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Original Research Paper

Active Biomonitoring of Atmospheric Metal Deposition by *Bryum* Species Around Almora, Nainital and Pithoragarh of Kumaon Hills, India

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ABSTRACT

Biomonitoring is a method of observing the impact of external factors on organisms and their development over a long period. The level of Zn, Cu, Cd and Pb has been determined in samples of the moss *Bryum cellulare* from the area of Almora, Nainital and Pithoragarh of Kumaon hills. A gradient was observed in metal load while projecting from forests to urban areas. Maximum metals were observed in locations proximate to higher traffic density areas, which integrated due to enhanced tourism during summer, followed by winter and monsoon season. In rural areas high value of Zn, Cu and Cd attributes to their use in fertilizers. Positive significant correlations were obtained between Pb-Zn and Zn-Cu suggesting a common origin of these metals. Interseasonal variability in metal deposition pattern shows that summer had maximum metal load followed by winter and rainy season.

INTRODUCTION

Significance of increasing heavy metal concentration in environment has created considerable attention among plant ecologists during the last few decades. Metal input through air pollution is the major threat for global warming, disproportion of seasonal fluctuation and iceberg melting (Marinova et al. 2006, Sardans et al. 2008). Therefore, monitoring of air contaminants of Kumaon hill is necessary to determine impact upon ecosystem and control measure required for abatement of their sources in their respective region (Saxena et al. 2008a). In recent decades the number and intensity of urbanization and anthropogenic sources, such as waste burning, fertilizers, vehicle emissions, agricultural and sewage sludge, have increased the overall environmental element concentration (Bargagli et al. 1998, Saxena et al. 2008b). This fact seems to be true for the Kumaon hills (India) where there are large numbers of vehicles and other human actives increased with time. This disturbs the harmony and beauty of Himalaya. Because of the specific ability to accumulate heavy metals, bryophytes are particularly suitable to indicate environmental load (Fernandez et al. 2000, Sardans & Penuelas 2005, Sun et al. 2009). The specific biology of mosses make them capable to receive and accumulate chemical substances predominantly from surrounding atmosphere without any selectivity parameter (Yurukova & Gecheva 2003, Fernandez et al. 2004). The bryophyte *Bryum* sp. has tremendous ability to absorb metals as dry fallout from the atmosphere (Schintu et al. 2005).

In present work, active monitoring (moss bags) has been used to determine total level of atmospheric deposition of heavy metals in Kumaon hills. This technique is very useful especially in such polluted areas where wild growing mosses are lacking (Makholm & Miadenoff 2005). Our earlier study confirms that moss *Bryum cellulare* is tolerant to high concentration of heavy metals (Saxena & Saiful 2006). Present study is an attempt to use moss *B. cellulare* to investigate the spatial distribution of the metal (Cu, Zn, Pb and Cd) contamination in Kumaon district during three different Indian continental seasons with the help of moss bags (transplant) technique during the year 2004.

MATERIALS AND METHODS

Study area: The Kumaon region is spread over 21,073 km^2 and has extensive tracts of natural forests until a few centuries back. The species is of wide range of distribution in Western Himalaya geographically ranging from 29°5'-31°25' N latitude to 79°43'-81°E longitude (IG 1931). The Kumaon hill is extensively divided into three major zones for the biomapping study namely Almora (longitudes 79°26'E-80°15'E; latitudes 29°15' N-30°29' N), Nainital (longitudes 79°51'E-80°18'E; latitudes 28°45' N-29°38' N) and Pithoragarh (longitudes 79°45'E-81°21'E; latitudes 29°32' N-30°47' N). The climate remains quite cold from October-April, and mild warm through May-June followed by monsoon rain till September. The average rainfall measured 80" and relative humidity range from 85 to 90% in the months of July and August. The maximum and minimum temperatures were recorded 27°C and 10°C in summer, and 15°C and 3°C in winter respectively.

