

# *Rinodina sophodes* (Ach.) Massal.: a bioaccumulator of polycyclic aromatic hydrocarbons (PAHs) in Kanpur City, India

Satya · Dalip K. Upreti · D. K. Patel

Received: 18 November 2010 / Accepted: 11 February 2011 / Published online: 5 April 2011  
© Springer Science+Business Media B.V. 2011

**Abstract** The aim of this study is to determine the possibility of using *Rinodina sophodes* (Ach.) Massal., a crustose lichen as polycyclic aromatic hydrocarbons (PAHs) bioaccumulator for evaluation of atmospheric pollution in tropical areas of India, where few species of lichens are able to grow. PAHs were identified, quantified and compared to evaluate the potential utility of *R. sophodes*. The limit of detection for different PAHs was found to be 0.008–0.050  $\mu\text{g g}^{-1}$ . The total PAHs in different sites were ranged between  $0.189 \pm 0.029$  and  $0.494 \pm 0.105 \mu\text{g g}^{-1}$ . The major sources of PAHs were combustion of organic materials, traffic and vehicular exhaust (diesel and gasoline engine). Significantly higher concentra-

tion of acenaphthylene and phenanthrene indicates road traffic as major source of PAH pollution in the city. Two-way ANOVA also confirms that all PAHs content showed significant differences between all sampling sites (P 1%). This study establishes the utility of *R. sophodes* in monitoring the PAHs accumulation potentiality for development of effective tool and explores the most potential traits resistant to the hazardous environmental conditions in the tropical regions of north India, where no such other effective way of biomonitoring is known so far.

**Keywords** *Rinodina sophodes* · PAHs · Bioaccumulation · Traffic level · Kanpur City · India

---

Satya  
Molecular Bio-prospection Department,  
Central Institute of Medicinal and Aromatic Plant  
(CIMAP-CSIR), Near Kukrail Picnic Spot,  
Lucknow 226015, India  
e-mail: satyalichen@gmail.com

D. K. Upreti (✉)  
Lichenology Laboratory, National Botanical  
Research Institute (CSIR), Rana Pratap Marg,  
Lucknow 226001, UP, India  
e-mail: upretidk@rediffmail.com

D. K. Patel  
Analytical Chemistry Section,  
Indian Institute of Toxicology Research (CSIR),  
Mahatma Gandhi Marg, Lucknow 226001, India

## Introduction

Polycyclic aromatic hydrocarbons (PAHs) are widespread environmental pollutants; they are formed during incomplete combustion and pyrolysis of organic materials, from both natural and environmental sources (Marr et al. 1999). The name “polycyclic aromatic hydrocarbons” commonly refers to a large class of organic compounds contains two or more aromatic (benzene) rings which are fused together when a pair of carbon atoms is shared between them (Guidotti et al. 2003).

Several findings also suggest that the PAHs have a pyrogenic origin and confirm that a traffic vehicle has a strong influence on PAH contamination in the area (Ma et al. 2005; Budzinski et al. 1997). The highest concentrations of atmospheric PAHs are usually found in urban areas due to increase in vehicular traffic and the scarce dispersions of atmospheric pollutants. The risk associated with human exposure to atmospheric PAHs is highest in the urban areas because of the density of population (Caricchia et al. 1999).

Recently, animal experiments have shown how carcinogenic activity of PAHs in vehicular exhaust extracts is associated with the fraction containing compounds composed of four to seven aromatic rings (Grimmer et al. 1983, 1984), and some of these compounds may cause tumours in humans (Boström et al. 2002).

Another study reported that PAH, phenol and selected trace element (Cd, Cu, Hg, Pb, S and Zn) determinations on detailed soil profiles are associated to plant bioindicators (including lichen: *Hypogymnia physodes*, moss: *Hylocomium splendens*, pine: *Pinus sylvestris*) from the three most representative habitats in the Holy Cross Mountains, South-Central Poland (Migaszewski et al. 2002). The results of this study indicate that content of PAHs, Cd, Cu, Hg, Pb, S and Zn in the soils and plant bioindicators examined has not changed considerably since 1998. Augusto et al. (2009) reported how spatial models based on PAHs intercepted by lichens can be used for fingerprinting of multisource atmospheric pollution in a regional area. Urban-industrial areas showed the highest atmospheric deposition of PAHs followed by urban > industrial > agricultural > forest.

Naeth and Wilkinson (2008) reported the effect of sampling direction and distance from a diamond mine on bioaccumulation in three lichen species, *Flavocetraria nivalis*, *Flavocetraria cucullata* and *Cladina arbuscula*. Thirty-three major and trace elements, sulphate (SO<sub>4</sub>), nitrate (NO<sub>3</sub>), ammonium (NH<sub>4</sub>), PAHs and phthalates were analyzed in lichen tissue and soil; result suggested that highest concentrations of Al, Cr, Cu, Fe, Ni, Ti and V in lichen were at the mine site regardless of direction. Highest concentrations for other elements were at the mine in at least two directions. Although present in lichen tissue, there

was no significant difference among sites for Hg, Mn, S and three phthalates. PAHs were below detection limits in lichen tissue.

