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Environmental Pollution and Leaf Cuticular Variations in *Newbouldia laevis* Seem. ex Bureau

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Abstract: Matured leaves of *Newbouldia laevis* were randomly collected from polluted and non-polluted habitats in two areas of Edo state. Microscopic examination of their epidermis revealed that the stomatal apertures in leaves from polluted habitats were closed while those from non-polluted habitats were opened. Also, epidermal cell aberrations and erosion were noticed in polluted specimens while the non-polluted specimens had normal tissue arrangements. Leaves from both populations were hypostomatic consisting of anisocytic stomatal type. The frequency of stomata for polluted and non-polluted samples was estimated to be 66.6 and 25.0%, respectively. Furthermore, the polluted population had an average leaf area of 54.98 cm^2 while the non-polluted population had 126.36 cm^2. It was suggested that foliar morphology of *Newbouldia laevis* could serve as a Phytometer to gauge the effects of air pollutants on the environment.

Key words: Pollution, hypostomatic, anisocytic stomata, *Newbouldia laevis*

INTRODUCTION

Newbouldia laevis Seem. ex Bureau is a glabrous erect shrub or tree that grows up to 15 m high. It is distinctly decorative, with shiny dark green leaves, commonly found in deciduous, secondary and fringing forests. They also grow in villages as good live fence or boundary tree around sacred groves and shrines (Irvine, 1961; Akobundu and Agyakwa, 1998). This taxon has cultural value in Nigeria especially in dressing the head of a king during coronation ceremony.

According to Lenntech (2005) traffic is held responsible for one-third of the greenhouse gas emissions. Emissions caused by traffic are mainly those of Carbon dioxide, Carbon monoxide, Nitrogen oxides, Volatile Organic Compounds (VOC) and small dust particles. Paul and Gail (1998) had earlier reported that the most dangerous substances are those smaller than 2.5 μ and 80% of these are produced by traffic, especially from diesel-power vehicles.

This present study has been carried out with a view to revealing the epidermal morphology of this species growing along roadsides, a place where a variety of gaseous particulate and photochemical pollutants from fossil fuel combustion bombard plants as they struggle to grow in heavy metal-laden soils. The present study has been carried out with a view to establishing the role, foliar morphology of *Newbouldia laevis* could play as a Phytometer to gauge the effects of air pollutants on the environment.

MATERIALS AND METHODS

Matured leaf samples of *Newbouldia laevis* were randomly collected in November-December 2006, from Oluku and Ebvoneka, Edo State, Nigeria. Oluku is an industrial and densely populated village situated on the Benin-Lagos highway at about 10 km away from Benin City while Ebvoneka is a sparsely populated rural community situated off Benin-Ifon road at about 20 km away from Benin City.

The population from Oluku represented a polluted habitat in which plants were affected by air pollutants from the high vehicular traffic and numerous light industries situated along the highway while the population from Ebvoneka, was devoid of these pollutants and represents a non-polluted habitat. Plant samples were collected from sites typical of the area roadsides.

Ten leaves per population were measured for their sizes and the leaf areas were determined according to Duncan and Hesketh (1968) and Remison and Lucas (1982). The epidermal peels for cuticular studies were obtained following the method outlined by Metcalfe and Chalk (1988). The leaves were placed with the outer surface facing downward on a flat surface and flooded with commercial bleaching agent. The epidermis was gently, carefully and gradually peeled off from the mesophyllic tissues of the leaves with the aid of sharp razor blade. The peels were stained with combination of safranin and delafield's haematoxylin and mounted

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temporarily on slides. Ten slides (each of adaxial and abaxial surfaces) were prepared per population. These slides were examined under the light microscope and data were collected from 10 microscopic fields selected at random from each slide. Line drawings were also made. The length and width of stomata pores and guard cells were measured and data were collected from 25 stomata per leaf surface. This was done in 10 replications and their means and stomatal index were determined. Data were analyzed statistically.

RESULTS AND DISCUSSION

The leaf size, area and surface morphology of *Newbouldia laevis* was shown in Table 1. The polluted population of this species had leaf necrosis and average leaf area of 54.98 cm^2 while that of the non-polluted population had an average leaf area of 126.36 cm^2. No necrosis was observed on the surfaces of the leaves. The natural dark green colour of leaves of the polluted population had faded compared to those of the non-polluted population. Eroded epicuticular wax and perforations were observed in both adaxial and abaxial surfaces in the polluted population of this taxon. These were however not observed in the samples obtained from the non-polluted microhabitats.

In both populations, stomata were only present in the abaxial surface of the epidermis. Hence, *Newbouldia laevis* is hypostomatic. The frequency of stomata (stomatal index) was estimated to be 66.6 and 25.0% for polluted and non -polluted samples, respectively. Shape of epidermal cell ranged from hexagonal to polygonal and the walls of the cells were found to be straight in the adaxial surface, while in the abaxial surface, they were slightly sinuous. On the other hand, the non-polluted leaves showed irregular shapes of epidermal cell in the adaxial and abaxial portions. Abundant peltate scales were observed in the adaxial surface of leaves of both populations as opposed to their abaxial surfaces. Hairs were absent in this species.

