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Studies on diversity of lichen, *Pyxine cocoes* to air pollution in Bhadravathi town, Karnataka, India

Naveen Danesh[1]*, E.T. Puttaiah[1] and B.E.Basavarajappa[2]

[1]*Department of Environmental Science, Gulbarga University, Gulbarga-585 106, India*

[2]*Department of Chemistry/ESTSC, Bapuji Institution of Engineering and Technology, Davanagere-577 004, India*

**Corresponding Author email : naveeanu@gmail.com*

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Abstract

Air pollution induced climate change affecting the pigmentation and diversity of lichen, *Pyxine cocoes* were monitored around the industrial area and traffic area of Bhadravthi using European guidelines. The obtained data has been discussed and results compared with data from that of Kuvempu University campus (control). From the present study, it was evident that the air pollutants emitted from the two major industries and other small scale industries affected the total chlorophyll (0.16 mg g^{-1}) and carotene pigments (0.11 mg g^{-1}) in *Pyxine cocoes*, as well as their diversity (approx 13) on two plants (*M. indica* and *P. pinnata*) in the vicinity of the industrial area. Further, as a result of vehicular pollution at traffic area resulted in the deterioration of total chlorophyll (0.11 mg g^{-1}), carotene pigments (0.07 mg g^{-1}) and diversity (approx. 17) of *Pyxine cocoes* compared to control site. The present study has thrown light on lichens sensitivity to the air pollution.

Key words
Epiphytic lichen, *Pyxine cocoes,* Air pollution

Introduction

The increase in atmospheric emissions from industrial and human sources, has led to an ever-increasing of atmospheric pollutant levels. Although appearing to be a single organism, a lichen is actually a symbiotic partnership between a fungus and one or more photosynthetic organisms, an alga or cyanobacterium (Dobson, 2000). Epiphytic lichens are very sensitive to air pollution and an alteration in air quality directly affects the lichen diversity; therefore, they are generally considered as good indicators of air quality (Crespo *et al.*, 2004; Shukla and Upreti, 2011). Lichens accumulates and releases nutrients (nitrogen and phosphorous) on to nutrient-poor soils in northern forests becoming an important aspect in sustaining ecosystems (Purvis, 2000). Since the lichen plots are re-measured periodically, trend analyses can indicate changes in lichen communities brought about by changes in climatic conditions or other environmental stressors (e.g air pollution or forest fragmentation). Because of their sensitive physiology, lichens are more likely to be affected by environmental changes than other plants (Bargagli *et al*., 1987; Taufikurahman *et al*., 2010). Lichens are highly valued ecological indicators known for their sensitivity to a wide variety of environmental stressors like air quality and climate change (Jovan, 2008).

The use of lichens in biomonitoring of particulate pollutants has gained increasing acceptance in recent years. Lichen biomonitoring is especially useful in urban areas, where high density of different emitting sources make monitoring of air pollution with conventional chemico-physical gauge an extremely difficult task due to variety of pollutants (Rout, 2010). A number of parameters are used to estimate the effect of air pollution on lichens (Ronen and Galun, 1984). Chlorophyll content and chlorophyll degradation are parameters commonly used to assess the impact of air pollution on lichens. The most obvious sign of pollution damage to lichens is bleaching of the thalli, caused by decomposition of chlorophyll. Metallic pollutants are known to disrupt the vital physiological processes (Shukla and Upreti, 2007). Chlorophyll in lichens is very sensitive to changes in environmental factors including air pollution (Boonpragob, 2002).

Air quality of the study area, abundance of lichens and concentration of chlorophyll pigments and carotene in epiphytic lichen *P.cocoes* at industrial area and traffic intersection (Rangappa circle) of Bhadravathi town were compared with that of a control area (Kuvempu University campus) keeping in view the aim to determine the effect of air pollution on lichen *P.cocoes*.