Lichens are peculiar plants in having special sensitivity for acidic gases but accumulate several organic compounds and elements in a large amount beyond their physiological need and thus can be utilized as biological indicators of air quality. To mitigate the adverse health and environmental effects of PAHs, it is essential to identify their sources, which in turn requires the identification and quantification of PAHs present in the affected area (Yang and Chen 2004). Species of *Rinodina* shows higher efficiency to accumulate different metals. *Rinodina olivaceobrunnea*, a common muscicolous species in Schirmacher Oasis, East Antarctica, accumulated Cr (46.01 µg g<sup>-1</sup>), Zn (71.44 µg g<sup>-1</sup>), Cu (82.33 µg g<sup>-1</sup>) and Fe (14,240 µg g<sup>-1</sup>) in fairly good amount (Upreti and Pandey 2000). *Rinodina sophodes* collected near Lucknow city in North India also exhibit accumulation of Pb (10.48 µg g<sup>-1</sup>), Cr (6.52 µg g<sup>-1</sup>), Zn (127.0 µg g<sup>-1</sup>), Fe (2,428 µg g<sup>-1</sup>), Cu (16.19 µg g<sup>-1</sup>) and Hg (6.52 µg g<sup>-1</sup>) (Saxena 2004). Satya and Upreti (2009) reported that concentration of carbon and sulphur content significantly affects the physiology of *R. sophodes* in Kanpur City.

In India, most of the studies on the accumulation of heavy metals by lichens are focused on foliose lichens (Shukla and Upreti 2007, 2008) which have efficient biomass and mainly of occurrence to the way of the urban regions. Regarding PAHs, only few studies using lichens (foliose) as biomonitors of these compounds have been found in India (Shukla and Upreti 2009; Shukla et al. 2009). The majority of these studies were conducted in natural and forest ecosystems or in urban environments. The crustose lichens on the other hand are highly resistant in the urban areas with slow growth pattern and abundance occurrence. However, the information about biomonitoring of pollutants using these crustose lichens is meager. Keeping this potentiality of crustose lichens in mind, present investigation have been made to determine the PAHs accumulation efficiency of *R. sophodes* (highly resistance in the study area) to assign best bioaccumulation in the tropical

regions of India for future prospective. No such other effective way of biomonitoring is known so far in India, which address the problems accurately emphasizing the implication of present investigations to sort out the environmental hazardous, effectively using potential trait of lichens in favor to cover the risk to the people and environment.

**Material and methods**

**Description of site and material**

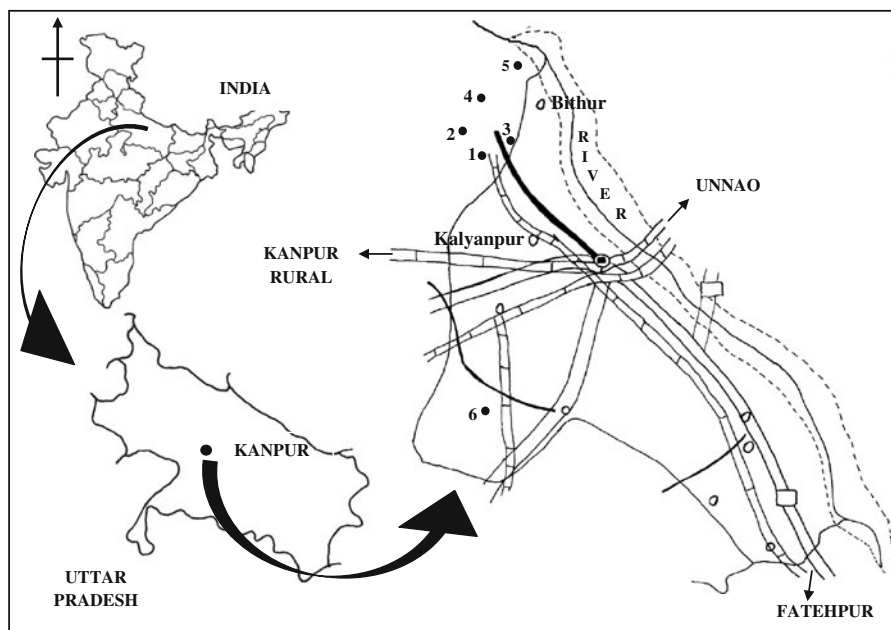
Kanpur City, with a population of about three million, is situated in north-central part of India at 88°22' E and 26°26' N in Gangatic plain. Kanpur is the largest and most populated industrial city in the state of Uttar Pradesh in India. *R. sophodes* (Ach.) Massal., an epiphytic crustose lichen found to be dominated in the area, was collected for estimation of its PAHs contents. Lichen samples were collected in December 2008 from *Mangifera indica* along with *Azadirachta indica* and *Artocarpous heterophyllus* tree trunk from up to 1.5 m height from the ground. Total six different peripheral regions of industrial and urban areas were selected for random collection of lichen samples (Fig. 1, Table 1). However, 18 samples,

triplicate from each of the six localities, were analyzed for the accumulation potential and physico-biochemical parameters. The samples were kept at low temperature and wrapped in aluminum foil until the analysis. The collected specimens were identified by their morphological, anatomical and chemical characters and voucher specimens were preserved in the lichen Herbarium of National Botanical Research Institute, Lucknow (LWG), India.