Sampling: Moss *B. cellulare* was collected from the forest cover of Mukteshwar, situated at an altitude of 2300 meters, which was treated as control site. A complete green patch of moss was transplanted in nylon bags at 19 study sites of investigated areas and sufficient amount of the same moss was taken for digestion to determine the baseline concentration of metals in each season. Each moss bag was suspended 20 cm above the ground in triplicate. These moss bags were transplanted cross section-wise in all the four directions and harvested after an exposure period of four months i.e., first week of November (for monsoon monitoring), March (for winter monitoring) and July (for summer monitoring).

Metal analysis: Upon return to the laboratory, harvested moss samples were oven dried at 40°C for 24 hours. Prior to analysis, adhering substrates and litter were removed by hand with great care to avoid metal contamination. Triplicate samples were digested with concentrated HNO_3 and $HClO_4$ in ratio of 4:1 v/v on a hot plate (Kolthoff 1971). The digestion was completed after all organic material had disappeared. The extract obtained was filtered and the filtrate was made up to a final volume of 50 mL by double distilled water and fraction was quantitatively analysed by atomic absorption spectrophotometer. Suitable blanks were used to check for possible contamination during extraction.

Data analysis: Samples were collected in triplicate to conduct the statistical analysis (Snedecor & Cochran 1967). ANOVA revealed significant differences in the metal concentration at different distances and seasons ($p \leq 0.05$) utilizing Dunkun's multiple range test (Karmer 1956) and were cartographically represented using program package Surfer (Golden Software Inc., U. S. A.). Pollution index (PI) of the study area was used to calculate pollution index value on the basis of each metal, total number of metals, total amount of metals and number of samples collected from predefined area of the district (Grodzinska et al. 1993).

RESULTS

After the exposure of moss during winter, summer and monsoon, the metal values, calculated in the seasonal and regional sampling, are represented cartographically for the year 2004 in Figs. 1 to 12. The cartographic sketch was drawn after statistically calculating the mean value of each metal and its significance (ANOVA at $P \leq 0.05$ and $P \leq 0.01$) compared with the control site, i.e., Mukteswar. Results are being characterized on exposure of moss *Bryum cellulare* in catchment areas of Nainital, Almora and Pithoragarh districts of Kumaon hills. It was found that elemental concentration in *B. cellulare* indeed reflected atmospheric elemental deposition.

Lead concentration in moss *B. cellulare* from the experimental area fluctuated both seasonally and regionally as evident from Figs. 1, 5 and 9). During winter, Nainital Tallital exhibited maximum value of 50.45 µg/g, in summer it was peaked at bus stand area of Nainital with 71.48 µg/g and in monsoon 35.43 µg/g was maximum at Mallital. The lower values were measured in moss *B. cellulare* from Mukteswar forest (7.46, 16.92 and 9.32 µg/g) during winter, summer and monsoon seasons respectively (Figs. 1, 5 and 9). The Tallital and bus stand areas of Nainital showed maximum value of Pb deposition in xeric the moss. The results of the present study at Jageshwar show that there is 83.89 % decrease in lead in monsoon with respect to summer season in year 2004 (Fig. 14).

Table 1: Comparison of pollution index (PI) value of year 2004 determined by using moss *B. cellulare* as a biomonitoring species exposed in Kumaon hills.

Catchment Sites	Value of Index
Mukteswar forest	-0.4003
Deenapnai	-0.08794
Jageswar	0.054387
Hwalbagh	0.795751
Almora (Mall Road)	1.54167
Kosi (G.B. Pant Institute)	1.636119
Almora (Petrol pump)	1.596236
Ranikhet (Golf court)	0.517413
Ranikhet (Bus stand)	0.024498
Ranikhet (Petrol pump)	1.086629
Nainital (Bus stand)	1.463346
Nainital (Mallital)	0.401092
Nainital (Tallital)	0.206162
Nainital (Capitol cinema)	0.831391
Bhowali (Bus stand)	1.104417
Ramgarh (Ramgarh malla)	-0.02735
Pithoragarh (Siltham)	0.02067
Pithoragarh (Market area)	1.126227
Pithoragarh (Bus stand)	0.204273

Pithoragarh bus stand shows 51.98 % increase in Zn in summer season in reference to winter, which was significant (Fig. 14). The monsoon season exhibits completely different picture for this metal in moss. The sites of Pithoragarh bus stand and Nainital Capitol cinema had 77 % decrease in Zn concentration with respect to summer in *B. cellulare* (Fig. 13). Significantly, control site Mukteswar forest showed around 56 % decreases in the metal in rainy season with reference to summer (Fig. 14).

