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Bryomonitoring of Atmospheric Elements in *Rhodobryum giganteum* (Schwaegr.) Par., Growing in Uttarakhand Region of Indian Himalayas

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ABSTRACT

Mosses are useful biological indicators of environmental contamination for a variety of metals originated from natural and artificial sources. The research aims on recent changes in air quality and estimate the atmospheric metal deposition and its seasonal/annual trend by using passive biomonitoring technique. The level of Cu, Cd, Zn and Pb was estimated in samples of the *Rhodobryum giganteum* (Schwaegr.) Par., from Mukteshwar, Nainital, Almora and Pithoragarh of Kumaon and Chamba, Mussoorie, Dhanaulti and Kampty Fall of Garhwal region of Uttarakhand, India.

The study considers the level and possible sources of atmospheric pollutants in catchment sites and their effect on naturally growing moss. The moss samples were collected from each season consist of fourth month i.e., winter, summer and monsoon from equi-distances during years 2008 to 2012. The high metal content was found at Nainital (Kumaon) and Mussoorie (Garhwal) due to heavy traffic activity.

The results showed that the collected moss species was capable of successfully delineating the expected differences in atmospheric metal deposition within the study area. The metal deposition loads were recorded in the order of Zn > Pb > Cu > Cd during the five years study period. A general increasing metal deposition trend was also observed, i.e., annual average (over seasons) metal loads in 2012 increased when compared with their loads in 2008. Development is almost always accompanied with negative changes in air quality and adverse impacts of air pollution on human health, agricultural production and natural ecosystems that need to monitor and do mitigate, therefore, such investigations is highly demanded. Present information will be very helpful for environmentalist/policy makers in making effective strategies to mitigate the environment related problems.

Keywords: Biomonitoring; Metal deposition; Moss; *Rhodobryum giganteum*; Seasonal and Annual trend.

INTRODUCTION

Bryophytes are used as active and passive bioindicators. Number of previous studies depicts the relationships between bryophyte communities and the environmental factors driving their ecology (Muotka and Virtanen, 1995; Virtanen *et al.*, 2001; Heino and Virtanen, 2006; Scarlett and O'Hare, 2006; Rodriguez *et al.*, 2014). There is a vast range of bryophytes (mosses) engaged in biomonitoring practices and are being used as one of the efficient tool for monitoring the climate changes (Gignac, 2001; Varela *et al.*, 2010). They are good air-quality monitors mainly due to the fact that their only water sources are atmospheric moisture and precipitations. Further, tolerance to desiccation allows them to survive long without water supply, and can continue photosynthesis under such conditions and, thus, stay at the study site longer, although exposed to all possible pollutants (Proctor and Tuba, 2002). Beyond these advantages, mosses are easy to work with due to their small size and low demands. Their cultivation, storage and analyses are also not expensive (Tingey, 1989). In recent past, there has been a surging curiosity regarding bryophyte responses to environment, due to their characteristic thallus composition as they are composed of tissue sheets. Therefore, they are in close vicinity with the environment and respond more rapidly to environmental changes than vascular plants, because they get water, nutrients and elements mainly from the atmosphere (Alam, 2013). In addition, bryophytes show a high degree of sensitivity too and are strongly resistant to numerous toxic substances, heavy metals and a wide range of persistent organic pollutants. Due to their unique uptake mechanisms they are regarded as excellent accumulation indicators in many studies (Zechmeister *et al.*, 2003). They are very handy tool in biological monitoring where the use of organisms provides information on certain aspect of biosphere like air-

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borne metal content. Bryomonitoring is inexpensive method in quantifying the possible emanation of metals as mosses are able to increase elements in extremely high concentration and provide easy detection of elements present even in very low concentration in the environment (Alam, 2013).

The focus of present study was the assessment of the metal pollution level in the Kumaon and Garhwal region of Uttarakhand by using *Rhodobryum giganteum* as a passive biomonitor, i.e., sampling design, sample collection and sample preparation/processing. Results of present moss survey allow examination of both seasonal and annual trends of metal concentration of past five years (2008–2012) with the help of moss *R. giganteum* and identification of areas of high metal deposition. Mosses are being used for the indication of heavy metal deposition in the air, with the help of spectroscopic analyses depiction of even absolute concentrations of their substances is possible (Figueira *et al.*, 2002; Srivastava *et al.*, 2014). The data obtained this way are suitable for the long-term survey of heavy metal deposition at the site under investigation (Grodzińska and Szarek-Łukaszevska, 2001; Lucaciu *et al.*, 2004; Chakrabortty and Paratakar, 2006) and such information will be very helpful for environmentalist/policy makers in making effective strategies to mitigate the environment related problems. In India, these different features of bryophytes in biomonitoring of atmospheric pollution are not utilized yet at large scale. Therefore, we propose a protocol that would enable the routine use of this tool for environmental monitoring. Many European country took part in the study concerning spatial differences in metal accumulation of bryophytes across boundaries (Schröder *et al.*, 2007) and there was great effort put in the studies relating to the biomonitoring of trace metals in moss material with emphases on recent or past environmental loads (Suchara and Sucharová, 2004; Saxena *et al.*, 2008a; Suchara and Sucharová, 2008).

METHODS

Study Area

The Uttarakhand comprises of a contiguous areas of Kumaon and Garhwal hills (Fig. 1). Kumaon experiences a sub tropical climate, and the weather is pleasant throughout the year. Summers, winters and monsoon constitute the basic seasons. Summers (March–June) are mild and the maximum and minimum temperature ranges from 46°C to 19°C respectively. Winters extend through the months from November to February and these months will experience a maximum of around 25°C and a minimum temperature of around 4°C. The monsoon experiences moderate rainfall from July to October.

The Garhwal climate is cool from November to February, with monsoon showers between July and October. Conditions are moderate from March to June and snowfall is seldom observed in January. During summer, maximum and minimum temperature hovers around 25.6°C–31.7°C and 7.2°C–12.8°C, respectively. In winter, temperatures range from 7.2°C–2.2°C and 1.0°C–4.4°C as maximum and minimum respectively.