**Sampling and sample preparation**

According to US EPA (1986) air-dried lichen sample (2 g) from each site was weighed, and a thimble (Whatman No. 42) was prepared for the extraction of PAHs in Soxhlet apparatus with 100 ml of Dichloromethane (Merck, AR) for 16 hours according to the procedure (EPA 8310). The extract was passed through anhydrous sodium sulphate (Qualigen, AR) to remove the moisture and concentrated up to 2 ml under vacuum in rotary evaporator (Buchi). The extracts were solvent-exchanged to cyclohexane before the silica gel column cleanup. The slurry of 10 g activated silica gel in methylene chloride was prepared and placed into a chromatographic column (ID 10 mm). The methyl was eluted from the

**Fig. 1** Map of Kanpur city showing localities explored for lichen and its location within India, Uttar Pradesh



**Table 1** Description of the sites selected for the collection of *R. sophodes* (Ach.) Massal. at Kanpur city, India

S. no.	Site	Location of the site	Pollution sources
1	Karsauli	27 v/h (mostly light vehicles, village area with low human interference)	Moderately polluted
2	Gadewa	32 v/h (light vehicles, agriculture area along with two brick field)	Moderately polluted
3	Ramnagar	152 v/h (grant trunk road, highly traffic activity, railway track with maximum human interference)	Highly polluted
4	Devpur	3–4 v/h (agricultural area)	Moderately polluted
5	Tatiyaganj	167 v/h (grant trunk road, high traffic activity, site having motor garage with maximum human interference)	Highly polluted
6	Sujanpur	56 v/h (soap and motor parts factory along with two brick field, huge human interference)	Highly polluted

column. The column was washed with 25 ml of *n*-pentane at an elution rate of about 2 ml min<sup>-1</sup> just prior to being eluted dry; 1 ml of sample extract in cyclohexane was loaded on the column. An additional 3 ml cyclohexane was used to complete the transfer. The column was washed in 25 ml of *n*-pentane without disturbing the upper layer and the washing discarded. PAHs on the column were eluted with 25 ml of methylene chloride/*n*-pentane mixture (2:3 *v/v*) in a conical flask (US EPA 1986). The final extract was solvent exchanged to acetonitrile (Merck, AR) and making up the final volume to 2 ml in amber colour volumetric flask. The sample was filtered through 0.2 µm and stored in dark at 5°C to avoid photo degradation of PAHs.

#### PAH determination

The end analysis was performed using high performance liquid chromatography consisting of 515 pump (Waters Milford, MA, USA) and ultra violet–visible detector 2487 (Waters) and chromatogram was observed at 254 nm. Millenium<sup>R</sup> software was used for processing. Reverse Phase C-18 (Symmetry column, Waters make; 250 nm × 4.6 mm i.d. and 5 µm particle size). Acetonitrile and Mill-Q water in the ratio of 70:30 *v/v* was used as solvent system at the flow rate of 1.5 mL min<sup>-1</sup> in isocratic mode at 27°C temperature.

#### Method performance

The analysis was carried out in triplicate and the recovery percent of individual PAHs were

determined through spiked sample method at 0.2 µg g<sup>-1</sup> level. For percent recovery 2 gm of lichen samples were spiked with 0.2 µg/mL contain individual PAHs. The standard was run in five times and limit of detection and limit of quantification were calculated (Huber 2003). Extraction and cleanup processor were performed according to the method described in sampling and sample preparation. The recovery was found between 80% and 90%. The PAHs were identified by their retention time and were quantified by their respective peak areas. Standard PAH calibration mixture (EPA 610, Cat. No. 4S8743) was procured from Sigma Aldrich, USA. Detection limit of the instrument is 10–30 µg kg<sup>-1</sup> for different PAHs.

#### Statistical analysis

The experiment was done as randomized block design. Two-way analysis of variance (ANOVA) and correlation coefficient was done with all the data to confirm the variability of data and significant difference between parameters (Gomez and Gomez 1984).