The comparative study on the seasonal basis revealed that summer is the season for high Cu in moss. Maximum value of copper was in Kosi and Ranikhet golf court during all the three seasons (Figs. 3, 7 and 11). A 25-50 % decrease in Cu content in moss *B. cellulare* was observed in the catchment areas of Nainital in winter season with respect to summer in year 2004. The catchments area of Mukteswar and Almora (Mall Road) exhibited 72 % decrease of Cu in rainy season in reference to summer (Fig. 14).

On basis of the indirect method of distribution study, samples containing somewhat higher amount of Cd are localized at Almora Mall Road and Hawalbagh. There was about 94.73 % increase in Cd at Ranikhet bus stand in winter with respect to summer was a striking outcome (Fig. 14).

Percent metal load and pollution index value: The Nainital (Bus stand), Kosi and Pithoragarh bus stand had maximum percent loading of metal up to 6.3, 6.46 and 7.27 % during the year 2004 (Fig. 14). For the present study Mukteswar forest was considered to be control site. There is a continuous decrease in percent metal load at Mukteswar, deploy that our consideration of taking Mukteswar as control site is significant. The percent (%) metal load at Mukteswar was -1.7015 in year 2004 (Fig. 14).

The pollution index (PI) value at Nainital bus stand, Almora Mall Road, Kosi, Almora petrol pump, Ranikhet petrol pump and Pithoragarh market area was +1.4633, +1.5416, +1.6361, +1.5962, + 1.0866 and +1.1262 respectively during year 2004 (Table 1).

DISCUSSION

Through active monitoring it is easy to depict the interseasonal and regional variability in metal deposition around the biomonitoring organism (Sloof 1993, Wolterbeek et al. 1995, Sun et al. 2009).

Lead: The high input of petrol through vehicular exhaust (Thoni et al. 1996, Halleraker et al. 1998) could be the reason for its high value near proximity to the town, along the roads or near Nainital bus stand which intensified during summer (Fig. 5). The results of the present study further justify the statement that at tourist places there is always increasing trend of vehicles; consequently increase in the metals associated with the vehicular pollution (Zechmeister 1998, De Caritat et al. 2001, Fernandez et al. 2002, Poikolainen et al. 2004). Many-fold increase in Pb value in summer (Fig. 5) could be due to enhanced traffic in summer season. In the arid seasons metals normally originate from soil into air and get accumulated more readily in mosses (Bargagli et al. 2002), which could be the reason for increase of metals in summer. The overall maximum percent of Pb load in location of Nainital bus stand, Almora petrol pump and Ranikhet golf court is in line with their maximum value in these areas (Fig. 14).

Summer is the tourist season for Kumaon hill, and due to increase in Indian economy the concept of holiday making is approachable to average Indian. These hill stations find themselves choked with tourist traffic in summer and indeed they are more crowded in each season except monsoon (Saxena & Saxena 2000, Gupta & Gupta 2001). A decrease in lead load during monsoon at Mall Road of Almora could be due to the atmospheric metal load, which is washed during heavy rains (Fig. 9).

Decrease in Pb concentration in rainy season could be attributed to decrease in tourist traffic, pollutant leaching and increase in growth and biomass more rapidly which reduces the metal percentage in leaves in proportion to biomass.

Zinc: Maximum percent increase of Zn was observed at the same spots similar to Pb. The value of regression correlation ($R^2 = 0.8819$) for Pb-Zn during year 2004 in Almora and Pithoragarh deployed their coexistence and common source (Fig. 13).

The locations closed to orchards sites like Kosi, Ranikhet golf court, which are used to grow fruits (apples, peach, plums, pear, oranges) in which Zn was applied as a nutrient, exhibited increase in Zn in summer (Fig. 6). Increase in Zn in this area is amply documented by the fact that zinc is used as element to promote the growth of orchards (Saxena & Saxena 2000) and was applied through foliar or aerial spray.