Materials and Methods

Study area : Bhadravathi is an industrial town situated at 13°49' 46" N to 75°42' 22" E in the Shivamogga district about 255 km from Bangalore of Karnataka, India. Two large-scale industries (Visweswaraya Iron and Steel Plant and Mysore Paper Mills Ltd.) in addition to numerous small-scale industries and number of vehicles plying on the poorly paved roads pollute the town.

Ambient air quality monitoring : Ambient air quality monitoring followed standard methods of the National Ambient Air Quality Monitoring (CPCB, 2003-04). Air sampling was carried out using APM-410 and APM-411 high volume air samplers make. The sampling frequency was done for 24 hrs, twice a week at uniform intervals and for a period of 2 consecutive years (July 2006 to June 2008) *i.e.*, overall 192 samples were taken.

Determination of suspended particulate matter (SPM) : SPM was measured by weight/volume and the mass/ quantity of SPM. In each case it was determined by weighing the filter paper before and after sampling with proper equilibrium each time (ISM, 1976).

Determination of sulphur dioxide (SO_2) : The ambient air was bubbled through the aqueous solution of potassium tetrachloromercurate (TCM) and the absorbance of the solution was measured by means of a spectrophotometer (Systronics 367, India) using the modified method of West and Geake (1956).

Determination of nitrogen oxides (NO_x) : The gas was collected in the absorber of the air sampler and the mixture was analyzed with the sodium arsenite method (Jacob and Hochheiser, 1958) by measuring the absorbance of highly colored azo-dye at 540 nm using a spectrophotometer (Systronics 367, India).

Epiphytic lichen diversity monitoring : European guidelines (Asta *et al*., 2002) were adopted to study the diversity of lichen on a defined portion of tree bark of *M.indica* and *P.pinnata* for 24 months at a frequency of one analysis each month. A monitoring quadrate consisting of four independent quadrate segments of five 10 × 10 cm^2 each (Fig. 1) was attached vertically to the tree trunk on North , East, South and West in such a manner that the lower edge of each segment was 100 cm above the ground. This was adopted as in urban centers where lichen cover is often restricted at the base of trees.

Pigment analysis : The chlorophyll content was calculated from absorbance values at 663 and 645 nm following the method of Arnon (1949). The total carotenoid content was calculated according to Parsons (1984) from absorbance values at 480 and 510 nm using spectrophotometer (Systronics 367, India).

Data pertaining to the air pollutants were subjected to one-way multifactorial analysis of variance (ANOVA) in order to determine significant differences between means ($P < 0.001$).

Results and Discussion

At both industrial and traffic sites of Bhadravathi town, the concentration of air pollutants was within the threshold limit for SPM (360 μg m^{-3}), SO_2 (80 μg m^{-3}) and NO_x (80 μg m^{-3}) prescribed by the Central Pollution Control Board (CPCB, 2004), but in comparison with control site (Table 1), air pollution was very high in the study area. Among the air pollutants, the annual average of 2006-07 and 2007-08 for SPM concentration was comparatively higher (236.56 and 232.30 μg m^{-3}) than the concentrations of NO_x (19.69 and 19.15 μg m^{-3}) and SO_2 (13.23 and 13.63 μg m^{-3})