Designing of Samples for Mapping

Samples of *Rhodobryum giganteum* (Schwaegr.) Par., were collected during three different seasons of the period of 2008 to 20012 in accordance with the Indian weather conditions (summer, winter and monsoon, at equi-distances of 0.5, 1 and 3 km on 8 stations (direction wise) within the study area. In contrast, biomonitoring in Europe may be carried out during four seasons' autumn, winter, summer and spring (Fernández *et al.*, 2007). Samples were also taken from the interior of the Mukteshwar (Kumaon) and Chamba (Garhwal) forest cover (considered as a control site for base line concentration). Sampling of healthy moss and its handling were carried out using plastic gloves and bags. After the end of the exposure period of one season (nearly after 122 days or 4 months), these moss sample was taken from each site for the metal analysis. The same process was repeated for every season for five consecutive years.

Identification and Characteristics

Moss *Rhodobryum giganteum* (Schwaegr.) Par. was collected from different sites of Kumaon (Mukteshwar, Nainital, Almora and Pithoragarh) Garhwal (Chamba, Mussoorie, Dhanolti and Kampty Fall) hills and was brought to laboratory in plastic bags. The specimens were duly identified on the basis of morphological examination. Plants are very large and robust, measured about 24–41 mm in loose tufts, growing on humus soil and also on logs in forest shade, bright yellow-green, brown below, with creeping subterranean stolons (Figure is positioned as supplementary material). Stems erect, usually branched by sub apical innovations, about 60 mm high with small, scaly, apprised leaves spreading up to 12.5–13.8 mm (Gangulee, 1969; Chopra, 1975; Smith, 1978; Saxena *et al.*, 2008b).

Sample Treatment and Chemical Analysis

Harvested moss samples were kept in plastic bags and were brought to the laboratory. Unwashed samples were carefully freed from dead material and attached litter. A jet of air was used to remove soil particles trapped in the moss. The apexes of green moss shoots (1–2 cm) were dried for 48 h at 40°C in air-oven, and the samples were homogenized. For the analysis, 0.5 g of homogenized moss tissue was digested 4:1 v/v using HNO_3 and $HClO_4$ at 120°C. Completion of digestion was conformed when liquid was colourless. The digested samples were filtered and the clear solution was diluted to 50 mL with bi distilled water and analyzed for Cd, Cu, Pb, and Zn by Atomic Absorption Spectrophotometer. Plant metal concentrations were expressed on a total dry weight basis. Suitable blank (contain HNO_3, $HClO_4$ and bi distilled water) were used to check for possible contamination during extraction.

Statistical Analysis and Distribution Maps

The statistical evaluation of complete data was carried out and all analyses were performed based on triplicate moss transplants of five consecutive years. Data was subjected to one-way analysis of variance (ANOVA) with differences in the metal concentration in different seasons (JMP 5.0, SAS Institute, Cary, NC, USA) to analyze the effect of each

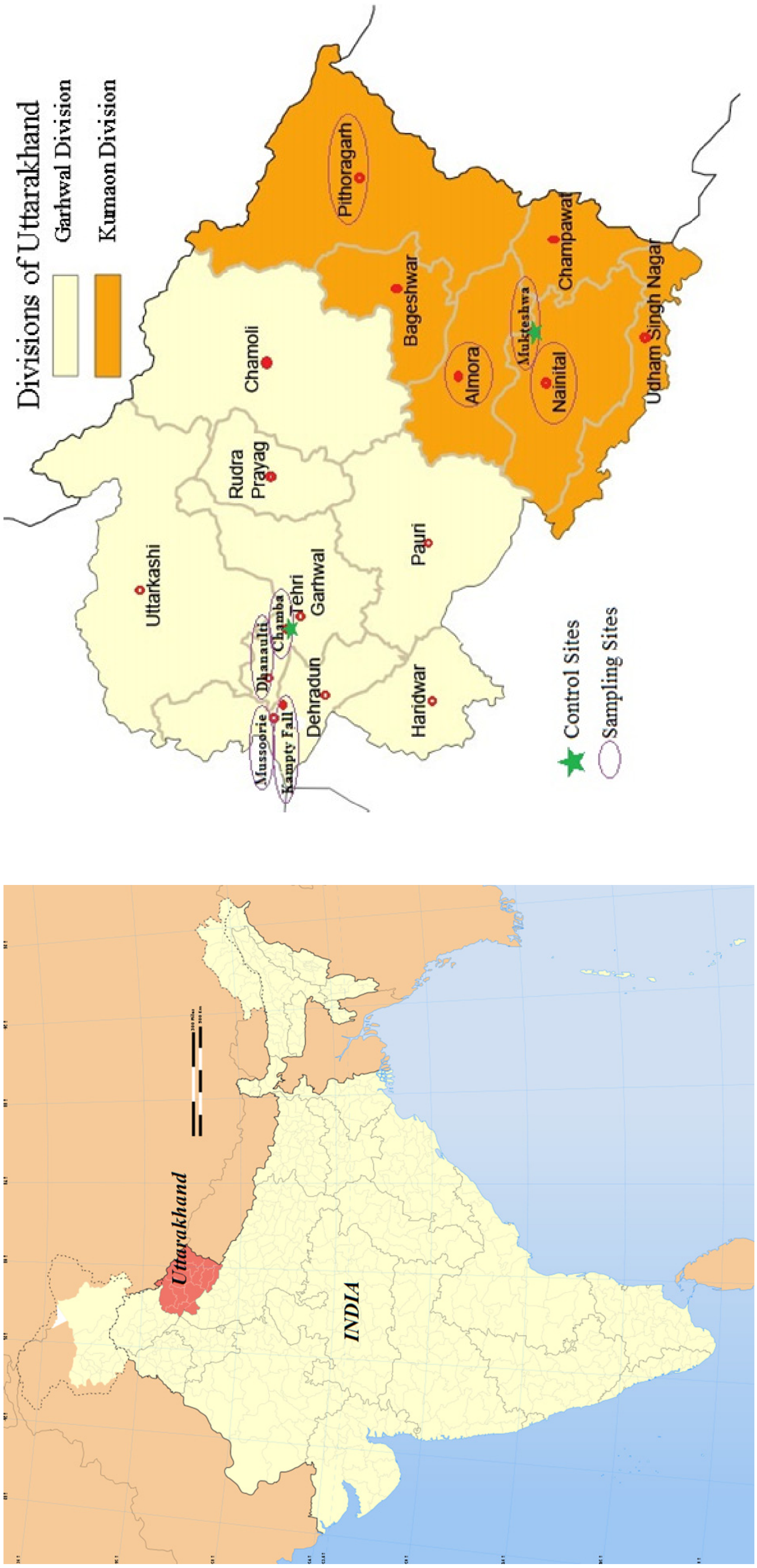

Fig. 1. Location map of sampling sites of Uttarakhand.

treatment separately. The treatment means (includes average of all directions during each season) were separated using Tukey HSD at 0.05% probability level.