A seasonal trend for Zn justifying that summer has the high value of Zn followed by winter and rain (Figs. 2, 6 and 10). Heavy rainfall could be one another way to explain the decrease in Zn load as most of the Zn was surface adsorbed.

Copper: In comparison to Pb and Zn, a moderate deposition rate of copper was observed in the same catchment area. Cu was high in populated as well as rural areas (Figs. 3, 7 and 11). Maximum value of copper was in Almora during all the three seasons. Therefore, present finding is further supported by the presence of abundant Cu mosses in these areas. A decrease in Cu in winter with respect to summer could be due to dry deposition, which increases on moving from humid to arid climates (Berg & Steinnens 1997, Couto et al. 2004).

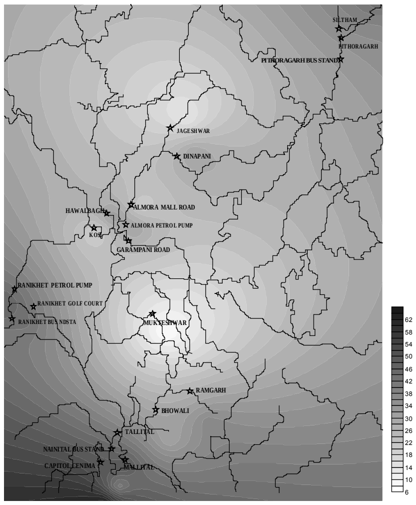

Fig. 1: Distribution map of Pb content in *B. cellulare* (µg g^{-1} dw) during winter 2004.

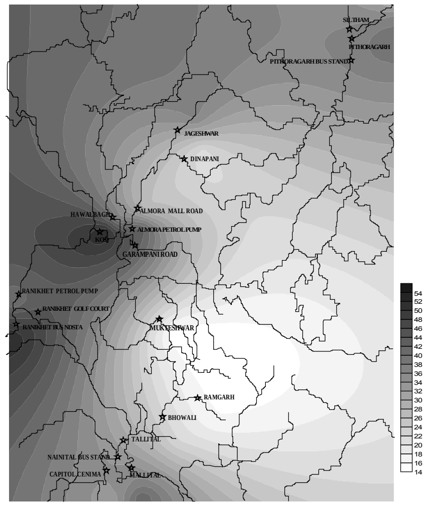

Fig. 2: Distribution map of Zn content in *B. cellulare* (µg g^{-1} dw) during winter 2004.

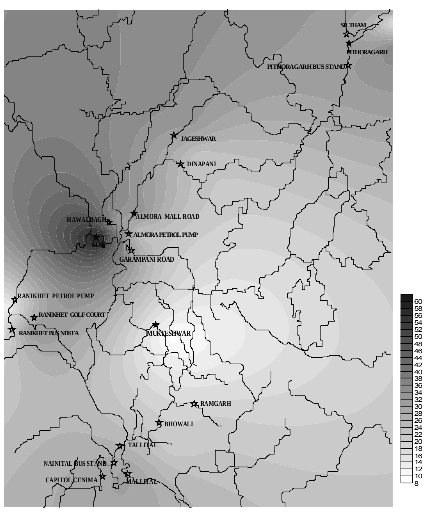

Fig. 3: Distribution map of Cu content in *B. cellulare* (µg g^{-1} dw) during winter 2004.

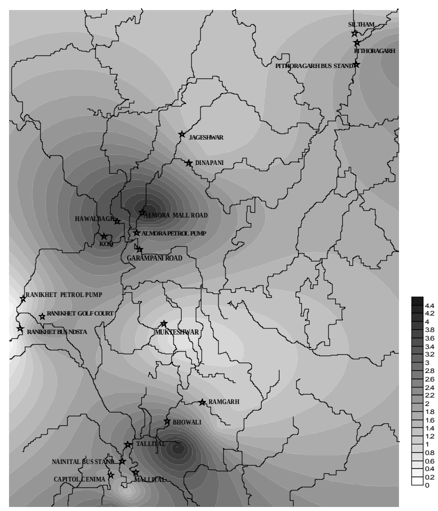

Fig. 4: Distribution map of Cd content in *B. cellulare* (µg g^{-1} dw) during winter 2004.