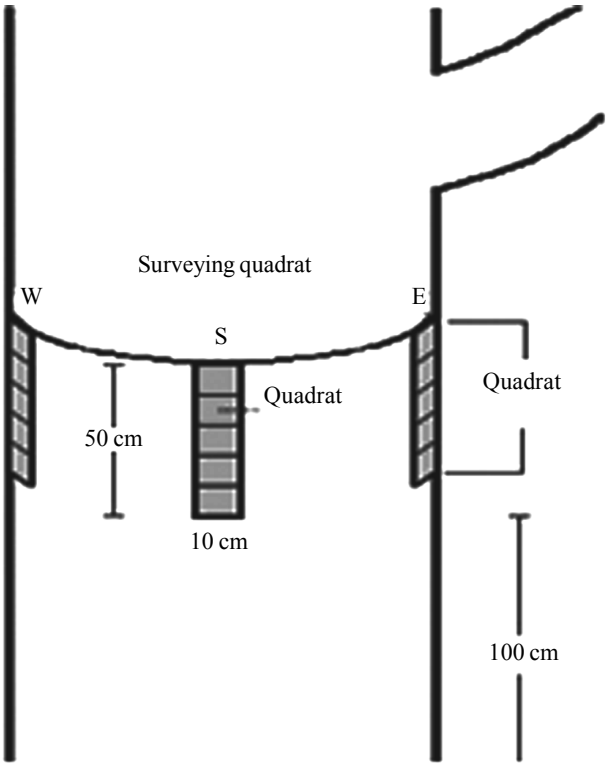

Fig. 1 : Recording quadrate composed of four quadrate segments each with five squares on the bark of trees *M.indica* and *P.pinnata*

respectively at industrial site. Traffic site also showed the same trend witnessing concentration of SPM (207.16 and 202.60 μg m^{-3}) higher than the concentrations of NO_x (17.97 and 18.51 μg m^{-3}) and SO_2 (8.91 and 10.57 μg m^{-3}) respectively for the respective years of the study period.

In all, a total of 9 genera of epiphytic lichens were identified: *Pyxine*, *Chrysothrix*, *Parmotrema*, *Teloschistes*, *Dirinaria*, *Graphis*, *Ramalina*, *Lecanora* and *Heterodermia*. In comparison, all 9 genera of epiphytic lichens were present at the control site, but lichens belonging to the genera *Ramalina* and *Teloschistes* were absent on *M. indica* at both the industrial and traffic site. Species of genera *Teloschistes*, *Ramalina* and *Heterodermia* were absent on the bark of *P. pinnata* while only six genera were found. These results suggest that the control site had high diversity of lichens with frequencies of 118 and 132 on the barks of *M. indica* and *P. pinnata*, respectively. Lichen species *Pyxine cocoes* (29) dominated while *Parmotrema* spp. (5) showed the least diversity on *M. indica*. *Dirinaria* spp. (21) was dominant while *Teloschistes* spp. had least diversity on *P. pinnata* (Table 2).

At industrial site, *Pyxine cocoes* (21 and 15) was most common on the bark of *M. indica* and *P. pinnata* and *Heterodermia* (01) and *Parmotrema* (01) were found in least numbers on *M.indica*. On *P. pinnata, lichen species Chrysothrix* noticed least diversity of 1. The total sum of frequencies of lichen genera were 42 and 41 on *M. indica* and *P. pinnata* plants indicating lichen diversity. On the other hand, at traffic site *Pyxine cocoes* (19 and 13) was most common on the bark of *M. indica* and *P. pinnata*. In contrast, lichen species *Dirinaria* (02) and *Lecanora* (2) had least diversity respectively on *M. indica* and *P. pinnata* plants. The total sum of frequencies of lichen genera were 52 and 37 on *M. indica* and *P. pinnata* plants. Industrial and traffic sites had LDV of 41.7 and 42.3 respectively, about three times lesser than the control site (122).

Direction wise lichen diversity on both plants *M.indica* and *P. pinnata* also showed variation at all the sampling sites. At Industrial site, the lichen diversity was high on East direction with 22 lichens on *M.indica* with an average of 2.44, followed by 14,5 and 3 lichens on North, South and West directions with average of 1.5, 0.55 and 0.33, respectively. On the other hand, 14 lichens found on South and West directions of *P. pinnata* plant with the average of 1.55 and 1.55 respectively, while 7 and 6 lichens were noticed on North and East sides of the plant with on average 0.77 ± and 0.66, respectively.

Lichen diversity at traffic area on four directions of *M. indica* and *P. pinnata* plants also showed varied results. In North direction 22 lichens were found with an average of 3.77 followed by 16, 9 and 7 on South, East and West directions with average of 2.86, 1.11 and 0.97, respectively. In contrast, on *P. pinnata* 18 lichens were identified with average of 2.78 on East side of the plant while, 9, 7 and 5 lichens on West, North and South directions with averages of 1.50, 0.97 and 1.50, respectively.

