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Structural Changes in Stomata in Plants Exposed to Air Pollution

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Abstract: *Air pollution, is now almost inescapable component of urban life effecting both plants and animals equally.The changes in the epidermal configuration reveal marked alteration in numberof stomata, epidermal cells and stomatal indices in leaf samples collectedfrom polluted zones that can be used as biomarkers of air pollution. These pollutants not only affect the morphology of plants but also alter the physiology. Reduction in various parameters of the two plant species studied frompolluted sites clearly indicate the deleterious effect of air pollution on plant health.It is evident from the present study that the air pollutants such as SPM, SO_x, NO_x and O_3 from automobile exhaust and industries along with many other unknown pollutants are responsible for bad air quality and are responsible for altering the epidermal structures in both Tridax and Datura plants growing in polluted zones.*

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I. Introduction

Continuously increasing vehicular pollution in the major cities of the world has led to serious environmental problems. Air pollution is now almost inescapable component of urban life effecting both plants and animals equally. The major pollutants are nitrogen oxides, carbon monoxide, carbon dioxide, sulphur oxides, volatile organic compounds and suspended particulate matter which contribute to 90% of air pollution. Apart from these small amounts of metals such as Cd, Cu, Hg and Zn have been discovered (NassimaOucher, 2015;Aksu ,2015).

60–70% of the pollution found in an urban environment is on account Motor vehicles (Singh, *et al*., 1995; Tripathi and M. Gautam 2007; Dwivedi and B. D. Tripathi 2008). Maximum pollution load in the major metros of India, is contributed by two wheelers alone (Central Pollution Control Board,2002).

Plants act as good indicators of air pollution. They play significant role in assimilation and accumulation of pollutants. Under polluted conditions, plants develop different morphological, physiological and anatomical changes (Inamdar and Chaudhri, 1984; Iqbal, 1985; Gupta and Ghouse, 1988;Gravano*et al*., 2003; Novak *et al*., 2003). Plants regulate the entry of gaseous molecules through stomatal movements. Injury to the plants by pollutants depends on the amount of pollutants entering the cells and their interaction with cellular constituents. Responses of plants to gaseous pollutants vary from species to species. Chronic exposure of leaves to pollution can break down the cuticle leading to excessive loss of water and also interferes with photosynthesis and growth. Adverse effects of air pollution on plants have been studied by various workers (Kaur, 2004;Dineva,2006; Rai and Kulshreshtha, 2006; Sher and Hussain 2006).

In the present investigation the influence of vehicular pollution on *Tridaxprocumbens* and *Daturametel* species was studied to understand their adaptive response to it. These plants were selected as they were found growing in the polluted sites.

Tridaxprocumbens, a member of Asteraceae is best known as a widespread weed in fields, meadows, croplands, disturbed areas, lawns, and roadsides in areas with tropical or semi-tropical climates.*Datura metel*a member of Solanaceaeis a perennial herb. Both the plants have medicinal importance.

II. Materials and methods

Mature leaves of the selected plant species were sampled in the early hours. Leaves were collected from the polluted and control site and were kept in ice box and brought to the lab. The epidermal peel of both adaxial (upper) and abaxial (lower) surface was carefully taken using the nail polish method to study the various epidermal related parameters using Optika Trinocular research microscope. Parameters like number of stomata, number of epidermal cells, size of the stomata, size of the guard cellsand subsidiary cells, stomatal type and stomatal indexwere studied. All the observations were made in triplicates.

Preparing the epidermal peel:

Leaves from the selected plants were collected, thoroughly washed and dried. A thick patch (at least one square centimeter) of clear nail polish was applied on both the leaf surfaces to be studied. It was then allowed to dry completely. A piece of clear cellophane tape was stuck to the dried nail polish patch and gently peeled from the leaf by pulling on a corner of the tape. This is the leaf epidermal impression. The nail polish peel was fixed on to a very clean microscopic slide. Using a scissors, trim away any excess tape. Label the slide and examine the leaf impression under 40X magnification. Search for areas where there are numerous stomata, and where there is no dirt, thumb prints, damaged areas, or large leaf veins. Count all the stomata in one microscopic field. Take at least three readings from other distinct microscopic fields and determine an average number per microscopic field. Other parameters like stomatal and epidermal cell size, number of subsidiary cells were also observed and measured