RESULTS AND DISCUSSION

The metal concentration data of each season were measured in mg g^{-1} DW and is summarized in Tables 1 to 4. In addition, Figs. 2 to 9 show the results as the annual average of metal loads of each element belongs to five consecutive years. The average Zn, Pb, Cu and Cd concentrations were significantly higher than those from the baseline (control site). The sites of undisturbed zone have been considered as control as the conditions in this region is almost ideal for the growth of *Rhodobryum giganteum* because of least population and least atmospheric pollution as compared to the sites in peripheral zone. Seasonal accumulation of metals (Zn, Pb, Cu and Cd) in *R. giganteum* was in the order of summer > winter > monsoon in each year (Tables 1–4). Metal load increased significantly from 2008 to 2012 in all undertaken sites of Kumaon and Garhwal region of Uttarakhand. Nevertheless, the reason for highly significant seasonal trend could be due to the peak consumption of gasoline during summer due to many fold increase in tourist activity (Gerdol *et al.*, 2000; Saxena *et al.*, 2013; Srivastava *et al.*, 2014). During monsoon, tourist activity decreases as well as pollutants are leached out by rains. Moreover, one cannot rule out the increase in growth (biomass) more rapidly during monsoon and thus, reduce the metal percentage in moss as compared to biomass.

Zn was measured high in moss near rural as well as in proximity to road along the highways. Its source can be explained as tear wear of vehicles and its engine, with very little from exhaust emission. Concentration of zinc was significantly higher in moss with that of baseline (Table 1). Zn values were higher at Nainital (Kumaon) and Mussoorie (Garhwal) during all the five consecutive years (Figs. 2 and 6). These areas were found influenced by concentration of Zn, could be that inhabitants of this area are engaged with agricultural practices and use Zn to promote growth of agricultural crops. It is also used in fungicides and pesticides; therefore, present finding is an agreement with the work of Otvos *et al.* (2003). Zn was also concentrated in areas of high traffic density and is a major constituent of roads themselves as asphalt pavement contains cobalt, nickel and zinc (Sardans and Penuelas, 2005; Saxena *et al.*, 2013). Movement of light and heavy trucks used for transportation may explain for the high zinc accumulation by the moss samples, In addition, it can be substantial from diesel exhaust (Gerdol *et al.*, 2000; Saxena *et al.*, 2008c, 2010).

It has been demonstrated from present study that Pb concentration in the vicinity of town are inversely correlated with the distance from the road. Catchments sites shows significantly higher concentration than those measured at the baseline (Table 2) and values were found maximum at Nainital (Fig. 3) and at Mussoorie (Fig. 7). This suggests that there is a significantly increase in the level of Pb is due to vehicular emissions. Present study is in agreement with finding made by Westerlund (2001); Adachi and Tainosho (2004); Vidovic *et al.* (2005). This study shows that Pb is still very widespread and values were high in spite of the introduction of lead free petrol. The same can be the fact that, the Pb values were decreased in per litre petrol but an increased volume of traffic upto 40 times results an increase in consumption of petrol, thus magnifying the Pb value along the road. Vehicle traffic seems to be main source of atmospheric Pb as reported in other places (Loppi and Bonini, 2000). Pb does not biodegrade and it remains in dusts, soils and in sediments.

Results for Cu were influenced by the extreme concentration at the same sites as that of Zn and Pb at Nainital and Mussoorie (Table 3), during all the five consecutive years (Figs. 4 and 8). These transplants were harvested in proximity of road where vehicular traffic was very high; therefore, motor driven transport are one of the suspected causes of Cu contamination (Srivastava *et al.*, 2014). High percentage of copper along the road could be derived from wear and tear in engines (Pearson *et al.*, 2000; Poikolainen *et al.*, 2004), and at residential areas through scrap from domestic, laundry waste and from kerosene oil (Loppi and Bonini, 2000). Copper is also an integral part of fungicides, agriculture practices, open burning of solid wastes (Gerdol *et al.*, 2000; Otvos *et al.*, 2003) and, therefore, its high value in agricultural areas cannot be ruled out. Use of $CuSO_4$ mixed kerosene could also be another source of Cu in rural areas as it is available on subsidized rate by government of India to the citizen comes under below poverty line (BPL). Present result is in agreement with the findings of Lopez *et al.* (1997), who also reported a similar rise in Cu level in inhabited areas.

Cd level was measured maximum in moss collected from Nainital and Mussoorie (Table 4). Cadmium metal is very easily leached out from surface and was found low in comparison to other metals (Figs. 5 and 9). It shows the biggest dissimilarity in distribution pattern of other metals, which suggests the probability that these originated from independent sources as described by earlier workers in their finding too (Scharova and Suchara, 1998; Grodzińska and Szarek-Łukaszevska, 2001). Use of coloured polythene bags, domestic wastes, discarded plastic and utensils perhaps could also be responsible for the Cd contamination in proximity to the towns. Other source of Cd in urban sites could not be ruled out from paints and enamels containing cadmium compounds and from manufacturing of coloured plastic bags. Perhaps Cd is also present in the petrol as mining impurities. Service shops related with metals are the other factors for this increase (Alam, 2013).

Excellent inter-elemental correlation was observed between almost all elements. It indicates that the analysed elements are mostly from the common sources or identical behaviour during long range atmospheric transport during undertaken period of time (Tables 5 and 6). However, exceptions are the pairs Cd with Zn, Pb and Cu, which probably are due to high variation in metal concentrations and different emission sources. Most of these elemental correlations represent pollution one or the other way.