Lichens on both *M.indica* and *P.pinnata* plants were found to be more at control site compared to the industrial and traffic sites. In the North direction of the *M. indica* plant, 45 lichens were recorded with average of 4.47. On the other hand, 25, 25 and 23 were found on East,West and South directions of the *M. indica* respectively with 3.23, 2.58 and 2.50. On comparison, East side of the *P. pinnata*, the highest number (53) of lichens were noticed overall with an average of 3.33. On North, South and West sides of the plant 29, 23 and 27 lichens were noticed with an average of 2.16, 1.87 and 2.87, respectively.

Table 3 shows the value of quantified parameters corresponding to the variation of concentrations from industrial and traffic sites to control site. From the observation it is clear that chlorophyll contents in *P.cocoes* are found to be lesser in industrial area (0.22, 0.14 and 0.36 mg g^{-1}) compared to control area (0.36, 0.26 and 0.52 mg g^{-1}). On the other hand, carotenoid reduced by 28.20% from control area to industrial area. Traffic area too witnessed deterioration of chlorophyll *a*, chlorophyll *b*, total chlorophyll (0.27, 0.14 and 0.41 mg g^{-1} f.wt. respectively) and carotenoid (0.32 mg g^{-1} f.wt.) in *P.cocoes* in contrast to control site.

Among all the lichen species used in India, *Pyxine cocoes* is found to be more toxitolerant and suitable for bio-monitoring studies (Shukla and Upreti, 2007; 2008; Bajpai *et*

Table 1 : Annual concentrations of SPM, SO_2 and NOx (μg m^{-3}) of industrial, traffic and control site of Bhadravathi town

Sampling sites	2006-07			2007-08		
	SPM	SO_2	NO_x	SPM	SO_2	NO_x
Control	20.08 ± 11.91*	0.07± 0.09*	0.35 ± 0.36*	20.15± 16.14*	0.033 ± 0.06*	0.28 ± 0.31*
Industrial	236.56 ± 83.22*	13.23 ± 4.75*	19.69 ± 7.88*	232.30 ± 81.63*	13.62 ± 6.09*	19.15± 6.88*
Traffic	207.16 ± 88.38*	8.91 ± 4.37*	17.97 ± 9.40*	202.60 ± 85.35*	10.57 ± 4.29*	18.51± 8.18*

Values are standard deviation of 96 analysis; *Significant at P<0.0001, number of samples analyzed are given in parenthesis

Table 2 : Variation in lichens diversity at control and industrial area of Bhadravathi town

