Air Quality Analytics and Monitoring: A study employing Transplanted Porina Lichen in Mumbai City

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Abstract

Present work aims to investigate bioaccumulation of heavy metals of anthropogenic origin employing lichens as active bioindicators in different locations of Mumbai. Lichens act as reliable bioindicators of terrestrial air quality due to lack of significant cuticle or epidermis combined with their extraordinary capability to grow in a large geographical area. Porina Internigrans (Nyl.) Mull. Arg. was collected from different locations of Bhimashankar for 3 consecutive years, identified at National Botanical Research Institute and transplanted at 30 locations of Mumbai for three months. Dried and powdered lichen samples, standard and control were digested and heavy metals were analyzed. Metals such as Al, Ba, Cd, Cr, Fe, Hg, Mn, Ni, Pb, Cu and Zn were analyzed and the correlation between exposure time and concentrations were statistically tested and enrichment factor (EF) and exposed-to-control (EC) ratio were calculated showing a spatial distribution in the concentration of heavy metals. Differences in concentrations of elements were accounted for by EC ratio, which allowed interpretation of changes in element concentrations.

In this study, some high levels of metals were observed which are attributed to discrete pollution events that occurred a short time before sampling and confirming the ability of lichen transplants for active monitoring.

Keywords: Anthropogenic, Enrichment factor, Bioaccumulation, Exposed-to-Control ratio.

Introduction

The present work has aimed to investigate bioaccumulation of heavy metals of anthropogenic origin using lichens as bioindicators in different locations of Mumbai city and secondly to confirm the ability of the transplanted lichens as potential bioindicators for biomonitoring of heavy metals of anthropogenic origin. Mumbai, most densely populated city of India, is overburdened with thousands of industry and millions of vehicles running every day on the roads across the length and breadth of the city. Despite these facts, to date, no serious attempt has been made to measure the atmospheric heavy metal deposition employing bioindicator.

Many definitions of heavy metals have been proposed based on their density, atomic number, atomic mass, toxicity etc. But the most common one is metals having density more than 5g cm^{-3}. Study of heavy metals is very important as many are toxic due to their stability and non-biodegradability. Hence, they make entry into the food chain. Anthropogenic activities can alter the balance of heavy metals in the environment and hence can alter biochemical and geochemical cycles. Major sources of heavy metals in urban areas are anthropogenic e.g. industrial effluents, solid waste disposal, metal reprocessing etc.

Lichens as bioindicators: The word lichen has a Greek origin which denotes the superficial growth on the bark of trees, rock as well as soil. Lichen species are collectively called as “Stone flower” in English, “Patthar ka Phool” in Hindi, “Dagad Phool” in Marathi etc. Lichens are composite organisms comprised of a mycobiont (fungus) and one or more photobiont (algae) living together in symbiotic association in which the algal partner produces essential nutrients and fungal partner provides mechanical strength. Development and establishment of lichen on a substratum are achieved by fruiting bodies (apothecia) produced by the fungal partner, which must germinate and find an algal partner before they can form a new thallus or may produce minute fragments (isidia or soredia) containing both partners, which can disperse quickly and colonize available habitat.

Biomonitoring employing Lichens: Biomonitoring can be defined as “A continuous observation of a geographical region with the help of a suitable bio-species that reflect changes over space and time”. The use of plant biomonitoring has proved to be a complementary method of investigation for pollutant analysis, as they constitute real biological integrators capable of providing a basis for assessment of environmental quality and/or contamination.[1-4] Over the past few years, research has been focused on the measurement of chemical compounds of plants as indicators of a particular environment state which provides a basis for determining the long-term impact of even low levels of pollution. Since these physiological changes appear before morphological and anatomical symptoms, they provide an early warning signal of modifications in environmental quality.

Lichens are perennial plants with a very slow growth rate which is mainly attributed to the growth of the mycobiont. There is no supply of nutrients from the central part of the growing part and the food produced by the photobiont at the growth site is used by the mycobiont. The pattern of growth, in general, is centrifugal, apical and marginal. In crustose and foliose lichen radical growth occurs while in fruticose lichens there is an increase in length.[5]

For identification of species, a key to genera and species provides concise information to identify the lichen species based on their morphological and anatomical character and chemicals present. A checklist of Indian lichens is available which describes approximately 2300 species belonging to 305 genera and 74 families in different regions of the country.[6-8] The sample collection site Bhimashankar situated in the Western Ghats has also been explored exhaustively for their lichen wealth.[9]

Being pioneers on the rock surface, lichens are important components of the ecosystem that establish life on rock and lichen deprived sites. As lichens colonize on rocks and barks or soil, they trap dust, silt, water and many other things including inorganic and organic materials such as PAHs and heavy metals, which lead to biogeophysical and biogeochemical changes on the work surface leading to soil formation. This also causes changes in their biochemical and physiological characteristics. Lichen occurs in all available substrata and all possible climatic conditions, but the lichen diversity of an area of interest or substratum is highly dependent on prevalent microclimatic conditions.