III. Resultsand Discussion

Tridaxprocumbens: Number of stomata on the upper epidermis of *Tridax* leaf collected from the polluted zone was 33%more than the control plantwhereas, on the lower epidermis the number of stomata decrease and the decrease was42.5% as compared to the control plants. The length showed a decrease by 43.8% on the upper epidermis whereas the decrease in length on the lower epidermis was only 7.5%. The breadth of the stomata was observed to be 8.5% more on the upper epidermis and a decrease of 35% was noted on the lower epidermis in the leaves collected from polluted zones as compared to the control plants.The size of the guard cells was 2% higher on the upper epidermis and 7% reductions was observed on the lower epidermis. Whereas there was 38% increase in the breadth of the guard cells on the upper epidermis and only 19% reduction was noted on the lower epidermis in the leaves collected from polluted zones as compared to the control. The stoma size showed a slight increase on the upper epidermis but on the lower surface the stoma showed a decrease in size. The stomatal index on the lower epidermis of the leaves collected from polluted site was more compared to the control plants. The number of subsidiary cells was observed to be four and the type of stomata in *Tridax* is anomocytic in all leaves collected from polluted zones and control plants. The subsidiary cells visually appeared larger on both the epidermal surfaces in leaves growing in polluted zones.

In *Datura* Number of stomata on the upper epidermis was less by 25% and more by 83%on the lower epidermis in the leaves of plants growing in polluted zones as compared to the control plants. The length of the stomata was observed to be 27% less on the upper epidermis and an increase of 9.8 % was observed on the lower epidermis in plants growing in polluted zones as compared to control plants. The breadth of the stomata decreased by 15.7% on the upper epidermis and a slight increase of 4.8% was observed on the lower epidermis in the leaves of plants growing in polluted zones as compared to control plants. The length of guard cells on the upper epidermis showed a decrease by 34% on the upper epidermis whereas the decrease was only 21% on the lower epidermis and the width of guard cells showed a decrease by 77% on both the epidermis in the leaves of plants collected from polluted zones as compared to control plants. The size of the subsidiary cells was smaller on both the surfaces on the leaves collected from polluted zones as compared to control. The number of subsidiary cells were observed to be three and the type of stomata is anisocytic in all leaves collected from polluted zones and control plants. Higher stomatal index was reported on both the surfaces of in the leaves of plants collected from the polluted site.

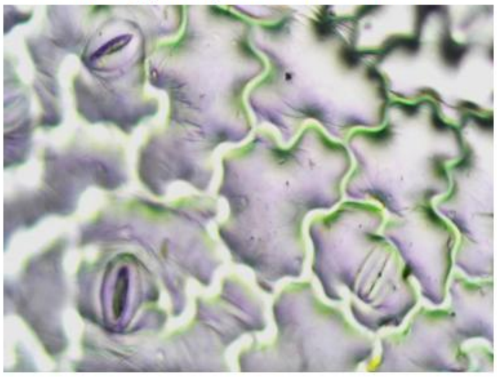
The number of epidermal cells were observed to be less on both the surfaces of *Tridax* leaves collected from polluted zones as compared to control.

Wide stomatal apertures were observed in the leaves of both *Tridax* and *Datura*collected from polluted areas as compared to control plants.

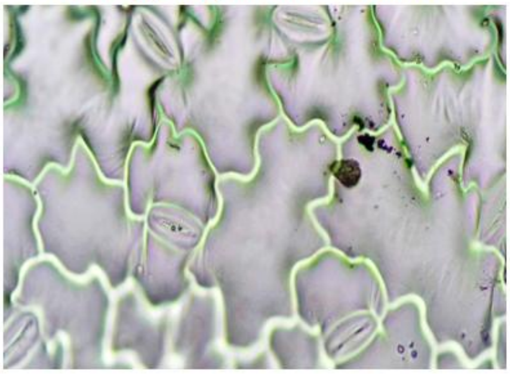
S. No.	Plants		*Tridax*				*Datura*			
	Parameters		Control		Polluted		Control		Polluted	
			UE	LE	U. E	LE	UE	LE	UE	LE
1	No. of Stomata		4	4.7	5.3	2.7	8	6	6	11
2	size of Stomata	L(µm)	7.3	8	4.1	7.4	6.2	5.1	4.5	5.6
		B(µm)	3.5	4.5	3.8	2.9	3.8	4.1	3.2	4.3
4	size of Guard Cell	L(µm)	4.9	5.7	5	5.3	3.1	4.7	2.7	3.3
		B(µm)	0.65	0.73	0.9	0.59	0.88	0.84	0.58	0.66
5	No. of Subsidiary cells		4	4	4	4	3	3	3	3
6	No. of Epidermal cells		9	10	8	6	10	8	6	6