Lichen	*M.indica*				*P. pinnata*			
	North	East	South	West	North	East	South	West
Pyxine cocoes	08 ± 0.81 IS	10 ± 1.41 IS	02 ± 0.00 IS	01 ± 0.81 IS	01 ± 0.81 IS	02 ± 0.00 IS	08 ± 1.41 IS	04 ± 3.55 IS
	16 ± 4.96 CS	04 ± 0.81 CS	04 ± 0.81 CS	05 ± 0.81 CS	04 ± 0.00 CS	12 ± 1.41 CS	02 ± 0.00 CS	02 ± 0.00 CS
	04±0.00 TS	02±0.81 TS	08±1.41 TS	05±4.24 TS	03 ± 0.10 TS	08± 1.63 TS	01±0.00 TS	01± 0.81 TS
Chrysothrix	02 ± 0.81 IS	03 ± 0.81 IS	-- IS	-- IS	01 ± 0.00 IS	-- IS	-- IS	-- IS
	03 ± 0.00 CS	04 ± 0.00 CS	02 ± 0.81 CS	-- CS	01 ± 0.00 CS	05 ± 0.00 CS	02 ± 0.00 CS	08 ± 1.63 CS
	01±0.00 TS	-- TS	-- TS	01±1.41 TS	01±0.00 TS	02± 1.63 TS	-- TS	03± 0.00 TS
Parmotrema	-- IS	01 ± 0.81 IS	-- IS	-- IS	03 ± 0.00 IS	02 ± .00 IS	01 ± 0.81 IS	04 ± 1.41 IS
	03 ± 0.00 CS	-- CS	-- CS	02 ± 0.81 CS	06 ± 0.81 CS	03 ± 0.00 CS	04 ± 0.00 CS	-- CS
	01±0.81 TS	03±0.81 TS	01±1.41 TS	01±1.41 TS	01±0.81 TS	02± 1.63 TS	-- TS	04± 0.81 TS
Teloschistes	-- IS	-- IS	-- IS	-- IS	-- IS	-- IS	-- IS	-- IS
	02 ± 0.81 CS	-- CS	-- CS	07 ± 2.12 CS	02 ± 0.00 CS	02 ± 0.00 CS	-- CS	01 ± 0.00 CS
	-- TS	-- TS	-- TS	-- TS	-- TS	-- TS	-- TS	-- TS
Dirinaria	01 ± 0.81 IS	01 ± 0.81 IS	-- IS	-- IS	-- IS	01 ± 0.81 IS	02 ± 0.81 IS	-- IS
	04 ±0.00 CS	02 ± 1.63 CS	06 ± 0.81 CS	-- CS	03 ± 0.00 CS	09 ± 0.00 CS	03 ± 1.41 CS	06 ± 0.00 CS
	01±0.81 TS	01±0.81 TS	-- TS	-- TS	-- TS	01± 0.81 TS	02± 0.00 TS	-- TS
Graphis	01 ± 0.00 IS	05 ± 0.81 IS	01 ± 0.81 IS	02 ± 0.81 IS	01 ± 0.00 IS	01 ± 2.15 IS	01 ± 0.81 IS	06 ± 0.81 IS
	03 ± 0.00 CS	06 ± 0.00 CS	-- CS	04 ± 1.63 CS	-- CS	02 ± 0.00 CS	04 ± 0.00 CS	04 ± 1.63 CS
	12±3.20 TS	02±1.60 TS	05±0.81 TS	03±0.00 TS	01±0.00 TS	05± 2.44 TS	01± 0.81 TS	01± 0.81 TS
Ramalina	-- IS	-- IS	-- IS	-- IS	-- IS	-- IS	-- IS	-- IS
	08 ± 1.63 CS	-- CS	04 ± 0.81 CS	-- CS	05 ± 0.81 CS	07 ± 0.00 CS	01 ± 0.00 CS	-- CS
	-- TS	-- TS	-- TS	-- TS	-- TS	-- TS	-- TS	-- TS
Lecanora	01 ± 0.81 IS	02 ± 0.81 IS	-- IS	-- IS	01 ± 0.81 IS	-- IS	02 ± 0.81 IS	-- IS
	03 ± 0.81 CS	-- CS	01 ± 0.00 CS	05 ± 0.81 CS	06 ± 1.63 CS	06 ± 0.81 CS	06 ± 0.95 CS	01 ± 0.81 CS
	02±1.40 TS	01±0.00 TS	02±1.60 TS	01±0.81 TS	01±1.41 TS	-- TS	01± 0.81 TS	-- TS
Heterodermia	01 ± 0.81 IS	-- IS	-- IS	-- IS	-- IS	-- IS	-- IS	-- IS
	03 ± 0.00 CS	09 ± 2.16 CS	06 ± 0.81 CS	02 ± 00 CS	02 ± 0.00 CS	07 ± 0.00 CS	01 ± 1.41 CS	05 ± 0.81 CS
	01±0.00 TS	-- TS	-- TS	02±0.0 TS	-- TS	-- TS	-- TS	-- TS
Sum of frequencies	14 ± 2.45 IS	22 ± 3.21 IS	3 ± 0.71 IS	3 ± 1.12 IS	7 ± 0.98 IS	6 ± 1.51 IS	14 ± 2.52 IS	14 ± 2.56 IS
	43 ± 4.55 CS	25 ± 3.19 CS	23 ± 2.45 CS	25 ± 2.63 CS	29 ± 2.15 CS	53± 3.22 CS	23 ± 1.96 CS	27 ± 2.84 CS
	22±0.70 TS	9 ± 0.61 TS	10±0.65 TS	11±0.54 TS	4±0.70 TS	18±0.2 TS	6±0.4 TS	9±0.5 TS