Concentration and distribution pattern of both Zn and Cu in *B. cellulare* were quite similar and both the metals were high in rural transplants located in vicinity of orchards. Cu contamination mainly originates from fertilizers, fungicides and pesticides used in agricultural areas (Otves 2003). A significant correlation was found between the Zn and Cu content ($R^2 = 0.8297$) in *B. cellulare* of Kumaon hill (Fig. 13). Cu pollution may also originate from domestic waste disposal. The use of

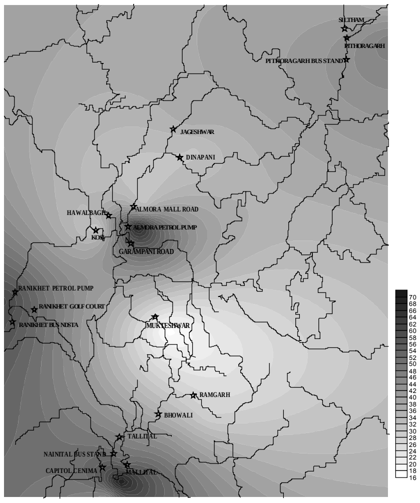

Fig. 5: Distribution map of Pb content in *B. cellulare* (µg g^{-1} dw) during Summer 2004.

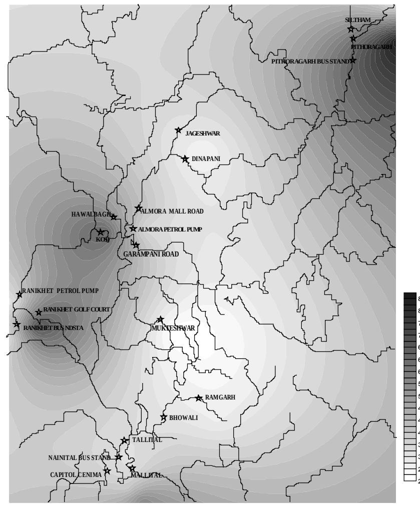

Fig. 6: Distribution map of Zn content in *B. cellulare* (µg g^{-1} dw) during summer 2004.

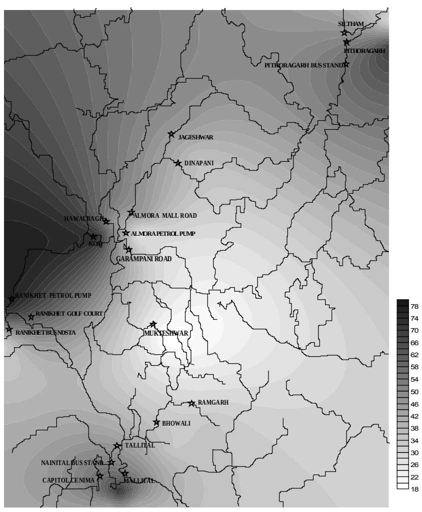

Fig. 7: Distribution map of Cu content in *B. cellulare* (µg g^{-1} dw) during summer 2004.

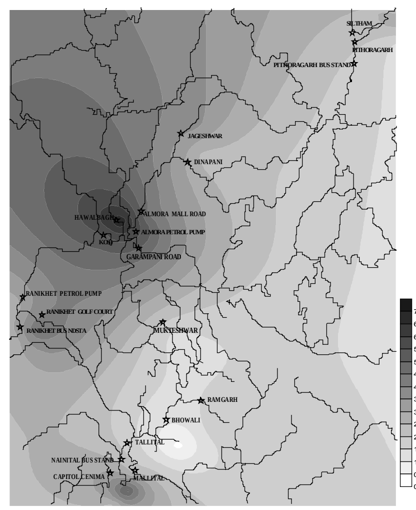

Fig. 8: Distribution map of Cd content in *B. cellulare* (µg g^{-1} dw) during summer 2004.

$CuSO_4$ mixed kerosene oil could also be one of the facts of increase of Cu concentration in domestic areas.