#### Results and discussion

As per EPA, 16 PAHs were analyzed in the study area: total of 10 individuals were found from six sites; the rest of them were found below detection limit (Table 2). Concentrations of individual PAHs were ranged between 0.010 and

**Table 2** PAH concentration ( $\mu\text{g g}^{-1}$ ) in lichen collected from different sites of Kanpur City, India

PAHs	Site 1	Site 2	Site 3	Site 4	Site 5	Site 6	Mean
Napthalene	ND	0.014	0.019	0.010	0.319	0.015	0.063
Acenaphthylene	0.059	0.052	0.107	0.092	0.014	0.099	0.071
Fluorene and Acenaphthene	0.037	0.160	ND	0.013	0.057	0.041	0.051
Phenanthrene	0.052	0.064	0.055	0.027	0.036	0.044	0.046
Anthracene	0.036	ND	0.066	0.022	0.026	0.066	0.036
Fluoranthene	0.034	ND	0.058	ND	ND	0.044	0.023
Pyrene	0.038	ND	0.052	0.025	0.042	0.012	0.028
Benz(a)anthracene and chrysene	ND	ND	0.107	ND	ND	0.133	0.04
$\Sigma$ PAHs	$0.256 \pm 0.021$	$0.290 \pm 0.056$	$0.464 \pm 0.037$	$0.189 \pm 0.029$	$0.494 \pm 0.105$	$0.454 \pm 0.041$	

Detection limit of the instrument = 10–30  $\mu\text{g kg}^{-1}$  for individual PAHs

ND not detected

0.319  $\mu\text{g g}^{-1}$ . The limits of detection for all PAHs were ranged from 0.008 to 0.050  $\mu\text{g g}^{-1}$  while limit of quantification were ranged between 0.010 to 0.107  $\mu\text{g g}^{-1}$  (Table 2). Out of the six localities, the  $\Sigma$ PAHs concentration in *R. sophodes* was maximum at site 5 (0.494  $\mu\text{g g}^{-1}$ ) followed by site 3 (0.464  $\mu\text{g g}^{-1}$ ) and site 6 (0.454  $\mu\text{g g}^{-1}$ ), respectively. The maximum mean concentration of acenaphthylene was found with 0.071  $\mu\text{g g}^{-1}$  followed by naphthalene and fluorene + acenaphthene having 0.063 and 0.051  $\mu\text{g g}^{-1}$ , respectively (Table 2). The common PAHs recorded from the study area are acenaphthylene and phenanthrene. Shukla et al. (2009) confirms that vehicular activity to be the main source of contamination. The source for acenaphthylene emission are varied and correspond to different sorts of combustion; this compound is mainly present in the gaseous phase (Dejean et al. 2009) while phenanthrene is mainly formed from motorized traffic, especially trucks (Blasco et al. 2006). It was observed that the concentration of benz(a)anthracene and chrysene are high (0.107 and 0.133  $\mu\text{g g}^{-1}$ , respectively) at sites 3 and 6 having heavy vehicular activity by the trucks as the site 3 was situated at Highway while site 6 near a brickfield. Unburned fuels and the conversion of 2-methylnaphthalene in the fuel to naphthalene are the main sources of naphthalene in motor vehicle exhaust (Marr et al. 2005). The average concentration of benz(a)anthracene and chrysene at sites 3 and 6 represent to diesel as well as natural gas combustion activities (Khalili et al. 1995).

Sites having moist and dense habitat showed maximum concentration of  $\Sigma$ PAHs 0.494  $\mu\text{g g}^{-1}$  (site 5), while dry habitat with sparsely growing *M. indica* trees exhibited minimum concentration of  $\Sigma$ PAHs 0.189  $\mu\text{g g}^{-1}$  (site 4). The collection of the samples was performed during winter season when the coal and wood are excessively used for domestic purposes. In general, PAH concentrations tend to be about one order of magnitude higher in winter than in summer. The main reason for these variations are meteorological factors, such as increased atmospheric stability in winter, higher emissions from domestic heating systems and reduced atmospheric reactivity of PAH compounds, e.g. reduced degradation by photo-oxidation and reaction with OH radicals, in winter (Zelano et al. 2006).

Table 2 shows the concentration obtained from the PAHs identified and quantified in *R. sophodes*. The obtained concentrations can be classified according to two groups of PAHs depending on their molecular weight. PAHs of low molecular weight having two rings accumulated in the concentration ranges of 0.010 to 0.319  $\mu\text{g g}^{-1}$ ; while three rings accumulated in between 0.013 and 0.160  $\mu\text{g g}^{-1}$ . The high molecular weight PAHs having above four rings accumulated between 0.012 and 0.133  $\mu\text{g g}^{-1}$  (Table 2). *R. sophodes*, a corticolous epiphytic lichen, exhibit maximum accumulation of 2–4-ringed PAHs, similar to the studies conducted by Shukla and Upreti (2009) on the saxicolous crustose-squamulose species of the

**Table 3** Concentration of total PAH ( $\mu\text{g g}^{-1}$ ) and ratios of different individual PAH compound in the *R. sophodes*, collected from Kanpur City, India

Sites	Fluo/Fluo+Pyr	Fluo/Pyr	Phen/Anthr	Anthr/Anthr+Phen	Naph/Phen
1	0.472	0.894	1.444	0.409	ND
2	ND	ND	ND	ND	0.218
3	0.527	1.115	0.833	0.545	0.345
4	ND	ND	1.227	0.448	0.370
5	ND	ND	1.384	0.419	8.861
6	0.785	3.666	0.666	0.600	0.340

ND not detected

genus *Acarospora*, *Dimelaena* and *Aspicilia* which accumulated uniform concentration of low molecular weight two- and three-ringed PAHs. The concentration of benz(*k*)fluorethene, benz(*b*)fluorethene, benz(*a*)pyrene, and dibenz(*a,h*)anthracene, indeno(1,2,3,*c,d*)pyrene, benz(*g,h,i*)perylene with high molecular weight (5–6-ringed) were not detected with the instrument used in this study. Guidotti et al. (2003) reported that PAHs with high molecular weight are exclusively adsorbed on suspended particulate matter in the atmosphere.