Variation of lichen diversity is shown in parenthesis (IS:Industrial Site, TS:Traffic Site and CS:Control Site; Values are mean ± SD

al., 2010). *P. cocoes* is a foliose lichen and has bigger size thallus, often found abundantly growing on trees and rocks in the tropical areas of India. From the present study, it was evident that the diversity of lichens and pigment profile of *P.cocoes* differed between the industrial and traffic sites with the control site. The variation in response could be directly attributed to the emissions from two large industries, number of small-scale industries and vehicular pollution at the study area. This observation is in confirm it with the earlier studies on lichens at Pauri City, Uttaranchal, India (Shukla and Upreti, 2007). A qualitative survey of the epiphytic lichens in the surroundings of Ulan Bator in October 2007 also showed similar trends (Hauck, 2009). Gombert *et al*. (2006) found lichens and tobacco plants as complementary biomonitors of air pollution in the Grenoble area of Isere, southeast France. Rout *et al*. (2010) found varied response of pigment profile and chlorophyll degradation of *Pyxine cocoes* lichen to air pollution scenario at Cachar district, Assam, India. Shukla and Upreti (2010) found lichen genera *Pyxine* to be tolerant to an unfavorable environment and exhibited luxuriant growth. A global increase in members of Physciaceae family (including *Phaeophyscia* and *Pyxine*) has been linked to climate change (van Herk *et al.,* 2002).In the present study, *P.cocoes* was the dominant lichen found at all the sites.

Dolney *et al*. (2009) found more than 20 lichen species Pennsylvania, US among all sample plots with the two species (*A. palmulata* and *P. squarrosa*) being sensitive to air pollution. It has been found that the epiphytic lichens are the first to be affected by environmental contamination and the fruticose lichens are the most sensitive towards

Table 3 : Variations in concentrations of pigments (mg g^{-1} f.wt.) in *P.cocoes* at sampling sites

Sampling sites	Chlorophyll *a*	Chlorophyll *b*	Total chlorophyll	Carotenoid
Control	0.36 ±0.06	0.26 ±0.20	0.52±0.30	0.39 ±0.30
Industrial	0.22 ±0.02	0.14 ±0.00	0.36 ±0.20	0.28±0.20
Traffic	0.27 ±0.10	0.14 ±0.30	0.41 ±0.14	0.32 ±0.15

Values are mean of 24 replicates ± SD

pollution followed by foliose and crustose forms (Awasthi, 2000). The same trend was seen in the present study. The fruiticose lichens, *Teloschistes* and *Ramalina* were found to be very sensitive. Nayaka *et al*. (2003) reported a correlation between air pollution induced environmental changes and lichen diversity in Bangalore.

The present study provides the information on how the industrial and vehicular air pollution affects the lichen diversity and the pigment distribution at one of the polluted town of Karnataka. The absence of naturally appearing lichens in polluted areas limits the spatial differences of polluted areas. Further, the results obtained will be baseline data for impact assessment at the study area Bhadravathi, which is nearer to the Western Ghat. Lisowska (2011) found the recolonisation of the former 'lichen desert' in the town centre, Poland where, species richness of lichens at study sites has increased with an improvement in the health of lichen thalli was noted and correlated with air quality improvement, mainly SO_2 decline in the last few decades and transport-related compounds, mainly NO_x and dust that have became the main pollutants in Polland. Keeping in view of the biomonitoring of air pollution using lichens, the present study is of greater importance. The present study suggests the need of improvement in the air quality by proper management at Bhadravathi to save the lichens.

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