CONCLUSION

Over all, the results suggest that the potential source of

Table 1. Average data of Zinc (mg g^{-1} DW) during five consecutive years (2008–2012) in *Rhodobryum giganteum* at different sites in Kumaon and Garhwal region of Uttarakhand.

Sites	SUMMER					MONSOON					WINTER				
	2008	2009	2010	2011	2012	2008	2009	2010	2011	2012	2008	2009	2010	2011	2012
KUMAON															
Baseline	0.094 ± 0.004^{d}	0.141 ± 0.003^{c}	0.218 ± 0.006^{b}	0.232 ± 0.004^{ab}	0.257 ± 0.029^{a}	0.008 ± 0.001^{d}	0.016 ± 0.002^{c}	0.022 ± 0.001^{b}	0.025 ± 0.002^{b}	0.029 ± 0.001^{a}	0.024 ± 0.002^{d}	0.034 ± 0.003^{c}	0.034 ± 0.002^{c}	0.045 ± 0.005^{b}	0.103 ± 0.002^{a}
Almora	0.546 ± 0.059^{d}	0.881 ± 0.077^{c}	1.061 ± 0.168^{b}	1.202 ± 0.213^{ab}	1.346 ± 0.143^{a}	0.459 ± 0.072^{d}	0.683 ± 0.088^{c}	0.822 ± 0.08b	0.913 ± 0.091^{b}	1.213 ± 0.161^{a}	0.534 ± 0.066^{d}	0.816 ± 0.094^{c}	1.049 ± 0.121^{b}	1.199 ± 0.130^{a}	1.316 ± 0.098^{a}
Nainital	0.967 ± 0.131^{d}	1.354 ± 0.236^{c}	1.591 ± 0.318^{b}	1.914 ± 0.359^{a}	2.056 ± 0.317^{a}	0.679 ± 0125^{d}	1.021 ± 0.281^{c}	1.178 ± 0.275^{bc}	1.378 ± 0.281^{b}	1.635 ± 0.345^{a}	0.788 ± 0.094^{c}	1.132 ± 0.210^{bc}	1.318 ± 0.207^{bc}	1.569 ± 0.361^{ab}	1.941 ± 0.706^{a}
Pithoragarh	0.869 ± 0.324^{d}	1.227 ± 0.298^{c}	1.527 ± 0.437^{bc}	1.693 ± 0.536^{ab}	1.859 ± 0.526^{a}	0.622 ± 0.207^{d}	0.903 ± 0.262^{c}	1.068 ± 0.322^{bc}	1.247 ± 0.399^{ab}	1.484 ± 0.466^{a}	0.708 ± 0.234^{c}	0.995 ± 0.344^{b}	1.178 ± 0.366^{b}	1.411 ± 0.479^{a}	1.631 ± 0.432^{a}
GARHWAL															
Baseline	0.091 ± 0.001^{d}	0.157 ± 0.001^{c}	0.168 ± 0.053^{bc}	0.228 ± 0.005^{ab}	0.270 ± 0.026^{a}	0.011 ± 0.001^{d}	0.015 ± 0.002^{c}	0.019 ± 0.001^{bc}	0.019 ± 0.001^{ab}	0.016 ± 0.002^{a}	0.027 ± 0.004^{a}	0.029 ± 0..009^{a}	0.028 ± 0.005^{a}	0.030 ± 0.005^{a}	0.032 ± 0.002^{a}
Dhanolti	0.578 ± 0.040^{d}	0.910 ± 00.096^{c}	1.133 ± 0.177^{b}	1.306 ± 0.248^{b}	1.617 ± 0.110^{a}	0.406 ± 0.082^{d}	0.643 ± 0.055^{c}	0.797 ± 0.02bc	0.910 ± 0.070^{b}	1.126 ± 0.139^{a}	0.489 ± 0.014^{d}	0.753 ± 0.076^{cd}	0.966 ± 0.112^{bc}	1.153 ± 0.168^{b}	1.473 ± 0.389^{a}
Mussoorie	0.987 ± 0.065^{e}	1.269 ± 0.092^{d}	1.541 ± 0.252^{c}	1.749 ± 0.185^{b}	1.968 ± 0.209^{a}	0.659 ± 0.155^{d}	0.876 ± 0.167^{c}	1.087 ± 0.217^{b}	1.250 ± 0.275^{b}	1.498 ± 0.293^{a}	0.733 ± 0.106^{d}	1.041 ± 0.127^{c}	1.260 ± 0.153^{bc}	1.421 ± 0.252^{b}	1.713 ± 0.327^{a}
Kampty Fall	0.839 ± 0.174^{e}	1.069 ± 0.130^{d}	1.298 ± 0.110^{c}	1.559 ± 0.192^{b}	1.851 ± 0.193^{a}	0.613 ± 0.208^{b}	0.892 ± 0.280^{c}	0.996 ± 0.301^{bc}	1.177 ± 0.357^{ab}	1.433 ± 0.411^{a}	0.648 ± 0.189^{d}	0.911 ± 0.213^{cd}	1.163 ± 0.267^{bc}	1.364 ± 0.397^{ab}	1.591 ± 0.424^{a}

Values are represented as mean ± SD.
*Levels not connected by the same letters in horizontal row are yearly significantly different during same season.

Table 2. Average data of Lead (mg g^{-1} DW) during five consecutive years (2008–2012) in *Rhodobryum giganteum* at different sites in Kumaon and Garhwal region of Uttarakhand.