Stomata in this taxon were anisocytic. Closed stomatal apertures were observed in the polluted population, whereas in the non-polluted populations, apertures were opened with average mean width of 0.37 μm. The reduced growth in size, development of necrosis and colour-fading appearance of leaves of *Newbouldia laevis* growing in the polluted environment could be due to the blockage of stomatal aperture occasioned by the combined effects of different pollutants they derived from effluents from exhaust of automobiles and the light industries. In the same vein, perforations in the epidermis of leaves of this population could be attributed to the abrasive force of dust particulate against them, since, they lacked hairs, which could have prevented the dusts from reaching the leaf cuticle. Perforations could enhance the entrance of

Table 1: Leaf size and epidermal characteristics of both populations of *Newbouldia laevis* (P.Beav.) Seemann ex bureau (Bignoniaceae)

Description		Population	
		Oluku	Ebvoneka
A. Leaf dimension			
Mean length (cm)		13.73±0.1	19.19±0.1
Mean width (cm)		5.43±0.2	8.78±0.2
Average leaf area (cm^2)		54.98±0.1	126.36±0.1
B. Leaf epidermal characteristics			
Nature of epidermal cell	U	Straight wall and range from Hexagonal to Polygonal shapes	Irregular in shape
	L	Slightly sinuous	Irregular in shape
Type of stomata	U	Absent	Absent
	L	Anisocytic	Anisocytic
Stomata index	U	Nil	Nil
	L	66.6%	25%
Type of epidermal hair	U	Absent	Absent
	L	Peltate scale	Peltate scale
Mean width of stomata pores (μm)	U	Absence of stomata	Absence of stomata
	L	-	0.37±0.1
Mean length of stomata pore (μm)	U	Absence of stomata	Absence of stomata
	L	-	0.62±0.1
Mean length of guard cell (μm)	U	Absence of stomata	Absence of stomata
	L	1.56±0.1	1.53±0.1
Mean width of guard cell (μm)	U	Absence of stomata	Absence of stomata
	L	1.81±0.1	1.65±0.1
Status of stomata pores	U	Absence of stomata	Absence of stomata
	L	Closed pores	Opened pores
Integrity of epicuticular wax	U	Subsequently eroded	Present
	L	Subsequently eroded	Present
Leaf surface appearance	U	Presence of necrosis	Absence of necrosis
	L	Presence of necrosis	Absence of necrosis

pathogenic organisms into the mesophyllic tissues to cause damages. This could also reduce the photosynthetic capacity of these leaves. This is similar to the earlier report of Adela (1990) that air pollutants injure trees by damaging their foliage and impairing the process of photosynthesis (food making), they also weaken trees making them more susceptible to other health problems such as insects and diseases. The loss of green colouration in leaves of this population might hinder food-manufacturing ability of the leaves whose stomatal apertures were also closed.

The large, healthy green leaves and opened stomatal apertures of the non polluted populations of this taxon could be a positive response to equable conditions they derived from steady and less disturbed microhabitat. It is believed that large leaves of this population will be able to capture enough amount of light for photosynthesis. This no doubt can serve as booster to other metabolic processes that enhance growth in the plant.

The variations in structure and shape of epidermal cells in adaxial leaf surface of the polluted population could be the effects of pollution on the leaves of this species, since, the epidermal cell-shapes in leaves of the non-polluted population maintained consistent irregular shape throughout the adaxial and abaxial surfaces of the leaves. The sinuous epidermal cell wall of the abaxial surface of polluted population could be as a result of stress posed on them by gaseous pollutants, since this was different from what was observed in the abaxial epidermis of non-polluted population. These tend to confirm the previous assertions of Gill *et al.* (1980), Pal *et al.* (2002) and Luzimar *et al.* (2005) on the alterations in the anatomical features of leaf surfaces by gaseous pollutants from automobiles. Pal *et al.* (2002) collected *Asparagus racemosus* Willd., *Azadirachta indica* A. Juss, *Bongainvillea spectabilis* Willd., *Cassia fistula* L., *Ficus reflexa* Thub, *Nerium odorium* Sol., *Polyalthia oliveri* Engl and *Thevetia neriifolia* Juss. Ex Steud. growing along roadsides of low and high traffic density and revealed collapsed epidermal cells, irregularly fused cell boundaries, eroded epicuticular wax and patches of a crust on the cuticle and reported that these could be attributed to automobile emissions which the plants were continuously exposed. Luzimar *et al.* (2005) investigated the anatomical and micromorphological alterations in leaves of *Eugenia uniflora* L. and *Clutia robusta* Pax which were exposed to stimulated acid rain. They reported erosion and morphological modification of epicuticular wax in both upper and lower epidermis of the leaves, necrosis in leaf margin, rupture of stomata edges and occasional leaf perforations.

The high stomatal frequency in the abaxial surface of leaves of the polluted population could be a physiological development to survive the unfriendly environmental conditions created by the varied degrees of pollutants that predominates busy roadsides. Thus confirming the previous assertion of Pal *et al.* (2002) on stomatal frequency.

In conclusion, the results from this study may be useful in the ecological and profitable management of this important cultural species in Nigeria.

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