Apart from morphology and anatomy of lichens, the high success of lichen in extreme climate has been attributed to the secondary metabolites produced by the fungal partner to protect the algal partner.[10] Lichens have a poikilohydric nature to survive in various climatic conditions as they have no mechanism to prevent desiccation; they desiccate and remain dormant when their environment dries out but can rehydrate when water becomes available again.[11]

Lichens usually absorb water directly through their body surface by aerosol, mist and water vapours; due to which they live long in dry areas even on stones and rocks used for the construction of monuments and other building artifacts. The ability to accumulate nutrients from rain or runoff enables them to colonize on the varied substratum.[12] The common natural substrata on which lichens can colonize and grow successfully include all categories of rocks (saxicolous), trees (corticolous), soil (terricolous), wood (lignicolous) and leaves (foliicolous) while man-made substrata include rubber, plastic, glass, stone-work, concrete, plaster, ceramic, tiles and bricks.

Lichens and mosses are reliable indicators of terrestrial air quality which due to lack of significant cuticle or epidermis make them well suited. Bioindicators and biomonitors absorb both nutrients directly from the atmosphere along with pollutants without having any visible signs of injury to the lichen thallus. These features combined with their extraordinary capability to grow in a large geographical area ranked them among an ideal and reliable bioindicator of air pollution. Being located above the ground, epiphytic lichens usually receive greater exposure to air pollution and do not have access to soil nutrient pools and as lichens usually grow on dead bark, therefore chances of contamination by nutrient cycling are also minimal.[13]

Higher plants (tracheophytes) are also employed in biomonitoring studies, but as they also accumulate pollutants from soil, interpretation of data requires more understanding of contributing factors.[14] The ability of lichen and bryophytes to sequester heavy metals yet remaining unharmed makes them good biomonitors.[15] Indeed, epiphytic lichens have been recognized as indicators of air pollution since the 1800s. Lichens are useful bioindicators, especially where technical instruments are not economically feasible.[16,17]

The most commonly used biomonitoring methods are community analysis, lichen tissue analysis and transplant studies. Anthropogenic activities cause the impoverishment of lichen communities including the local disappearance of the most demanding species. The biomonitoring capability of lichens has been studied in depth for various contaminants.[18] The use of a single species in the same survey is recommended to minimize data variability.[19] Lichens exhibit different levels of sensitivity to pollution depending upon growth forms in the following series: Crustose (flat, tightly adhered, crust-like lichens) < Foliose (leafy lichens) < Fruticose (shrubby lichens), though there are exceptions to the gradation, foliose lichens are better accumulators in comparison to others.[20-21]

Metal absorption employing Lichens as bioindicator: In a developing country like India, biomonitoring is in its preliminary stage and needs more elaborate and extended programs (like Nation-wide projects) to utilize biomonitoring techniques for pollution monitoring in vastly different geographical conditions where employing instruments is not feasible.

In heavy metal and trace element deposition studies, the use of lichen transplants has gained importance owing to the advantage of being a substratum-free surface during a defined period and the possibility of exposing lichens in sites where they are naturally absent. The absorption of metals in lichens involves different mechanisms such as intercellular absorption through an exchange process, intracellular accumulation or entrapment of particles containing heavy metals. Heavy metal content in lichen thallus tends to alternate over time in a phase of accumulation and subsequent release. The metal absorption in lichens is influenced by geographical variations (latitude, longitude and altitude), temporal variations (seasonal changes), acid precipitation, soil dust and local pollution sources (commercial, industrial, vehicular, mining areas).[22]

Based on its application, biomonitoring can be passive and active. The former is based on biomaterial occurring naturally in nature whereas later is based on biomaterials transplanted into research area for a specific period. In India, lichen diversity varies due to the vast geographical area. Therefore, there are different lichen species (based on their distribution and sensitivity) which may be utilized as an indicator species and their indicator value may be further employed in air quality studies of an area of interest.

In India, till date very few biomonitoring studies have been conducted in active biomonitoring studies which have provided the air pollution level of various major cities of India which include Kolkata, Bengaluru, Pune and Lucknow, but not of Mumbai. Hence, the work was carried out in Mumbai but with active monitoring as in Mumbai due to its high pollution, most of the lichens have disappeared except few regions like Aarey Milk Colony, Sanjay Gandhi National Park etc.

In the present study, the level of few common heavy metals such as Al, Ba, Cd, Cr, Fe, Hg, Mn, Ni, Pb, Sn and Zn was determined in selected lichen species using active monitoring by lichen transplant technique.

Material and Methods

Biomonitoring methods: Passive biomonitoring is the most suitable method for carrying out pollution study of any region. But in Mumbai passive biomonitoring cannot be performed as leaving few places, Mumbai city is deprived of lichens. Hence, active biomonitoring by transplanting a lichen collected from the unpolluted location is the best suitable method (Fig. 1-6).

