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Effects of Re-Suspended Roadside Dusts on the Phylloplane Microflora

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Abstract: In this study, the changes of phylloplane microbial community due to depositions of dust on the leaf surface of Ficus benghalensis was observed. Accumulation of dust particles due to traffic vehicles lead to lowering of micro fungi count on phylloplane unlike the non-roadside site. Metal analysis of traffic induced dust shown the presence of heavy metals like Pb,Zn,Cu etc. Reduction of microbial colony on leaf surface may be caused due to high exposure on metals and other pollutants present on the dust.
Keywords: phylloplane, SEM, microbial count, air pollution, road side dust.

I. INTRODUCTION

Presence of high amount of nutrients and other essential elements make phylloplane a good habitat for different micro-flora. Bacteria and different filamentous fungi are found to colonize on the phylloplane surface and these inhabitants are known as epiphytes. Research evaluated that the total size of microbial colonization has been estimated about 6.4 × 10^8 km in case of terrestrial leaf surface area [1]. Anthropogenic activities like road construction, sand milling, stone grinding, etc. also generate enough dust which settles down on the phylloplane of the roadside trees. The growing number of fossil fuel driven vehicles produces excessive smoke containing tar particles and other metal pollutants due to the incomplete burning of fuel. It is a well-known fact that a huge amount of traffic generated dust settled down on the upper and lower phylloplane of roadside plant and absorb by plant leaves [2]. Ficus benghalensis and Polyathia longifolia can capture dust ranging between 0.12 mg/cm^2 to 1.89 mg/cm^2 in urban areas of Kolkata mainly in Salt Lake region on both adaxial and abaxial surface.

These pant-microbes associations can be destroyed by the air pollutants. Studies have been done on the air pollutants effects on leaf surface microorganisms in the roadside urban environment [3-6] and from the vehicular pollution effects also [5, 7]. Heavy metal pollution derived from industry and chemical sprays also left a harmful effect on leaf surface microbes population according to researchers [8-10]. The leaf phylloplane is faced to a very hostile situation due to changes in temperature, UV radiation and relative humidity. Along with these physiochemical constraints, different anthropogenic activity can be a reason of difficulty for this host-microbe relationship. This stress hampers the productivity and growth of the phylloplane Microflora population, which can disturb the plant defensive mechanism against the pathogenic attacks [14]. The re-suspended dust on leaf surfaces of roadside plants make contact with phylloplane microorganisms and directly affect their colonizing activity [5]. Different type of non-pathogenic activity i.e. nitrogen fixation, degradation of waxes, production of growth hormones and also biological control of pathogenic bacteria [11-13] hampered by the air pollutants. The present study has been carried to understand the effect of re-susespended dust on phylloplane microflora of Ficus bengalensis.

II. MATERIAL AND METHODS

Kolkata, being one of the quickest developing cities in India has been experiencing air contamination for a long time. More or less 15 million people is staying here and the amount is in increasing every day. Increasing amount of high rise, roads, bridges as well as growing transportation system causes rapid urbanisation on this city, which are the major contributor of particulate emission in air.

For comparative analysis two sites were selected, i.e. Dunlop (site 1/S1) (22°38'N and 88°61'E), and Rajarhat (site 2/S2) (22°35'N and 88°28'E). Heavy load of dust generated by traffic and other activities were seen during the pick time (5000-6000 vehicles/hour in Dunlop. But another site, Rajarhat having low depositions due less amount of vehicle moving.Selected plants are more than 500 meters away from the actual roadside, mainly covered by huge buildings (non-roadside environment). This site was considered as less polluted and non-roadside location. Ficus benghalensis was chosen for the for it's high occurrence on roadside. For metal sample analysis, firstly dried and powdered leaf sample was taken 0.5 gram. Then in a digestion tube it was kept for overnight and digested with 20 ml of concentrated Nitric acid. All the samples were measured at triplicate. Then the filtered extracts were made up to 50 ml with distilled water. Then the sample concentration (ppm) was measured on Perkin Elmer (A400) atomic absorption spectrophotomer. SEM images was taken as described by Ram *et al*. [15].

From an undisturbed leaf, a very small portion ware cut. In the next step it was air dried in a close chamber. Small strips were cut from marginal area. This leaf strips ware mounted on aluminium stab and coated by thin layered highly conductive gold (about 200°A), in an ion sputter coater (GIB2). Then the image of the coated sample was taken in scanning electron microscopy (Hitachi, S530). The air quality of the study sites was collected from the available data in the website of West Bengal Pollution control board (WBPCB) as they have monitoring station in both sites mantioned here

III. RESULT AND DISCUSSION

From the result, it was seen that the levels of particulate matter present on the air was quite higher from the national standards (Table 1) and for NO_2 also, air quality of Dunlop showed a high value than the national air-quality standard. And Table 2 shows the amount of dust depositions on the leaf samples of both sites.