Sites	SUMMER					MONSOON					WINTER				
	2008	2009	2010	2011	2012	2008	2009	2010	2011	2012	2008	2009	2010	2011	2012
KUMAON															
Baseline	0.085 ± 0.003^{b}	0.107 ± 0.006^{ab}	0.122 ± 0.010^{a}	0.118 ± 0.010^{a}	0.132 ± 0.024^{a}	0.005 ± 0.002^{a}	0.006 ± 0.002^{a}	0.009 ± 0.001^{a}	0.010 ± 0.005^{a}	0.011 ± 0.003^{a}	0.013 ± 0.002^{a}	0.018 ± 0.004^{a}	0.017 ± 0.002^{a}	0.017 ± 0.002^{a}	0.017 ± 0.002^{a}
Almora	0.521 ± 0.088^{e}	0.851 ± 0.115^{d}	1.023 ± 0.131^{c}	1.196 ± 0.138^{b}	1.332 ± 0.106^{a}	0.395 ± 0.074^{d}	0.593 ± 0.108^{c}	0.711 ± 0.115^{c}	0.884 ± 0.117^{b}	1.030 ± 0.122^{a}	0.517 ± 0.120^{d}	0.759 ± 0.081^{c}	0.946 ± 0.171^{b}	1.079 ± 0.126^{ab}	1.212 ± 0.120^{a}
Nainital	0.908 ± 0.193^{d}	1.246 ± 0.126^{c}	1.498 ± 0.198^{b}	1.684 ± 0.279^{ab}	1.869 ± 0.304^{a}	0.591 ± 0.138^{d}	0.887 ± 0.168^{c}	1.094 ± 0.233^{b}	1.260 ± 0.243^{b}	1.540 ± 0.302^{a}	0.682 ± 0.143^{d}	1.042 ± 0.151^{c}	1.250 ± 0.186^{b}	1.401 ± 0.214^{b}	1.614 ± 0.291^{a}
Pithoragarh	0.779 ± 0.275^{d}	1.139 ± 0.301^{c}	1.290 ± 0.362^{bc}	1.522 ± 0.368^{ab}	1.719 ± 0.446^{a}	0.607 ± 0.235^{d}	0.821 ± 0.233^{c}	0.958 ± 0.253^{bc}	1.106 ± 0.333^{b}	1.385 ± 0.379^{a}	0.698 ± 0.263^{e}	0.889 ± 0.254^{d}	1.052 ± 0.315^{c}	1.214 ± 0.406^{b}	1.514 ± 0.387^{a}
GARHWAL															
Baseline	0.087 ± 0.006^{c}	0.110 ± 0.004^{b}	0.108 ± 0.002^{b}	0.135 ± 0.003^{a}	0.139 ± 0.008^{a}	0.010 ± 0.001^{b}	0.018 ± 0.001^{a}	0.021 ± 0.004^{a}	0.022 ± 0.003^{a}	0.023 ± 0.003^{a}	0.022 ± 0.005^{d}	0.030 ± 0.001^{c}	0.041 ± 0.005^{b}	0.044 ± 0.006^{ab}	0.049 ± 0.003^{a}
Dhanolti	0.526 ± 0.092^{e}	0.797 ± 0.095^{d}	0.958 ± 0.139^{c}	1.114 ± 0.156^{b}	1.249 ± 0.116^{a}	0.391 ± 0.076^{e}	0.581 ± 0.090^{d}	0.741 ± 0.088^{c}	0.922 ± 0.092^{b}	1.054 ± 0.096^{a}	0.404 ± 0.075^{d}	0.643 ± 0.056^{c}	0.834 ± 0.137^{b}	1.012 ± 0.133^{a}	1.085 ± 0.102^{a}
Mussoorie	0.916 ± 0.198^{d}	1.191 ± 0.093^{c}	1.412 ± 0.183^{b}	1.605 ± 0.228^{b}	1.811 ± 0.274^{a}	0.587 ± 0.158^{e}	0.803 ± 0.158^{d}	0.995 ± 0.231^{c}	1.154 ± 0.245^{b}	1.444 ± 0.239^{a}	0.607 ± 0.135^{d}	0.966 ± 0.168^{c}	1.121 ± 0.193^{c}	1.294 ± 0.258^{b}	1.595 ± 0.270^{a}
Kampty Fall	0.706 ± 0.223^{d}	1.045 ± 0.343^{c}	1.257 ± 0.359^{bc}	1.454 ± 0.418^{ab}	1.665 ± 0.444^{a}	0.549 ± 0.224^{d}	0.758 ± 0.199^{cd}	0.905 ± 0.205^{bc}	1.099 ± 0.309^{ab}	1.302 ± 0.396^{a}	0.590 ± 0.245^{d}	0.786 ± 0.260^{c}	0.990 ± 0.331^{b}	1.136 ± 0.374^{b}	1.411 ± 0.403^{a}

Values are represented as mean ± SD.
*Levels not connected by the same letters in horizontal row are yearly significantly different during same season.

Table 3. Average data of Copper (mg g^{-1} DW) during five consecutive years (2008–2012) in *Rhodobryum giganteum* at different sites in Kumaon and Garhwal region of Uttarakhand.