Sampling, identification and transplantation: The lichen species were selected for the active biomonitoring study of Mumbai using transplantation method. Porina Internigrans (Nyl.) Mull. Arg. is easy to collect intact, hence the contamination of the thallus by parts of the substrate is reduced. Lichen samples were collected in acid-free paper bags from different locations of Bhimashankar forests, from same tree barks, together with a small piece of the substrate, at least 1.5 m above the soil, far from any pollution sources. Sampling was done as per literature methods with necessary precautions.[23]

Figure 1-3: Porina Internigrans Lichen on mango tree bark in Bhimashankar forest

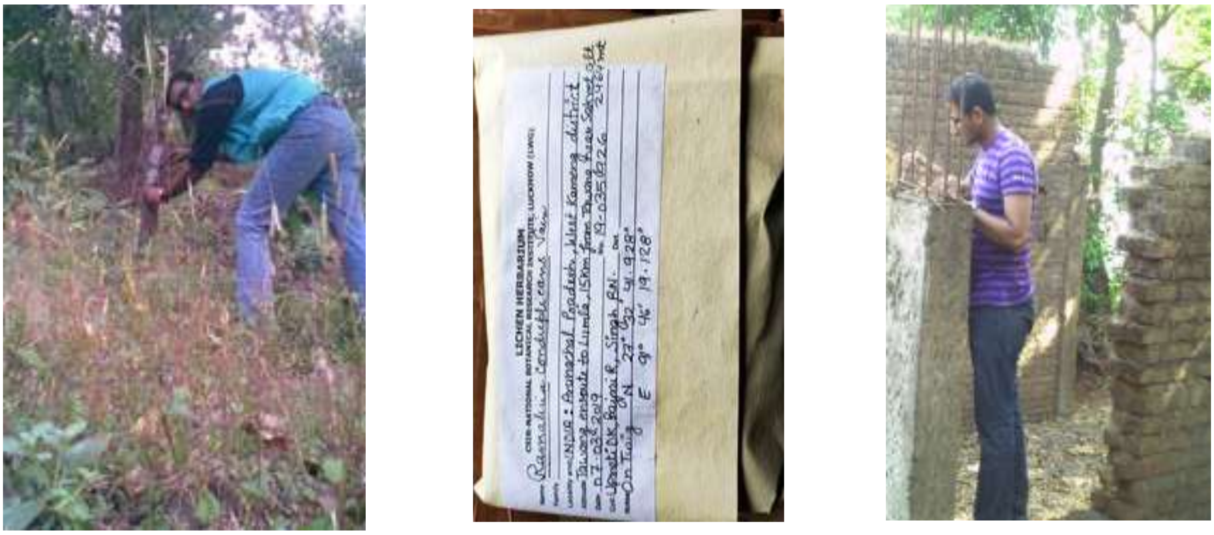

Figure 4-6: Sampling and Transplantation of Porina Internigrans Lichen

After collection, they were immediately packed along with information such as latitude, longitude etc. Sampling was done in December month of each year starting from 2009 to 3 successive years. Only a few samples were collected from a single location to avoid wiping of species from the collection site. (figs. 4-6) Collected lichen sample was identified using colour tests, Thin Layer Chromatography (TLC), morphological and anatomical characteristics. A voucher of the species was deposited in the Lichen Herbarium of National Botanical Research Institute (NBRI), Lucknow (figs. 7-9).

Transplanted lichens were then exposed for three months, January to March. For transplantation, lichen samples were glued with Araldite along with substratum on cardboard and were hung on poles at 5 feet, one facing the source of pollution and one opposite to the source of pollution. Other geographical factors (latitude, longitude & altitude) and location description (residential, commercial and vehicular traffic zones) were also recorded. In total 20 locations were used to determine average levels of heavy metals (Fig. 10).

Elemental analysis using ICP-OES: For analysis, the collected lichen samples were sorted and any adhering particulate matter was carefully removed with plastic tweezers to avoid any metallic contamination from any external source. Then samples were freeze-dried, without washing to avoid losing particles trapped on the lichen surface, pulverized and homogenized with mortar and pestle. 500 mg of dried powdered lichen sample, along with reagent blank, was digested with 6:1 v/v mixture of concentrated HNO_3 and H_2O_2 in microwave digestion system MARSXpress (CEM Corporation). The solution was filtered through Whatman Filter Paper No. 42. The digested solution was diluted and heavy metal content was estimated using ICP-OES (Thermo-iCAP-6000 series) using IAEA-SRM-336 & IRMM-CRM-842 as reference material. Control samples were directly subjected to the same procedure described above without transplantation. Every investigation was done no less than 5 times to accomplish more prominent exactness in results and to acquire factually solid information (Fig. 11).