Table 1: Pollution levels in study sites and standard parameters of air pollutants.

Sites	SO_2	NO_2	SPM	RSPM
Dunlop	7.84	63.86	181.04	100
Rajarhat	4.975	42.775	151.25	88
Standard*	60	60	140	60

All values in µg per m^3
*Air quality standard values of West Bengal pollution control board.

Table 2: Amount of dusts deposited on the Phylloplane of *F. benghalensis* at two sites

Sites	Dust adsorbed mg/cm^2
Dunlop	0.382 ±0.235
Rajarhat	0.078 ±0.044

Both the surface of phylloplane generally accumulates dust. Presence of fungal hyphae and conidiophore emerging from the phylloplane was observed on the lower surface of the leaf sample phylloplane, collected from Rajarhat (Figure 1). But no such structure was seen on the leaf sample collected from Dunlop (roadside environment). The upper surface of all this phylloplane was covered by dust, including the stomatal pore. It might be a cause of less microbial association on the leaf surface collected from Dunlop, as the main pathway of nutrient and water to the pathogens are the stomatal pore of the phylloplane. That means Phyllosphere microbes showed a lowering of microbial colony number with increase of air pollutant.

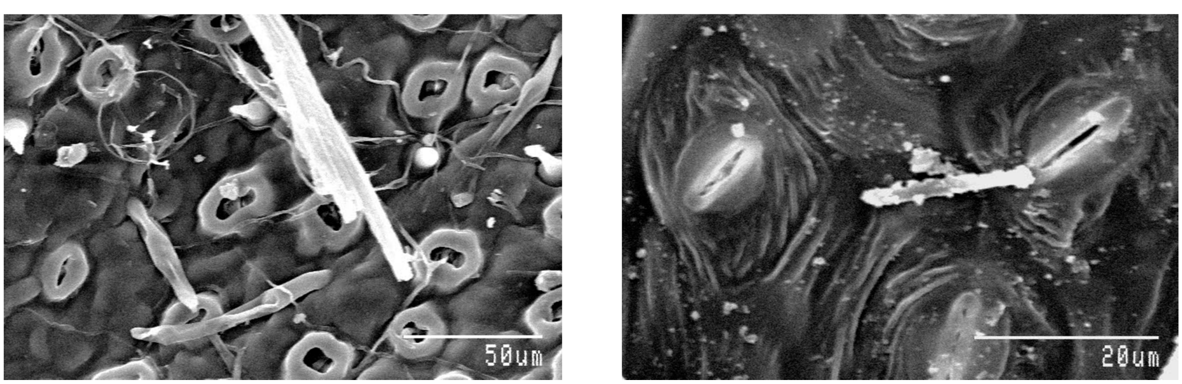
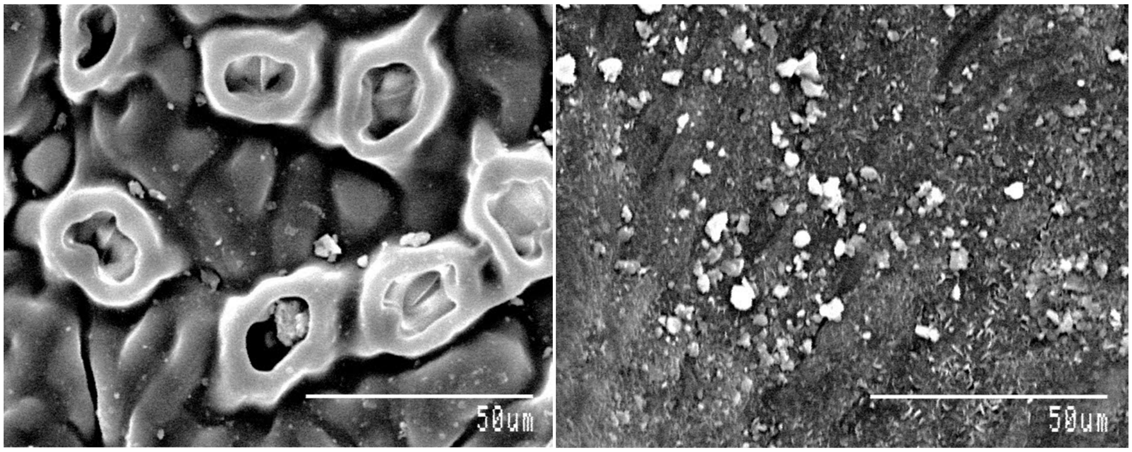


Figure 1: SEM microgragh showing the conidiophore emerges out near the stoma and hyphal growth of fungi near the stoma of F.benghalensis. of S2



(A) Stomata blocked by dust particles B) upper surface of leaf covered by dust particles

Figure 2: Scanning electron micrograph of stomatal dust deposition on the phylloplane of roadside plant of S2 site.

