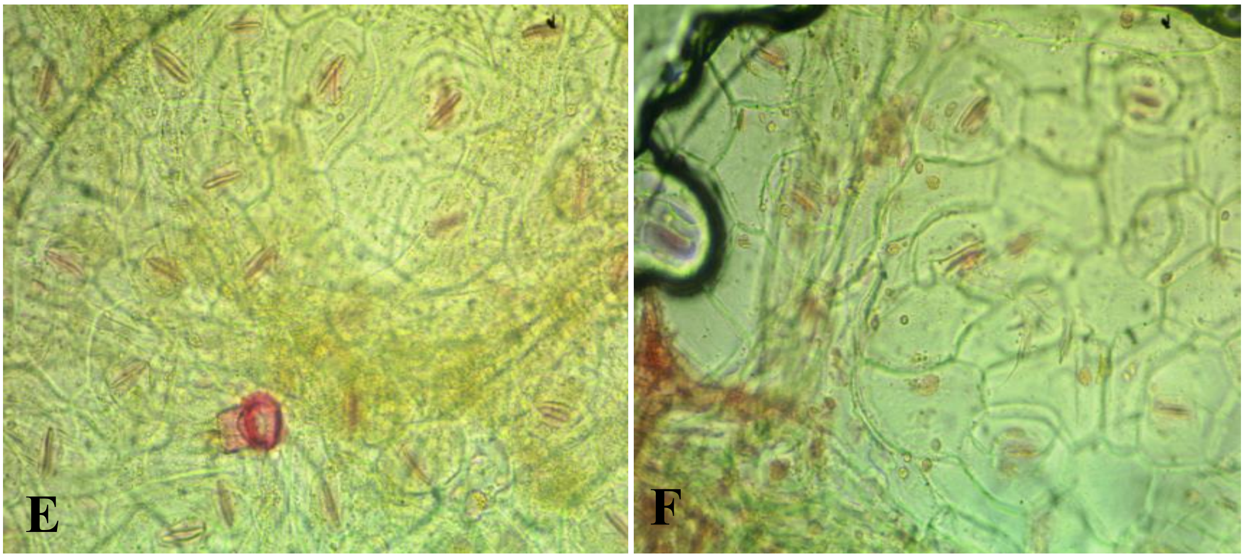