High Power Microscope | **Garmin – GPS** | **Lichen Removal Tools**

Figure 7-9: Lichen collection Tools / Apparatus

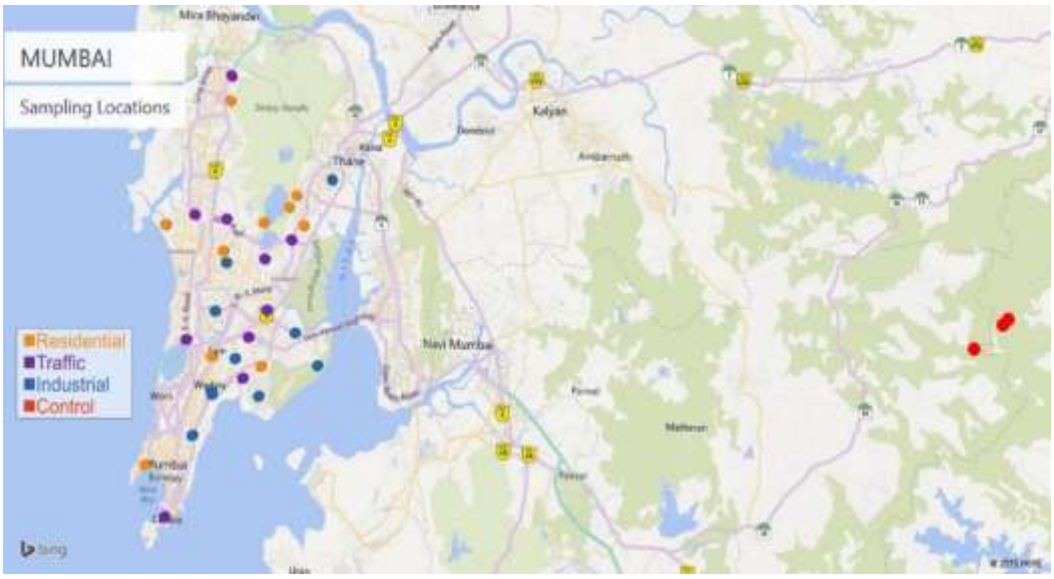

Figure 10: Control & Transplant locations

Results and Discussion

The correlation between exposure time and concentration values was statistically tested for each heavy metal. The exposed-to-control (EC) ratio was calculated using standard methods showing a spatial distribution in the concentration of heavy metals.[24-26] Table 1 shows a range of EC-ratio values which is taken as standard while evaluating the metal contamination of anthropogenic origin. Also, table 2 provides information about the various locations selected for lichen transplantation for active biomonitoring of anthropogenic metals.

All traffic sites showed normal to severe accumulation. We were quite satisfied by reporting that all residential sites showed normal metal concentration and hence EC-ratio was also in the normal range. From table 3 and table 4 we can observe that for traffic sites, order of accumulation of anthropogenic metals was Ni > Fe > Cd > Cr > Mn > Al > Hg = Ba > Zn > Cu > Pb (Fig. 12).

Table 1
EC ratio scale

EC Ratio	Inference
0-0.25	Severe Loss (SL)
0.25-0.75	Loss (L)
0.75-1.25	Normal (N)
1.25-1.75	Accumulation (A)
>1.75	Severe Accumulation (SA)

Figure 11: ICP-AES at Pondicherry University

Table 2
Lichen transplant locations

S.N.	Description of Transplanted Location	S.N.	Description of Transplanted Location
1	Mahada colony, Vashi Naka, Chembur (East)	11	Sindhi Society, Bhakti Bhavan, Chembur (East)
2	DAV College, Bhandup (West)	12	Jain Shivam CHS, Jivdaya Road, Ghatkopar (East)
3	Vikas Tea Shop, Khindipada, Mulund (West)	13	E-Block, Badhwar Park, CPP Marg, Cuff Parade
4	Hanging Garden, Malabar Hill	14	Dairy No. 66, Aarey Colony, Goregaon (East)
5	Mahavir Kripa Jain Society, Sion (West)	15	Sejal Park, Ghartanpada, Dahisar (East)
6	Bhandup Cabin: Vihar Lake Eastern End, Powai	16	Lazeej Biryani, IRB Road, Chandivali, Powai
7	Abhinav Nagar Bus stop, Opposite CTI & RC, Borivli (East)	17	Sagarika CHS, Near Bandra-Worli Sea Link, MWETP, Bandra (West)
8	Mamta Bakery, Lokhandwala Township, Aakurli Road Kandivali (East)	18	Fishing Training Institute, Trombay Jetti, Trombay
9	Amarnath Tower, Fisheries University Road, Varsova andheri (West)	19	Railway Police Training Ground, Mulund (East)
10	NITIE MDP Hostel, Powai	20	Avani Mobile Shop, Kalina, Opp. The University of Mumbai, Santacruz (East)

Table 3
Concentration of different elements analyzed by ICP-AES in ppm

Element	Control	Residential	Traffic	Element	Control	Residential	Traffic
Ba	6.14	6.37	7.29	Cu	4.08	3.37	4.54
Cd	1.14	1.48	1.77	Zn	13.11	12.15	15.05
Cr	1.17	1.18	1.74	Mn	16.98	15.58	20.84
Hg	0.27	0.22	0.32	Al	582.41	522.71	697.9
Ni	1.11	1.09	2.06	Fe	536.47	600.21	922.6
Pb	4.21	3.24	4.14				

Table 4
EC-Ratio of different elements

Element	Residential	Traffic	% Rise	Inference
Ni	0.98	1.86	89.80	High Accumulation
Fe	1.12	1.72	53.57	High Accumulation
Cr	1.01	1.49	47.52	High Accumulation
Al	0.9	1.2	33.33	Accumulation
Cd	1.29	1.55	20.16	Accumulation
Mn	0.92	1.23	33.70	Accumulation
Hg	0.85	1.19	40.00	Accumulation
Cu	0.83	1.11	33.73	Accumulation
Pb	0.77	0.98	27.27	Normal
Zn	0.93	1.15	23.66	Normal
Ba	1.04	1.19	14.42	Normal