UE – Upper epidermis, LE – Lower epidermis

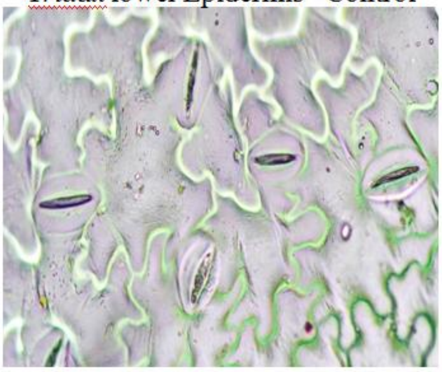
Tridax Upper Epidermis - Control



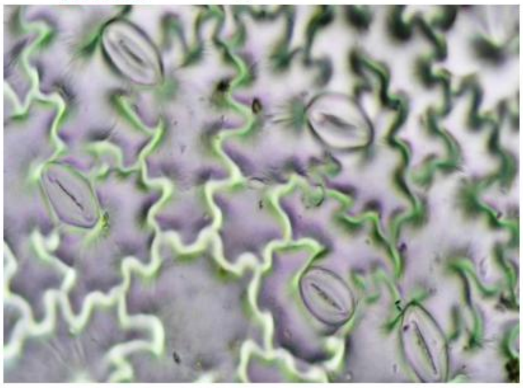
Tridax Upper Epidermis - Polluted



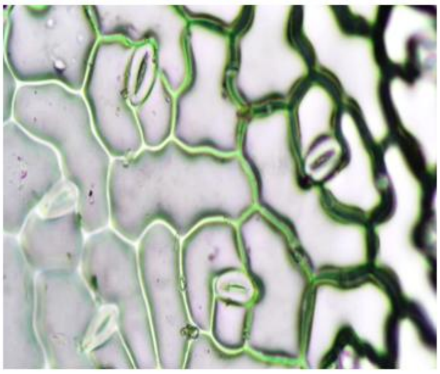
Tridax lower Epidermis - Control



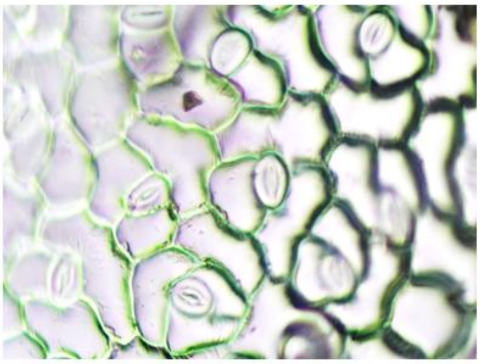
Tridax lower Epidermis - Polluted



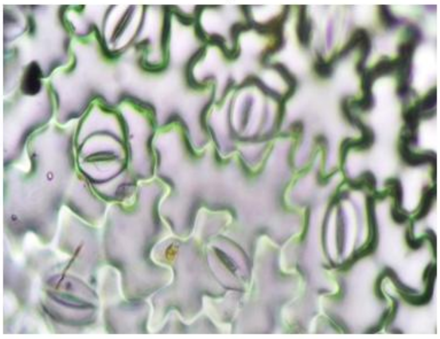
*Datura*Upper Epidermis - Control



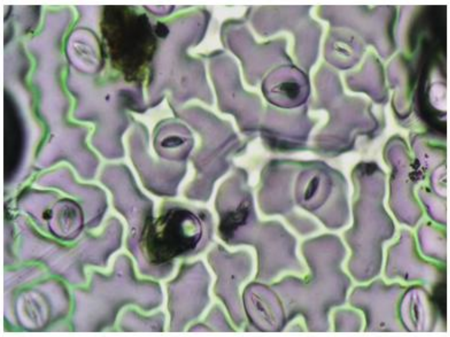
*Datura*Upper Epidermis - Polluted



Datura Lower Epidermis- Control



*Datura*Lower Epidermis - Polluted



IV. Discussion

The use of plants as indicators of air pollution has long been accepted. Many plantsrespond quickly to low concentrations of air pollutants in predictable ways. As a result,plants are considered to be more sensitive to air pollutants than are animals and humans asthey are constantly exposed to air pollutants.

Increase in number of stomata were observed on the upper epidermis of *Tridax* and lower epidermisof*Datura* plants and decrease in number was observed on the lower epidermis of *Tridax* and the upper epidermis of *Datura* was noted in plants growing in polluted zones as compared to control plants.Rai and Kulshrestha2006 suggested that air pollutants inhibit cell elongation, leaf area and consequently the increase in cell frequency resulted in reduction in the size of stomata and epidermal cells.

Decrease in number of epidermal cells both on the upper and lower epidermis of *Tridax*and on the upper epidermis of *Datura* was observed as compared to control in plants growing in the polluted zones. Reduction in the size of stomata and epidermal cells in the plant growing in polluted areas were reported by (Trivedi and Singh 1990) Kulshreshtha*et al*., 1980,1994, Sharma and Roy 1995, Aggarwal 2000, Kaur 2004, Dinva2006, Raj and Kulshreshtha 2006). Significant reduction in the size of epidermal cells and stomata was due to inhibited cell elongation, leaf area and increase in cell frequency as suggested by (Raj and Kulshreshtha2006. Satyanarayana*et al*., 1990;salgare and thorat,1990). Considered reduction in size as an adaptive response of plant to inhibit the pollutant from returning into cells or plants.