Sites	SUMMER					MONSOON					WINTER				
	2008	2009	2010	2011	2012	2008	2009	2010	2011	2012	2008	2009	2010	2011	2012
KUMAON															
Baseline	0.077 ± 0.002[b]	0.085 ± 0.005[b]	0.088 ± 0.001[b]	0.569 ± 0.312[a]	0.779 ± 0.186[a]	0.009 ± 0.002[c]	0.013 ± 0.002[bc]	0.016 ± 0.003[ab]	0.018 ± 0.001[ab]	0.019 ± 0.002[a]	0.015 ± 0.006[b]	0.017 ± 0.002[b]	0.019 ± 0.001[b]	0.021 ± 0.005[b]	0.051 ± 0.010[a]
Almora	0.464 ± 0.088[d]	0.653 ± 0.106[c]	0.780 ± 0.058[c]	0.950 ± 0.100[b]	1.145 ± 0.118[a]	0.307 ± 0.065[e]	0.417 ± 0.055[d]	0.528 ± 0.059[c]	0.639 ± 0.112[b]	0.822 ± 0.108[a]	0.378 ± 0.026[e]	0.539 ± 0.097[d]	0.712 ± 0.084[c]	0.849 ± 0.049[b]	0.995 ± 0.068[a]
Nainital	0.825 ± 0160[d]	1.095 ± 0.183[c]	1.277 ± 0.191[bc]	1.504 ± 0.256[ab]	1.671 ± 0.331[a]	0.540 ± 0.156[d]	0.615 ± 0.056[cd]	0.780 ± 0.092[c]	0.963 ± 0.123[b]	1.248 ± 0.210[a]	0.576 ± 0.163[d]	0.886 ± 0.162[c]	1.023 ± 0.196[b]	1.177 ± 0.187[b]	1.394 ± 0.226[a]
Pithoragarh	0.614 ± 0.193[d]	0.885 ± 0.224[c]	1.075 ± 0.287[b]	1.157 ± 0.24[b]	1.419 ± 0.323[a]	0.527 ± 0.200[d]	0.674 ± 0.286[c]	0.790 ± 0.309[bc]	0.905 ± 0.312[b]	1.110 ± 0.298[a]	0.526 ± 0.194[d]	0.718 ± 0.270[c]	0.866 ± 0.318[bc]	1.033 ± 0.356[b]	1.284 ± 0.207[a]
GARHWAL															
Baseline	0.064 ± 0.003[c]	0.092 ± 0.005[b]	0.119 ± 0.016[a]	0.135 ± 0.012[a]	0.144 ± 0.013[a]	0.005 ± 0.001[b]	0.009 ± 0.001[b]	0.015 ± 0.003[a]	0.016 ± 0.002[a]	0.019 ± 0.002[a]	0.008 ± 0.001[e]	0.017 ± 0.002[d]	0.022 ± 0.002[c]	0.032 ± 0.002[b]	0.104 ± 0.002[a]
Dhanolti	0.416 ± 0.042[d]	0.614 ± 0.070[c]	0.750 ± 0.048[bc]	0.881 ± 0.062[b]	1.112 ± 0.125[a]	0.262 ± 0.060[d]	0.366 ± 0.060[d]	0.502 ± 0.044[c]	0.617 ± 0.099[bs]	0.757 ± 0.095[a]	0.328 ± 0.048[e]	0.486 ± 0.081[d]	0.621 ± 0.059[c]	0.769 ± 0.081[b]	0.950 ± 0.053[a]
Mussoorie	0.699 ± 0.137[d]	1.026 ± 0.178[c]	1.188 ± 0.214[c]	1.399 ± 0.227[b]	1.596 ± 0.277[a]	0.473 ± 0.132[d]	0.630 ± 0.127[c]	0.749 ± 0.142[bc]	0.879 ± 0.157[b]	1.124 ± 0.215[a]	0.539 ± 0.166[d]	0.862 ± 0.150[c]	1.000 ± 0.127[c]	1.160 ± 0.183[b]	1.365 ± 0.239[a]
Kampty Fall	0.552 ± 0.197[e]	0.858 ± 0.226[d]	1.023 ± 0.284[c]	1.159 ± 0.261[b]	1.376 ± 0.304[a]	0.437 ± 0.175[d]	0.573 ± 0.233[c]	0.679 ± 0.246[bc]	0.748 ± 0.257[b]	1.020 ± 0.287[a]	0.489 ± 0.179[d]	0.684 ± 0.275[c]	0.860 ± 0.288[b]	0.935 ± 0.268[b]	1.190 ± 0.243[a]

Values are represented as mean ± SD.
*Levels not connected by the same letters in horizontal row are yearly significantly different during same season.

Table 4. Average data of Cadmium (mg g^{-1} DW) during five consecutive years (2008–2012) in *Rhodobryum giganteum* at different sites in Kumaon and Garhwal region of Uttarakhand.

Sites	SUMMER					MONSOON					WINTER				
	2008	2009	2010	2011	2012	2008	2009	2010	2011	2012	2008	2009	2010	2011	2012
KUMAON															
Baseline	0.006 ± 0.002^{a}	0.049 ± 0.043^{a}	0.036 ± 0.037^{a}	0.023 ± 0.004^{a}	0.030 ± 0.010^{a}	0.002 ± 0.001^{c}	0.004 ± 0.001^{bc}	0.005 ± 0.001^{bc}	0.008 ± 0.001^{ab}	0.010 ± 0.005^{a}	0.005 ± 0.001^{c}	0.007 ± 0.001^{bc}	0.010 ± 0.002^{bc}	0.013 ± 0.003^{ab}	0.019 ± 0.007^{ab}
Almora	0.099 ± 0.019^{d}	0.165 ± 0.033^{c}	0.221 ± 0.028^{bc}	0.258 ± 0.034^{ab}	0.297 ± 0.035^{a}	0.043 ± 0.017^{c}	0.075 ± 0.020^{bc}	0.111 ± 0.046^{bc}	0.169 ± 0.093^{ab}	0.224 ± 0.099^{a}	0.065 ± 0.025^{c}	0.097 ± 0.037^{c}	0.146 ± 0.080^{bc}	0.227 ± 0.119^{ab}	0.314 ± 0.132^{a}
Nainital	0.103 ± 0.074^{c}	0.157 ± 0.064^{bc}	0.211 ± 0.096^{bc}	0.300 ± 0.152^{ab}	0.389 ± 0.200^{a}	0.025 ± 0.012^{d}	0.043 ± 0.020^{cd}	0.064 ± 0.023^{bc}	0.078 ± 0.030^{b}	0.125 ± 0.035^{a}	0.038 ± 0.011^{c}	0.062 ± 0.019^{bc}	0.077 ± 0.028^{b}	0.099 ± 0.027^{b}	0.161 ± 0.018^{a}
Pithoragarh	0.103 ± 0.026^{c}	0.174 ± 0.012^{bc}	0.227 ± 0.028^{bc}	0.330 ± 0.096^{ab}	0.438 ± 0.173^{a}	0.025 ± 0.007^{c}	0.047 ± 0.010^{bc}	0.072 ± 0.011^{abc}	0.087 ± 0.033^{ab}	0.091 ± 0.065^{a}	0.037 ± 0.007^{d}	0.051 ± 0.014^{cd}	0.097 ± 0.018^{b}	0.129 ± 0.015^{ab}	0.156 ± 0.028^{a}
GARHWAL															
Baseline	0.009 ± 0.001^{c}	0.011 ± 0.001^{c}	0.022 ± 0.001^{b}	0.025 ± 0.001^{a}	0.027 ± 0.002^{a}	0.002 ± 0.001^{c}	0.002 ± 0.001^{c}	0.003 ± 0.001^{bc}	0.004 ± 0.001^{ab}	0.005 ± 0.001^{a}	0.002 ± 0.001^{b}	0.003 ± 0.001^{b}	0.003 ± 0.001^{b}	0.008 ± 0.001^{a}	0.009 ± 0.001^{a}
Dhanolti	0.071 ± 0.040^{c}	0.118 ± 0.032^{bc}	0.138 ± 0.023^{b}	0.163 ± 0.026^{b}	0.220 ± 0.021^{a}	0.019 ± 0.007^{d}	0.033 ± 0.008^{cd}	0.044 ± 0.007^{bc}	0.056 ± 0.005^{b}	0.099 ± 0.015^{a}	0.034 ± 0.011^{d}	0.051 ± 0.009^{c}	0.069 ± 0.009^{b}	0.081 ± 0.009^{b}	0.134 ± 0.022^{a}
Mussoorie	0.071 ± 0.020^{d}	0.120 ± 0.019^{c}	0.139 ± 0.023^{bc}	0.168 ± 0.032^{b}	0.244 ± 0.031^{a}	0.031 ± 0.012^{e}	0.052 ± 0.019^{d}	0.068 ± 0.018^{c}	0.082 ± 0.017^{b}	0.109 ± 0.012^{a}	0.038 ± 0.010^{c}	0.061 ± 0.019^{bc}	0.077 ± 0.017^{b}	0.090 ± 0.013^{b}	0.152 ± 0.034^{a}
Kampty Fall	0.089 ± 0.020^{d}	0.107 ± 0.016^{cd}	0.119 ± 0.017^{c}	0.147 ± 0.021^{b}	0.214 ± 0.010^{a}	0.022 ± 0.003^{e}	0.035 ± 0.007^{d}	0.050 ± 0.008^{c}	0.069 ± 0.013^{b}	0.086 ± 0.012^{a}	0.032 ± 0.005^{d}	0.046 ± 0.012^{cd}	0.060 ± 0.015^{bc}	0.076 ± 0.012^{b}	0.120 ± 0.025^{a}