Some of the PAH species are known to be animal and/or human carcinogens, so evaluating the healthy risk that is associated with the exposure to both particulate PAHs and gaseous PAHs is important. Gaseous PAHs usually contain more fractions of less carcinogenicity and lower molecular weights, while particulate PAHs contain more fractions of higher carcinogenicity and higher molecular weights. In principle, PAHs with high molecular weights are often more carcinogenic than those with low molecular

weights (Park et al. 2002; Tsai et al. 2002). In the present study, only benz(*a*)anthracene and chrysenes identified as carcinogenic with high molecular weight PAHs. The lower molecular weight PAHs (two- to three-ringed group of PAHs such as naphthalenes, fluorenes, phenanthrenes and anthracenes) have significant acute toxicity to aquatic organisms, whereas the high molecular weight PAHs (four to seven rings, from chrysenes to dibenz(*a,h*)anthracene) do not (Park et al. 2002; Tsai et al. 2002). The ratios between some specific PAHs are good indicators for the sources of the environmental contamination (Park et al. 2002; Tsai et al. 2002). In the present study, Phe/Ant ratios are <10, while the Flu/Pyr ratios are >1.0, strongly indicates that the PAHs originated by pyrogenic and combustion except site 1 having 0.89  $\mu\text{g g}^{-1}$  (Tables 3 and 4). Similarly, Blasco et al. (2006) also reported that the ratio of Phe/Ant are lower than 10, while the Flu/Pyr ratios are higher than 1.0, strongly indicating that the PAHs had pyrogenic origins. Phe/Ant ratios <10, Flu/Pyr ratios >1 and BaA/BaA and Chr

**Table 4** Value of PAHs and different sources of PAHs

Ratio	Values	Sources	Reference
Fluo/Pyr	>1	Combustion	Baumard et al. (1998)
	<1	Petroleum	
Phen/Anthr	<10	Combustion	Baumard et al. (1998)
	>10	Petroleum	
Naph/Phen	$\ll$ 1	Petroleum	Dahle et al. (2003)
Anthr/(Anthr+Phen)	>0.1	Combustion	Yunker et al. (2002)
	<0.1	Petroleum	
Fluo/(Fluo+Pyr)	>0.5	Grass, coal and wood combustion	Yunker et al. (2002)
	0.4–0.5	Gasoline, diesel and crude oil combustion car and diesel trucks	
	<0.4	Petroleum	
B(a)P/B(ghi)P	>0.6	Non traffic sources	Pandey et al. (1999)
	<0.6	Traffic sources	



ratios >0.35 suggest a pyrogenic origin (Baumard et al. 1998). The ratios of Anthr/Anthr + Phen and Naph/Phen were found >0.10 and <1, respectively, except site 5 having 8.861 g g<sup>-1</sup>. Budzinski et al. (1997) suggested that, Anthr/Anthr + Phe ratios <0.10 are usually attributed to petroleum sources, while ratios >0.10 indicate the combustion sources. The ratios of Naph/Phen ≪1 indicate to petroleum emission (Dahle et al. 2003). Similar to Yunker et al. (2002) both the sites 3 and 6 showed Fluo/Fluo + Pyr ratios >0.5, characteristic of coal wood combustion as source of PAHs. The leaves are the main sinks of airborne PAH compounds in plants, while uptake of PAHs from soils via the root system appears to be negligible. The most important anatomical feature of plants that affects the rate of interception and accumulation of airborne PAHs is total leaf area. Volatile PAHs enter plants primarily through gaseous diffusion via open stomata, although absorption by the waxy leaf surface accounts for a portion of total PAHs in tissues. The waxy surface of leaves intercepts both particulate and vapour phase contaminants. Absence of waxy cuticle in lichens and sometimes peculiar granular thallus surface of *R. sophodes*, a poliotolarent lichen, serves as the best accumulator for ambient organic pollutant like PAHs.

The two-way ANOVA also revealed that PAHs showed a significant interaction between all sampling points (significant at P 1%, Table 5). Correlation of various PAHs showed mostly negative significant correlation (P 1%) between all organic pollutants while fluoranthene is positively correlated with benz(a)anthracene and chrysene ( $r = 0.812132$ , Table 6).