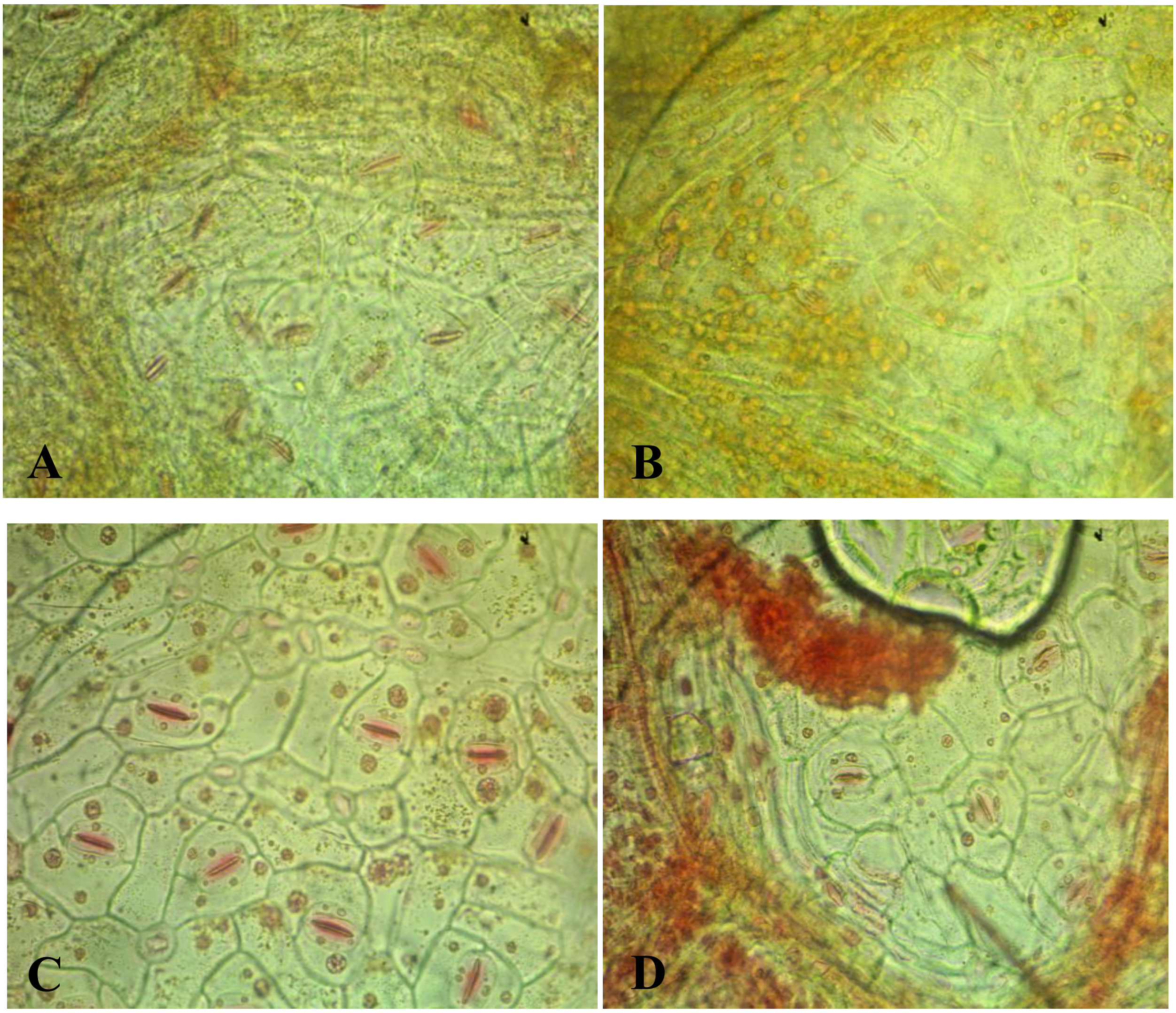