In the current investigation, the level of aluminium was normal in residential as well as in traffic zones and the level of barium was in the normal range in residential as well as traffic zones. Barium is used in various industrial processes such as vacuum tubes, spark-plug alloys, getter alloys, fray's metal and as a lubricant for anode rotors in X-ray machines. We observed that lichens showed a normal level of cadmium in residential zones but accumulated high level in traffic zones. Cadmium is released into the environment through mining and smelting, usage of phosphate fertilizers, plating, steel production, pigments and burning of fossil fuels such as coal or oil and incineration of municipal waste such as plastics and nickel-cadmium batteries.

Similar to cadmium, lichens showed a normal level of chromium in residential sites but accumulated high level in traffic zones. Chromium originates mainly from coal combustion, manufacture of steel, incineration, oil combustion for electric generation and cement production.

Lichen accumulates iron more readily than most of the other metals in most of the transplanted sites. We observed that levels of iron were at a normal level in residential sites but accumulated high level in traffic zones. Iron is released into the environment from natural deposits, industrial wastes, refining of iron ores and corrosion of materials made up of iron and its alloys. Mercury showed the least variation in its concentration and was quite low in all sites. Levels of mercury in the environment are increasing due to discharge from hydroelectric, mining, pulp and paper industries. Incineration of municipal and medical waste and emission from coal-fired power plants also contribute to it. The level of magnesium was well below the normal range in residential as well as traffic zones.

Manganese compounds occur in many rocks and soils. It is also released into the environment iron and steel production plants, power plants and coke ovens. Nickel, a by-product of coal combustion, industrial processes and automobile exhaust was highest in traffic sites which completely depicts its cause whereas it was well below normal in residential transplant locations.

Results confirm that lichens are very efficient accumulators of lead. We observed that levels of lead were in the normal range in residential sites and traffic zones. Lead enters into the environment as a pollutant through toys, dust and chips from old paints, traditional medicines, car exhaust, metal smelting, industries using lead compounds and battery manufacturing. Copper showed normal accumulation in all transplanted lichens except some industrial locations where it was well above normal level but not severe.

The main anthropogenic source of copper is from coal-fired power stations, metal production, waste incineration, sewage treatment processes, plumbing, utensils, food preservatives, as an anti-fouling agent in paints and application of agricultural chemicals. The level of zinc was well below the normal range in residential as well as traffic zones. Zinc emission occurs from zinc smelters and automobile exhaust. High level of zinc confirms its anthropogenic origin. The maximum concentration of Zn was less than many other metals studied (Figures 12-15).

Conclusion

In conclusion, the use of lichens for transplants and the measurements allowed the interpretation of the accumulation trends. The initial content has to be measured before each exposure experiment, as an inter-annual variability can be observed, mainly for anthropogenic metals. In the course of this study, some high levels of metals were observed which are attributed to discrete pollution events that occurred a short time before sampling. This study confirms the ability of lichen transplants for active monitoring.

The present level of pollutants will be a useful baseline for future studies of the ambient air in Mumbai city. Also, through this study, it can be proposed that in-line with many European and other developed and developing countries, biomonitoring can be used periodically to check air-quality in different cities of India.