The present study affirms that the major sources of PAHs were combustion of organic materials, traffic and vehicular exhaust (diesel and gasoline engine). In the past, control strategies for motor vehicles emphasized improvements in engine and emission control technologies. Previous studies were providing good tools for the prevention of PAHs from the ambient environment. In the past, control strategies for motor vehicles emphasized improvements in engine and emission control technologies. Results suggested that fuels have been reformulated to reduce vehicle emissions. For example, oxygenated compounds, such

**Table 5** PAHs concentration (µg g<sup>-1</sup>) at different sites of Kanpur city, India

Sites	PAHs concentrations µg/g FW											
	Naph	Acy	Fl and Ace	Phen	Anthr	Fluo	Pyr	B(a)A and Chry	B(k)F	B(b)F	B(a)P	D(a,h)A
1	0	0.059 ± 0.040	0.037 ± 0.064	0.052 ± 0.040	0.036 ± 0.003	0.034 ± 0.003	0.038 ± 0.034	0	0	0	0	0
2	0.014 ± 0.005	0.052 ± 0.022	0.160 ± 0.046	0.064 ± 0.018	0	0	0	0	0	0	0	0
3	0.019 ± 0.020	0.107 ± 0.055	0	0.055 ± 0.048	0.066 ± 0.057	0.058 ± 0.050	0.052 ± 0.024	0.107 ± 0.093	0	0	0	0
4	0.010 ± 0.000	0.092 ± 0.002	0.013 ± 0.000	0.027 ± 0.000	0.022 ± 0.000	0	0.025 ± 0.022	0	0	0	0	0
5	0.319 ± 0.553	0.014 ± 0.025	0.057 ± 0.010	0.036 ± 0.016	0.026 ± 0.044	0	0.042 ± 0.074	0	0	0	0	0
6	0.015 ± 0.017	0.099 ± 0.077	0.041 ± 0.001	0.044 ± 0.040	0.066 ± 0.000	0.044 ± 0.001	0.012 ± 0.001	0.133 ± 0.028	0	0	0	0
ANOVA	**	**	**	**	**	**	**	**	ns	ns	ns	ns

Mean concentrations and SD of three individual replicates along with two-way ANOVA

ns not significant, ± standard deviation

\*\*\* p = 1%

**Table 6** Values of correlation coefficient between the various polycyclic aromatic hydrocarbons in *R. sophodes*

PAH	Naph	Acy	Fl and Ace	Phen	Anthr	Fluo	Pyr	B(a)A and Chry	B(k)F	B(b)F	B(a)P	D(a, h)A	IP and B(g, h, i)P
Naph	1	-0.537	0.075	-0.029	0.131	-0.452	-0.338	-0.310	0	0	0	0	0.687
Acy		1	-0.378	0.021	-0.605	0.213	-0.032	-0.044	0	0	0	0	-0.561
Fl and Ace			1	0.453	-0.256	-0.531	-0.247	-0.386	0	0	0	0	0.118
Phen				1	0.014	0.015	-0.759	0.381	0	0	0	0	0.516
Anthr					1	0.177	-0.096	0.584	0	0	0	0	0.385
Fluo						1	0.430	0.812**	0	0	0	0	0.079
Pyr							1	-0.036	0	0	0	0	-0.476
B(a)A and Chry								1	0	0	0	0	0.382
B(k)F									1	0	0	0	0
B(b)F										1	0	0	0
B(a)P											1	0	0
D(a, h)A												1	0
IP and B(g, h, i)P													1

\*\*  $P = 1\%$  correlation

nd not detected (detection limit of the instrument: 10–30  $\mu\text{g kg}^{-1}$ )

*Naph* naphthalene, *Acy* acenaphthylene, *Fl and Ace* fluorene and acenaphthene, *Phen* phenanthrene, *Anthr* anthracene, *Fluo* fluoranthene, *Pyr* pyrene, *B(a)A and Chry* benzo(a)anthracene and chrysene, *B(k)F* benzo(k)fluoranthene, *B(b)F* benzo(b)fluoranthene, *B(a)P* benzo(a)pyrene, *D(a, h)A* dibenzo(a, h)anthracene, *IP and B(g, h, i)P* indeno(1,2,3, c, d)pyrene and benzo(g, h, i)perylene

as methyl *tert*-butyl ether, have been added to gasoline (Kirchstetter et al. 1996) and the sulphur and aromatic contents of diesel fuel have been reduced (Westerholm and Li 1994). It is possible that some types of fuel reformulation might help to reduce PAH emissions from motor vehicles (Marr et al. 1999). Polycyclic aromatic hydrocarbons are a particularly recalcitrant group of contaminants and are known to be highly persistent in the environment (Glick 2003). Another study have been reported that plants can be removed, destroy or sequester hazardous substances from the environment by phytoremediation method (Glick 2003; Raskin and Smith 1997).

## Conclusions

*R. sophodes* exhibits its luxuriant growth in tropical areas of India where few other crustose and foliose lichens are able to grow. The species has ability to colonize both on thin smooth to thick, rough barked trees of *M. indica*, *A. indica* and *A. heterophyllus*. These trees are mostly planted in the proximity of urban areas as avenue trees.