Figure 1: Leaf epidermal features of different concentrations of detergent effluents on *Abelmoschus esculentus* A. Abaxial surface of control treatment B. Adaxial surface of control treatment C. Abaxial surface of 5% treatment D. Adaxial surface of 5% treatment E. Abaxial surface of 10% treatment F. Adaxial surface of 10% treatment.



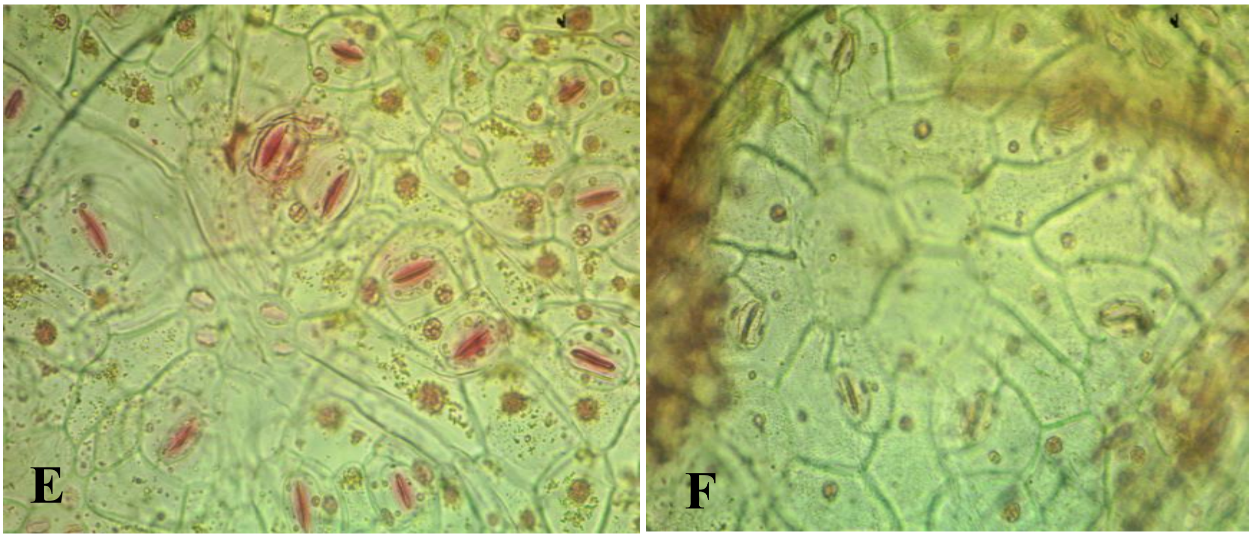


Figure 2: Leaf epidermal features of different concentrations of detergent effluents on *Abelmoschus esculentus* A. Abaxial surface of 15% treatment B. Adaxial surface of 15% treatment C. Abaxial surface of 20% treatment D. Adaxial surface of 20% treatment E. Abaxial surface of 25% treatment F. Adaxial surface of 25% treatment

Discussion

In Nigeria, large amounts of pollutants are always present in the water and sediment because of industrial effluent discharge into receiving water bodies. This study demonstrated that higher concentrations of detergent effluent contamination had a positive impact on *Abelmoschus esculentus* growth parameters, as evidenced by decreased biomass, plant height, stem girth, number of leaves, the weight of fresh and dry shoots, and weight of fresh and dry roots. According to this study on *Abelmoschus esculentus*, growth was more reduced at 15% and 25% effluent concentration. This could be due to ingredient that may be present in the effluent, and this could lead to changes in soil conditions, the result is in agreement with the findings of Mammi *et al.* (2011).

The study clearly shows that low concentrations of the detergent effluent can be non-toxic to okra growth, but those high concentrations can be hazardous, potentially leading to reduced biomass production. Wahid *et al.* (2000) reported that the growth and yield of soybean cultivars were greatly reduced because of wastewater effluents from a chemical industry in Lahore that was highly saline and had extremely high electrical conductivity, which corresponded with this work where detergent effluent affect the growth of Okra at higher concentration

According to the FAO (1992) guideline, toxicity and miscellaneous problems, can be expected from use of wastewater effluents. The plant's leaf epidermis responded differently to the different detergent effluent concentrations. This was similarly noted in *Amarantus hybridus* which was watered with pharmaceutical effluents and crude oil (Ogunkunle *et al.*, 2013). Significant reduction in stomata size was seen from 5% to 25% effluent treatments, as well as an impact on the number of leaves.

Additionally, the toxic effect of the detergent was seen at effluent concentrations of 15% a decrease on the size of epidermal cells on adaxial surface while 25% recorded the highest value of size of epidermal cells, the reduction on the size of epidermal cell size could be as a result of the grossly effect of the effluent and the impactfulness of the effluent at 15% concentration. This agrees with findings of Omosun *et al.* (2008) who reported low stomata index in the plants treated with pharmaceutical effluent and this also go in line with the work of Okanume *et al.* (2017). Paracytic stomata types was found in all the treatments which conforms with the findings of Osawuru *et al.* (2011). Which observed from 5% to 25% effluent concentrations in the form of reduction of trichome density and number of epidermal cells on both surfaces, this perhaps may be a strategy to reduce stress from physiological processes on the adaxial surface since it is the surface receiving much of the solar radiation and engage in photosynthesis more.

According to this study, agricultural crops could still benefit from using modest concentrations of detergent effluents in order to lessen the toxicity of the contaminants. Industrial waste that has not been treated poses major risks to plants and eventually to human health. It can also be deduced that *Abelmoschus esculentus* anatomical structure and plant growth are all inhibited in some way by the detergent effluent's increased concentration. *Abelmoschus esculentus* cultivated in effluent-contaminated soil underwent apparent, significant changes that could be used as a gauge for monitoring environmental pollution.

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