Decrease in the size of Guard cells was observed on the upper epidermis of both *Tridax* and *Datura* leaves of the polluted zones. Wide open stoma was observed in both plants growing in polluted zones. Stomatal opening response to so_2 was reported by Rennenberg and Hersch bach, (1996); Robinson *et.al*., (1998) and was related to increased rate of transpiration increases transpiration due to wide stomatal opening. Much convoluted subsidiary cells were observed in the *Tridax* leaves growing in polluted sites.similar results were observed in *pongamiapinnata* exposed to exhaust polluting which could be due to lowering of pH in cytoplasm of guard cells, change in turgor of stomatal complex (Kondo*et al*., 1980).

Different plant species respond differently when exposed to same concentrations of SO_2(Biggs and Davis 1980). It can cause opening of stomata in one species and closing in another (Mudd, 1975). Rai and Mishra, (2013) illustrated that the plants growing along the roadsides have modified leaf surface characters including stomata and epidermal cells due to the stress of automobile exhaust emission with high traffic density in urban areas. A surprising outcome of some recent studies is the suggestion that CO_2 pollution may disrupt the control of water relations in some species because their stomata do not close sufficiently in CO_2-enriched air(Mansfield,1998).changes in epidermal trait, leaf surface structures and leaf morphology in plant species were also observed by many researchers (Shafiq *et al*., 2011; Shafiq *et al*., 2012; Shafiq and Iqbal, 2012; Power, 2013; Kabir *et al*., 2014; Iqbal *et al*., 2015; Pawar, 2016).

V. Conclusion

It is evident from the present study that the air pollutants such as SPM, SO_x, NO_x and O_3 from automobile exhaust and industries along with many other unknown pollutants are responsible for bad air quality. These pollutants not only affect the morphology of plants but also alter the physiology. Reduction in various parameters of two shrub species studied at different sites clearly indicates the deleterious effect of air pollution on plant health and their adaptive response to vehicular pollution.It is evident from the present study that the air pollutants such as SPM, SO_x, NO_x and O_3 from automobile exhaust and industries along with many other unknown pollutants are responsible for altering the epidermal structures in both *Tridax* and *Datura* plants growing in polluted areas.

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