In India, biomonitoring in most of the tropical areas *Phaeophyscia hispidula* and *Pyxine cocoes* are most frequently used lichens as active biomonitoring studies. However, in carrying out biomonitoring studies on lichens growing in natural environment, *R. sophodes* serves as the best plant material as it not only efficiently accumulates both metals and PAHs but also able to grow more frequently in polluted areas than its foliose brethrens.

**Acknowledgements** The authors are thankful to the Director, National Botanical Research Institute, Lucknow, India for providing necessary laboratory facilities to work. Dr. Satya is thankful to Council of Scientific and Industrial research, New Delhi, India for providing Senior Research Fellowship.

## References

- Augusto, S. C., Máguas, J., Matos, M. J., Pereira, A., Soares, C., & Branquinho. (2009). Spatial modeling of PAHs in lichens for fingerprinting of multisource atmospheric pollution. *Science of the Total Environment*, 43, 7762–7769.
- Baumard, P., Budzinski, H., & Garrigue, P. (1998). Polycyclic aromatic hydrocarbons in sediments and



- mussels of the western Mediterranean sea. *Environmental Toxicology Chemistry*, 17, 765–776.
- Blasco, M., Domeno, C., & Nerín, C. (2006). Use of lichens as pollution biomonitors in remote areas: Comparison of PAHs extracted from lichens and atmospheric particles sampled in and around the somport tunnel (Pyrenees). *Science of the Total Environment*, 40, 6384–6391.
- Boström, C. E., Gerde, P., Hanberg, A., Jernström, B., Johansson, C., Kyrklund, T., et al. (2002). Cancer risk assessment, indicators, and guidelines for polycyclic aromatic hydrocarbons in the ambient air. *Environmental Health Perspectives*, 110, 451–488.
- Budzinski, H., Jones, I., Bellocq, J., Pierad, C., & Garrigues, P. (1997). Evaluation of sediment contamination by polycyclic aromatic hydrocarbons in the Gironde estuary. *Marine Chemistry*, 58, 85–97.
- Caricchia, A. M., Chiavarini, S., & Pezza, M. (1999). Polycyclic aromatic hydrocarbons in the urban atmospheric particulate matter in the city of Naples (Italy). *Atmospheric Environment*, 33, 3731–3738.
- Dahle, S., Savinov, V. M., Matishov, G. G., Evenset, A., & Naes, K. (2003). Polycyclic aromatic hydrocarbons (PAHs) in bottom sediments of the Kara Sea shelf, Gulf of Ob and Yenisei Bay. *Science of the Total Environment*, 306, 57–71.
- Dejean, S., Raynaud, C., Meybeck, M., Della Massa, J. P., & Simon, V. (2009). Polycyclic aromatic hydrocarbons (PAHs) in atmospheric urban area: monitoring on various types of sites. *Environmental Monitoring and Assessment*, 148, 27–37.
- Glick, B. R. (2003). Phytoremediation: Synergistic use of plants and bacteria to clean up the environment. *Biotechnology Advance*, 21, 383–393.
- Gomez, K. A., & Gomez, A. A. (1984). *Statistical procedure for agricultural research*. New York: Wiley.
- Grimmer, G., Brune, H., Deutsch-Wenzel, R. P., Dettbarn, G., & Misfeld, J. (1984). Lichens as polycyclic aromatic hydrocarbon bioaccumulators used in atmospheric pollution studies. *Journal of National Cancer Institute*, 72, 185–90.
- Grimmer, G., Brune, H., Deutch-Wenzel, R. P., Naujack, K.-W., Misfeld, J., & Timm, J. (1983). On the contribution of polycyclic aromatic hydrocarbons to the carcinogenic impact of automobile exhaust condensate evaluated by local application onto mouse skin. *Cancer Letter*, 21, 105–113.
- Guidotti, M., Stella, D., Owczarek, M., DeMarco, A., & De Simone, C. (2003). Lichens as polycyclic aromatic hydrocarbon bioaccumulators used in atmospheric pollution studies. *Journal of Chromatography*, 985, 185–90.
- Huber, W. (2003). Basic calculations about the limit of detection and its optimal determination. *Accred Quality Assurance*, 8, 213–217.
- Khalili, N., Scheff, P., & Holsen, T. (1995). PAH source fingerprints for coke ovens, diesel and, gasoline engines, highway tunnels, and wood combustion emissions. *Atmospheric Environment*, 29, 533–542.
- Kirchstetter, T. W., Singer, B. C., Harley, R. A., Kendall, G. R., & Chan, W. (1996). Impact of oxygenated gasoline use on California light-duty vehicle emissions. *Environmental Science Technology*, 30, 661–670.
- Ma, L. L., Chu, S. G., Wang, X. T., Cheng, H. X., & Lin, X. Xu. (2005). Polycyclic aromatic hydrocarbons in the surface soils from outskirts of Beijing, China. *Chemosphere*, 58, 1355–1363.
- Marr, L. C., Dzepina, K., Jimenez, J. L., Reisen, F., Bethel, H. L., Arey, J., et al. (2005). Sources and transformations of particle-bound polycyclic aromatic hydrocarbons in Mexico City. *Atmospheric Chemistry Physics Discussion*, 5, 12741–12773.
- Marr, L. C., Kirchstetter, T. W., Harley, R. A., Miguel, A. H., Hering, S. V., & Hammond, S. K. (1999). Characterization of polycyclic aromatic hydrocarbons in motor vehicle fuels and exhaust emissions. *Environmental Science Technology*, 33, 3091–3099.
- Migaszewski, Z. M., Galuszka, A., & Paslawski, P. (2002). Polynuclear aromatic hydrocarbons, phenols, and trace metals in selected soil profiles and plant bioindicators in the Holy Cross Mountains, South-Central Poland. *Environmental International*, 28, 303–313.
- Naeth, M. A., & Wilkinson, S. R. (2008). Lichens as biomonitors of air quality around a diamond mine, Northwest Territories, Canada. *Journal of Environmental Quality*, 37, 1675–1684.
- Pandey, P. K., Patel, K. S., & Lenicek, J. (1999). Polycyclic aromatic hydrocarbons: Need for assessment of health risks in India? Study of an urban-industrial location in India. *Environmental Monitoring and Assessment*, 59, 287–319.
- Park, S. S., Kim, Y. J., & Kang, C. H. (2002). Atmospheric polycyclic aromatic hydrocarbons in Seoul, Korea. *Atmospheric Environment*, 36, 2917–2924.
- Raskin, R. D., & Smith, D. E. (1997). Salt, Phytoremediation of metals: Using plants to remove pollutants from the environment. *Current Opinion Biotechnology*, 8, 221–226.
- Satya, & Upreti, D. K. (2009). Correlation among Carbon, Nitrogen, Sulphur and physiological parameters of *Rinodina sophodes* in Kanpur City, India. *Journal of Hazardous Materials*, 163, 1088–1092.
- Saxena, S. (2004). *Lichen flora of Lucknow district with reference to air pollution studies in the area* (pp. 1–131). Ph.D. thesis, University of Lucknow (submitted to).
- Shukla, V., & Upreti, D. K. (2007). Physiological response of the lichen *Phaeophyscia hispidula* (Ach.) Essl., to the urban environment of Pauri and Srinagar (Garhwal), Himalayas, India. *Environmental Pollution*, 150, 295–299. doi:10.1016/j.envpol.2007.02.010.
- Shukla, V., & Upreti, D. K. (2008). Effect of metallic pollutants on the physiology of lichen, *Pyxine subcinerea* Stirton in Garhwal Himalayas. *Environmental Monitoring and Assessment*, 141, 237–243.
- Shukla, V., & Upreti, D. K. (2009). Polycyclic aromatic hydrocarbon (PAH) accumulation in lichen, *Phaeophyscia hispidula* of Dehradun City, Garhwal Himalayas. *Environmental Monitoring and Assessment*, 149, 1–7.
- Shukla, V., Upreti, D. K., Patel, D. K., & Tripathi, R. (2009). Accumulation of polycyclic aromatic