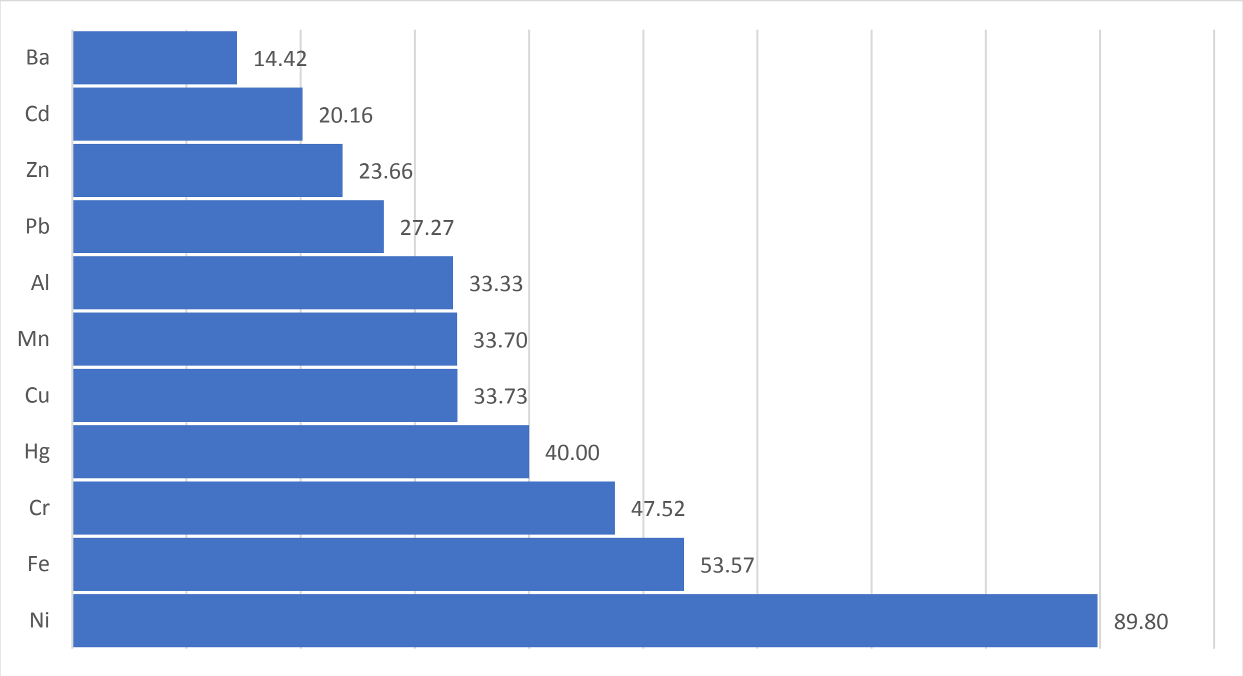

Figure 12: Percentage increase in accumulation of anthropogenic elements

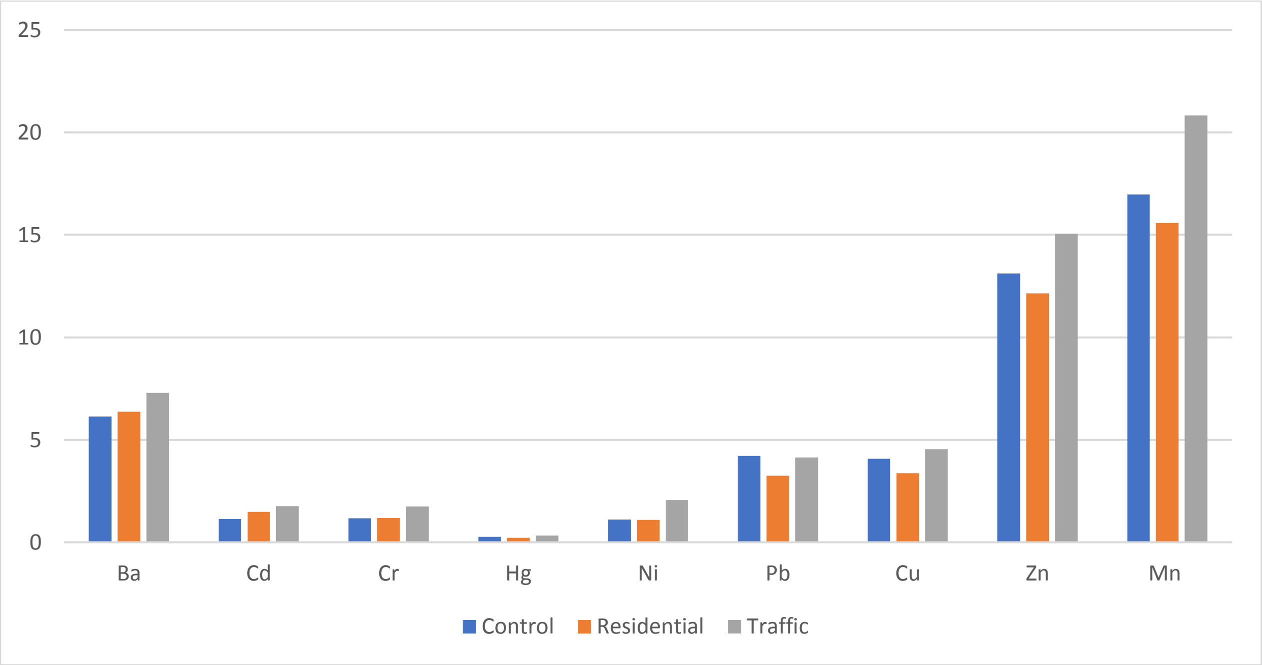

Figure 13: Concentration of Micro-level elements in ppm

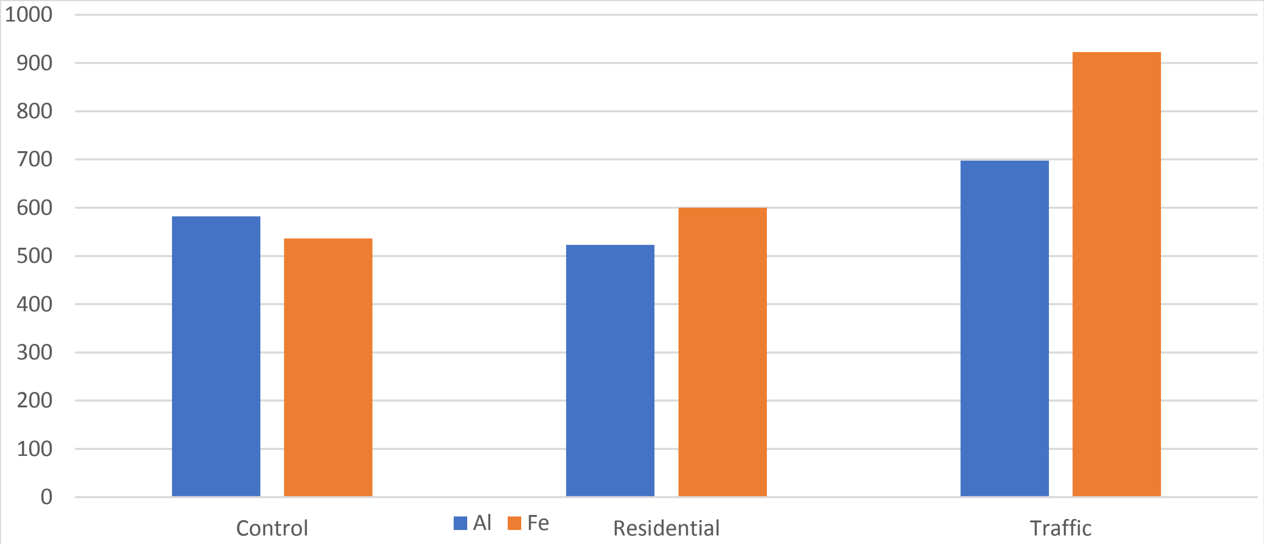

Figure 14: Concentration of Macro-level elements in ppm

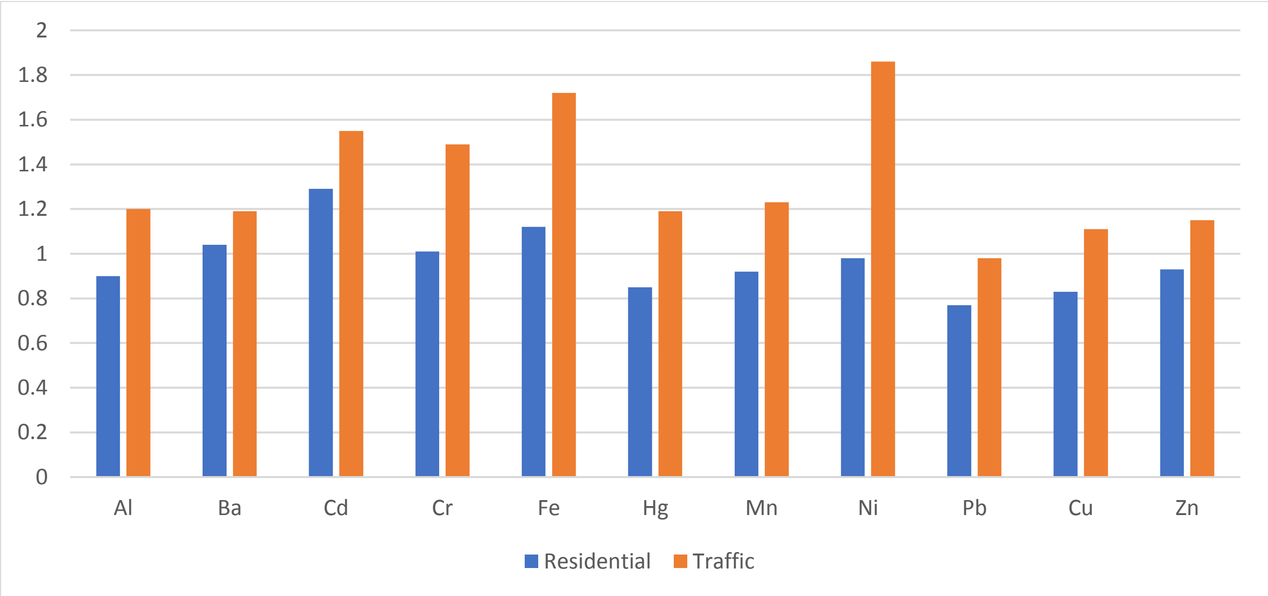

Figure 15: Comparison of EC ratio of elements

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References

1. Alaimo M.G., Dongarrà G., Melati M.R., Monna F. and Varrica D., Recognition of environmental trace metal contamination using pine needles as bioindicators - The urban area of Palermo (Italy), *Environmental Geology*, **39(8)**, 914–924 **(2000)**

2. Bačkor M. and Loppi S., Interactions of lichens with heavy metals, *Biologia Plantarum*, **53(2)**, 214–222 **(2009)**

3. Bajpai R. and Upreti D.K., New records of lichens of Mahabaleshwar and Koyna areas of Satara District, Maharashtra, India, *Geophytology*, **40(1-2)**, 61–68 **(2011)**

4. Blasco M., Domeno C., Lopez P. and Nerin C., The behaviour of different lichen species as biomonitors of air pollution by PAHs in natural ecosystems, *Journal of Environmental Monitoring*, **13(9)**, 2588–2596 **(2011)**

5. Brodo I.M., Substrate Ecology, The Lichens, New York, Academic Press, 401–441 **(1973)**

6. Frati L., Brunialti G. and Loppi S., Problems Related to Lichen Transplants to Monitor Trace Element Deposition in Repeated Surveys: A Case Study from Central Italy, *Journal of Atmospheric Chemistry*, **52(3)**, 221–230 **(2005)**