Values are represented as mean ± SD.
*Levels not connected by the same letters in horizontal row are yearly significantly different during same season.

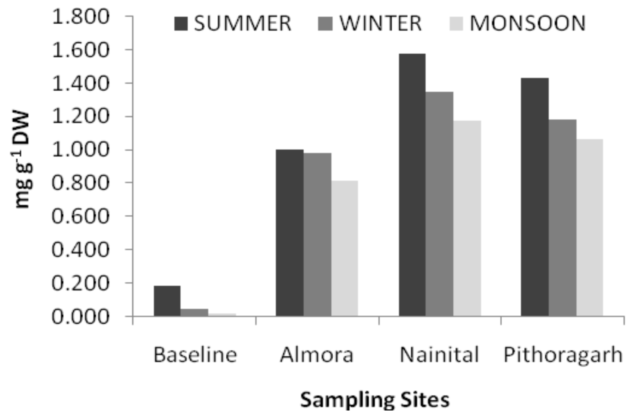

Fig. 2. Annual average metal load of zinc (mg g^{-1} DW) during different seasons at Kumaon region (2008–2012).

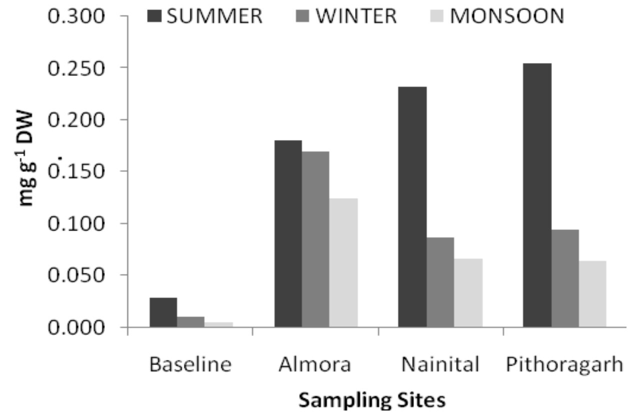

Fig. 5. Annual average metal load of cadmium (mg g^{-1} DW) during different seasons at Kumaon region (2008–2012).

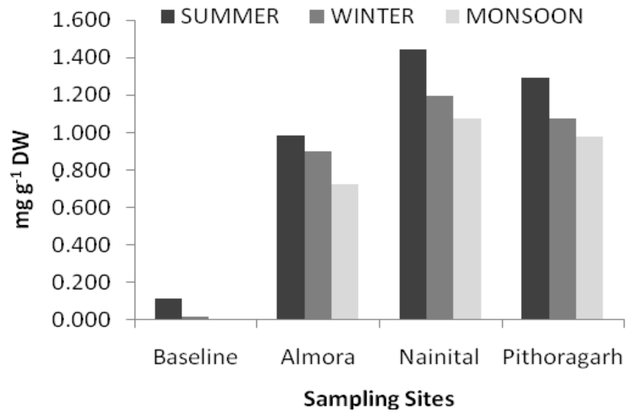

Fig. 3. Annual average metal load of lead (mg g^{-1} DW) during different seasons at Kumaon region (2008–2012).

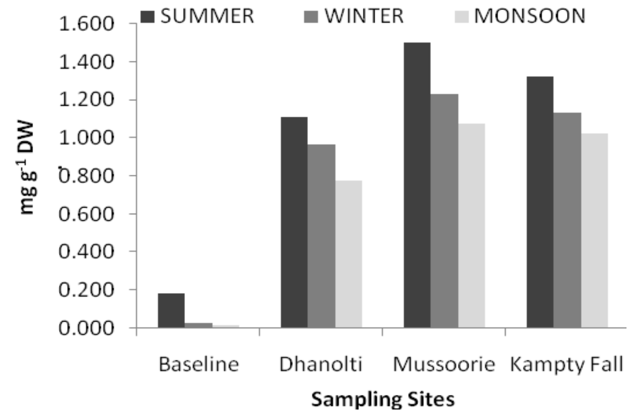

Fig. 6. Annual average metal load of zinc (mg g^{-1} DW) during different seasons at Garhwal region (2008–2012).