Variation of level of heavy metal depositions was seen on both of the sites. Sites having high particulate matter depositions was showing high amount of metal accumulations. Fe was the most heavily deposited metals present on phylloplane of both the sites (Figure 3). And it was followed by Zn, Cu and other metals. One of the major findings of the studies are presence of fungal hyphae and conidiophore on the phylloplane of the plant of Non-roadside environment but it was absent in Dunlop area. The lowering of the microfungal community occurred due to its collection from busy road crossings. Another finding is dust depositions of phylloplane caused blocking of stomata and hampered the plant-microbial commensalism. Populations of microbes generally very much sensitive towards both gaseous and solid particulates. Metal analysis of the deposited dust shows that damaging effect of the particulates hamper the relationship between plant and microbes. The result of this work has much similarity with work of Joshi [16]. Loss of microbial community due to applications of chemical spray on phylloplane was reported by De Jager et al. [17]. This type of result was also true for lichen also as stated by Monge-Najera et al. [18]. Sandhu et al [19] was reported about the harmful effect of the air pollutants over the other group of bacteria and also about some air borne phenol tolerant microbes too. Bacterial colony population on some dusty Hemlock trees was also fluctuates with high pollutants levels [20]. According to Mohamed and Abo-amer [21] that higher heavy metal concentration and bacterial and fungal populations is inversely proportionate with each other. But it was seen that fungal populations can show high growth rate on some roadside environment also because of its acidic concentration. It is one of the favourable conditions for the fungal growth. Phyllosphere of Tillandsia leaf of an urban environment changes in terms of species variation and colony count due to high pollutant load, according to Brighigna et al [22]. Study revealed that microflora distribution is generally common on the lower surface of the phylloplane. This result is also supported by the findings of Mafia et al [23], according to his research the lower surface of the phylloplane of Eucalyptus spp is infected by pathogen Quambalaria eucalypti. Same result was shown in the work of Leben [24] and Surico [25]

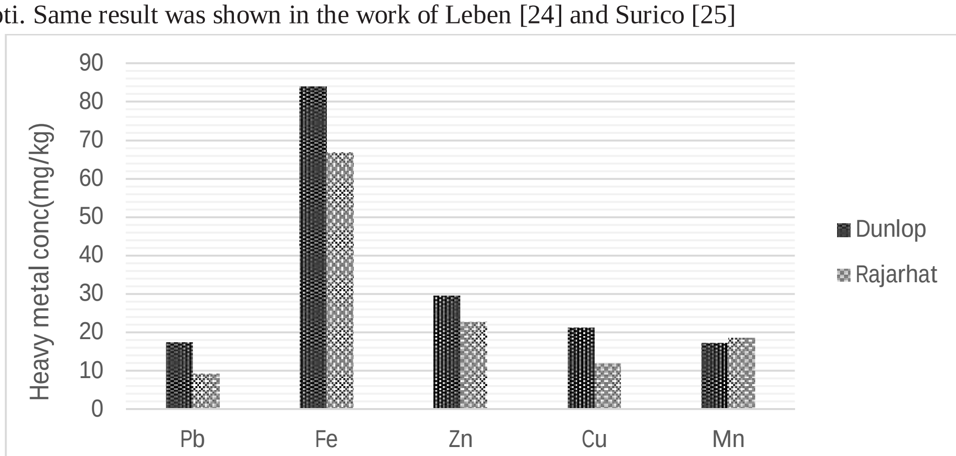


Figure 3**:** Variation of the concentration (mg/kg) of Pb,Cu,Zn,Fe and Mn phylloplane of roadside(S_1) and non-roadside(S_2).

IV.CONCLUSION

Direct microscopy of leaf surface revealed absence of fungal hyphae on the phylloplane of the plant which was collected from the roadside environment. Blocking of stomatal passage by heavy amount of dust was also observed. It can be a major limiting factor of collecting nutrient and water by the microbial community. As the stomatal passage may act as a key passage of transport. It indicates microbial sensitivity towards dust as well as heavy metals. Metals and other elements present in air act as an nutrients to some extent but after that it generally convert into an toxic substances. A detailed study is needed to expose the complex relationship between air pollutants and microbes, and to describe the mechanism of interaction.

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