7. Garty J., Biomonitoring Atmospheric Heavy Metals with Lichens: Theory and Application, *Critical Reviews in Plant Sciences*, **20(4)**, 309–371 **(2001)**

8. Hawksworth D.L. and Rose F., Qualitative Scale for estimating Sulphur Dioxide Air Pollution in England and Wales using Epiphytic Lichens, *Nature*, **227(5254)**, 145–148 **(1970)**

9. Kranner I., Beckett R. and Varma A., Protocols in Lichenology, Berlin, Heidelberg, Springer Berlin Heidelberg, 580 **(2002)**

10. Loppi S., Pirintsos S. and De Dominicis V., Soil Contribution to the Elemental Composition of Epiphytic Lichens (Tuscany, Central Italy), *Environmental Monitoring and Assessment*, **58(2)**, 121–131 **(1999)**

11. Martin E. and Hine R., A Dictionary of Biology, Oxford University Press **(2015)**

12. Minganti V., Capelli R., Drava G., De Pellegrini R., Brunialti G., Giordani P. and Modenesi P., Biomonitoring of Trace Metals by Different species of Lichens (Parmelia) in North-West Italy, *Journal of Atmospheric Chemistry*, **45(3)**, 219–229 **(2003)**

13. Monna F., Aiuppa A., Varrica D. and Dongarra G., Pb Isotope Composition in Lichens and Aerosols from Eastern Sicily: Insights into the Regional Impact of Volcanoes on the Environment, *Environmental Science and Technology*, **33(15)**, 2517–2523 **(1999)**

14. Nash T.H., Lichen Biology, 2nd Edition, Cambridge University Press **(2008)**

15. Nieboer E., Lichens as monitors of atmospheric deposition, Atmospheric inputs of pollutants to natural waters, Michigan: An Arbor Science Publishing, 339–388 **(1981)**

16. Purvis O.W., Dubbin W., Chimonides P.D.J., Jones G.C. and Read H., The multi-element content of the lichen Parmelia sulcata, soil and oak bark in relation to acidification and climate, *Science of The Total Environment*, **390(2–3)**, 558–568 **(2008)**

17. Bergagli R., Trace elements in terrestrial plants, an ecophysiological approach to biomonitoring and biorecovery, Verlag, Berlino: Springer-Verlag, 324 **(1998)**

18. Chaffer R.J., The Weathering of natural stones, Atmospheric Input of Pollutants to Natural Waters: Symposium : 178th National Meeting : Extended Abstracts, London: Chemistry, American Chemical Society, Division of Environmental **(1972)**

19. Howell R.K., Influence of air pollution on quantities of caffeic acid isolated from leaves of Phaseolus vulgaris, *Phytopathology*, **60(11)**, 1626–1629 **(1970)**

20. Seaward M.R.D., Lichens and Sulphur dioxide air pollution: field studies, *Environmental Reviews*, **1(2)**, 73–91 **(1993)**

21. Shukla V., Upreti D.K. and Bajpai R., Lichens to Biomonitor the Environment, Springer India **(2015)**

22. Shukla V., Patel D.K., Upreti D.K., Yunus M. and Prasad S., A comparison of metallic contents in lichen Pyxine subcinerea, its substratum and soil, *International Journal of Environmental Science and Technology*, **10(1)**, 37–46 **(2013)**

23. Bargagli R. and Nimis P.L., Guidelines for the use of Epiphytic Lichens as Biomonitors of Atmospheric Deposition of Trace Elements in Nimis P., Scheidegger C. and Wolseley P., Eds., Monitoring with Lichens — Monitoring Lichens, Dordrecht: Springer Netherlands, 295–299 **(2002)**

24. Singh K.P. and Sinha G.P., Indian lichens an annotated checklist, Botanical Survey of India **(2010)**

25. Frati L., Brunialti G. and Loppi S., Problems Related to Lichen Transplants to Monitor Trace Element Deposition in Repeated Surveys: A Case Study from Central Italy, *Journal of Atmospheric Chemistry*, **52(3)**, 221-230 **(2005)**

26. St. Clair S.B., St. Clair L.L., Mangelson N.F. and Weber D.J., Influence of growth form on the accumulation of airborne copper by lichens, *Atmospheric Environment*, **36(36–37)**, 5637–5644 **(2002)**

27. St. Clair S.B., St. Clair L.L., Weber D.J., Mangelson N.F. and Eggett D.L., Element Accumulation Patterns in Foliose and Fruticose Lichens from Rock and Bark Substrates in Arizona, *The Bryologist*, **105(3)**, 415–421 **(2002)**

28. Upreti D.K., Taxonomic, Pollution monitoring and ethnolichenological studies on Indian lichens, *Phytomorphology*, **51**, 477–497 **(2001)**

29. Upreti D.K., Agarwal S.K., Kaushik J.P. and Kaul K.K., Diversity of lichens in India, Perspective in Environment, 71–79 **(1998)**

30. We T., The Algal Partner, CRC Handbook of Lichenology, Boca Raton, CRC Press, 39–92 **(1988)**.

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