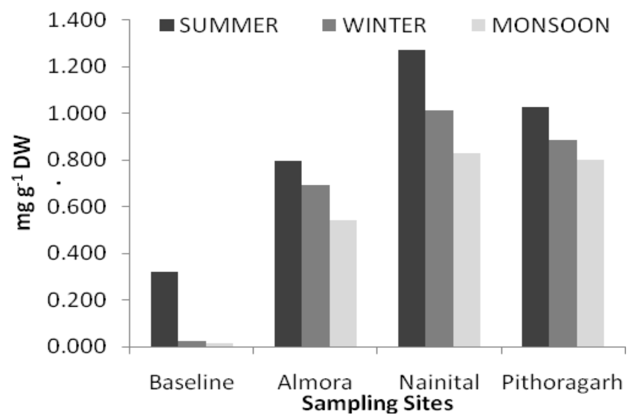

Fig. 4. Annual average metal load of copper (mg g^{-1} DW) during different seasons at Kumaon region (2008–2012).

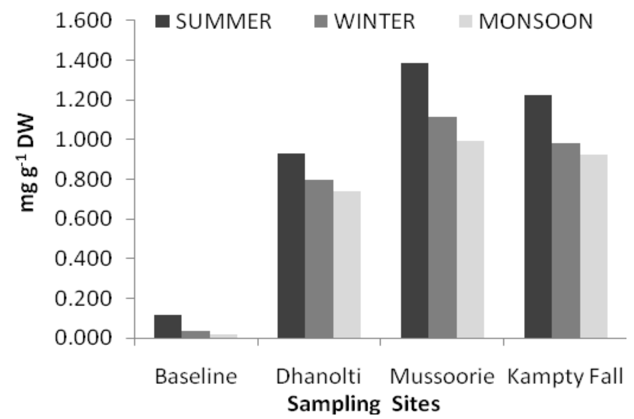

Fig. 7. Annual average metal load of lead (mg g^{-1} DW) during different seasons at Garhwal region (2008–2012).

metals was automobiles and which might have resulted from the high tourist activity in summer and winter depicting high pollution level in proximity to the cities. Although, there has been a fall in amount of lead in petrol per litre, but increase in automobile numbers has increased its consumption which ultimately has increased its concentration in the atmosphere. Besides, climatic factors could also modify the effects of metals and should be taken into consideration in any biomonitoring surveys but this role is as yet unclear. In addition, laundry and municipal waste, open burning of solid waste, agricultural practices also important sources of increasing metal load.

The present work also encourages the use of *Rhodobryum giganteum* as biomonitors using the biomapping approach and is proved to be a perfect tool for the assessment of atmospheric metal loads. Their habitat diversity, structural

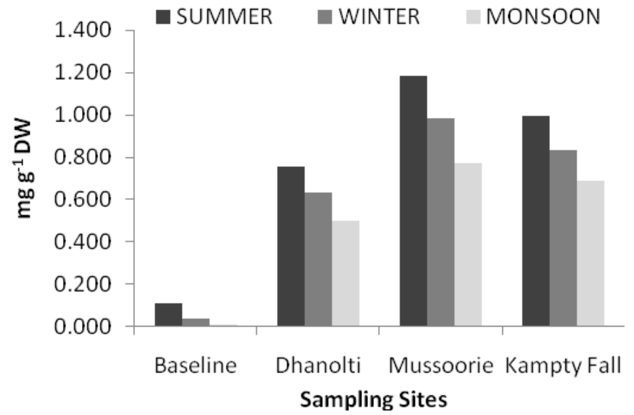

Fig. 8. Annual average metal load of copper (mg g^{-1} DW) during different seasons at Garhwal region (2008–2012).

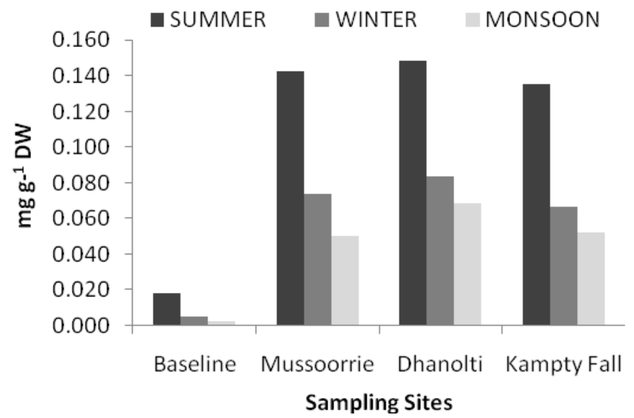

Fig. 9. Annual average metal load of cadmium (mg g^{-1} DW) during different seasons at Garhwal region (2008–2012).

Table 5. Inter-elemental correlation matrix of moss *Rhodobryum giganteum* from Kumaon hills.

	Zn	Pb	Cu	Cd
Zn	100			
Pb	996	100		
Cu	0.968	0.964	100	
Cd	0.569	0.582	0.566	100

Table 6. Inter-elemental correlation matrix of moss *Rhodobryum giganteum* from Garhwal hills.

	Zn	Pb	Cu	Cd
Zn	100			
Pb	0.992	100		
Cu	0.982	0.985	100	
Cd	0.766	0.747	0.764	100

simplicity, totipotency and rapid multiplication rate, make them an ideal organism for studies related to pollution. The research highlighted the seasonal and annual metal pollution and the extent of contamination in the study area. Study also examined the distribution pattern of under taken metals and discussed its possible sources. Furthermore, the chemical analysis of undertaken bryophytes proved that biomonitoring is the rapid and comparatively economical tool for surveying heavy metal deposition in terrestrial ecosystem and in surrounding ambient air. Furthermore, bryophytes (mosses) are supportive and functional monitoring tool, which could be helpful in biomonitoring studies around similar sources of metal pollution and to notify nearby residents about potential threatening extension of metal deposition.

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SUPPLEMENTARY MATERIAL

Supplementary data associated with this article can be found in the online version at http://www.aaqr.org.

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