- hydrocarbons in some lichens of Garhwal Himalayas, India. *International Journal of Environmental Waste Management*, 5, 104–113.
- Tsai, P. J., Shieh, H. Y., Lee, W. J., & Lai, S. O. (2002). Characterization of PAHs in the atmosphere of carbon black manufacturing work places. *Journal of Hazardous Materials*, 91, 25–42.
- Upreti, D. K., & Pandey, V. (2000). Determination of heavy metals in lichens growing on different ecological habitats in Schirmacher Oasis, East Antarctica. *Spectroscopy Letter*, 33, 435–444.
- US EPA (1986). *Test methods for evaluating solid waste* (Vol 1B). Laboratory Manual Physical/Chemical Methods. Washington, DC.
- Westerholm, R., & Li, H. (1994). A multivariate statistical analysis of fuel-related polycyclic aromatic hydrocarbon emissions from heavy-duty diesel vehicles. *Environmental Science and Technology*, 28, 965–972.
- Yang, H. H., & Chen, C. M. (2004). Emission inventory and sources of polycyclic aromatic hydrocarbons in the atmosphere at a suburban area in Taiwan. *Chemosphere*, 56, 879–887.
- Yunker, M. B., MacDonald, R. W., Vingarzan, R., Mitchell, H. R., Goyette, D., & Sylvestre, S. (2002). PAHs in the Fraser River basin: A critical appraisal of PAH ratios as indicators of PAH source and composition. *Organic Geochemistry*, 33, 489–515.
- Zelano, V., Torazzo, A., Berto, S., Ginepro, M., Prenesti, E., & Ferrari, A. (2006). Biomonitoring of traffic originated PAHs in the air. *International Journal of Environmental Analytical Chemistry*, 86, 527–540.