Cadmium: The enrichment ratios for Cd in moss were not constant throughout the year; therefore, interpretation of enrichment of Cd is complicated (Ross 1990). Beside as a nutrient, farmers also implicate zinc as Cd-based insecticides or fungicides.

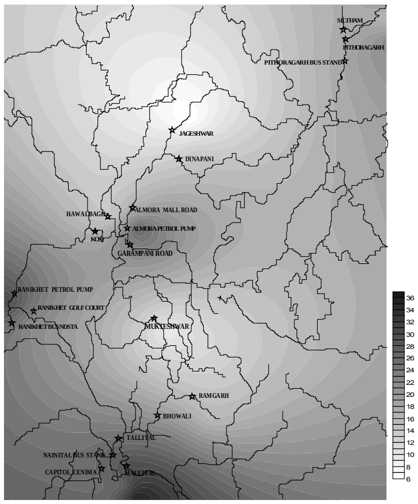

Fig. 9: Distribution map of Pb content in *B. cellulare* (µg g^{-1} dw) during monsoon 2004.

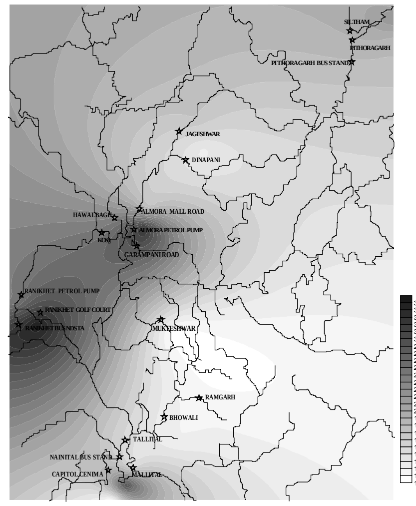

Fig. 10: Distribution map of Zn content in *B. cellulare* (µg g^{-1} dw) during monsoon 2004.

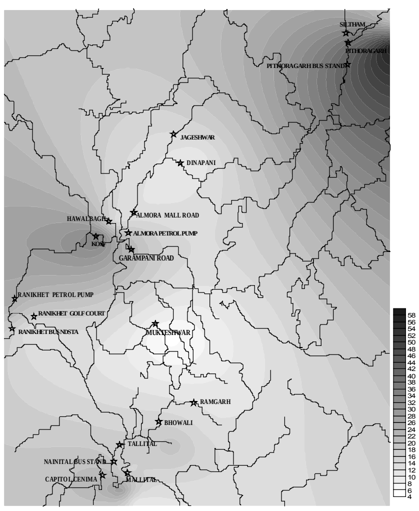

Fig.11: Distribution map of Cu content in *B. cellulare* (µg g^{-1} dw) during monsoon 2004.

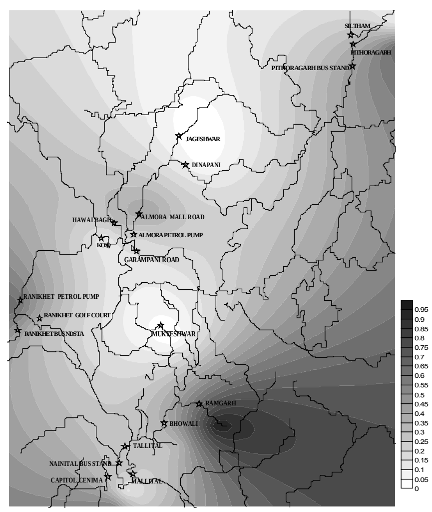

Fig. 12: Distribution map of Cd content in *B. cellulare* (µg g^{-1} dw) during monsoon 2004.

Cadmium is a metal which is very easily leached out (Fernández et al. 2009) and the similar results were observed in monsoon season in *B. cellulare*, where almost all catchment areas show significant ($P \leq 0.01$) decrease in Cd value in rainy season with respect to summer (Figs. 4, 8 and 12). Indeed the low value was even in the urban areas. There is a high value of Cd at Ranikhet (petrol pump) in winter (Fig. 8). There is a frequent use of breaks in the hilly region compared to the land

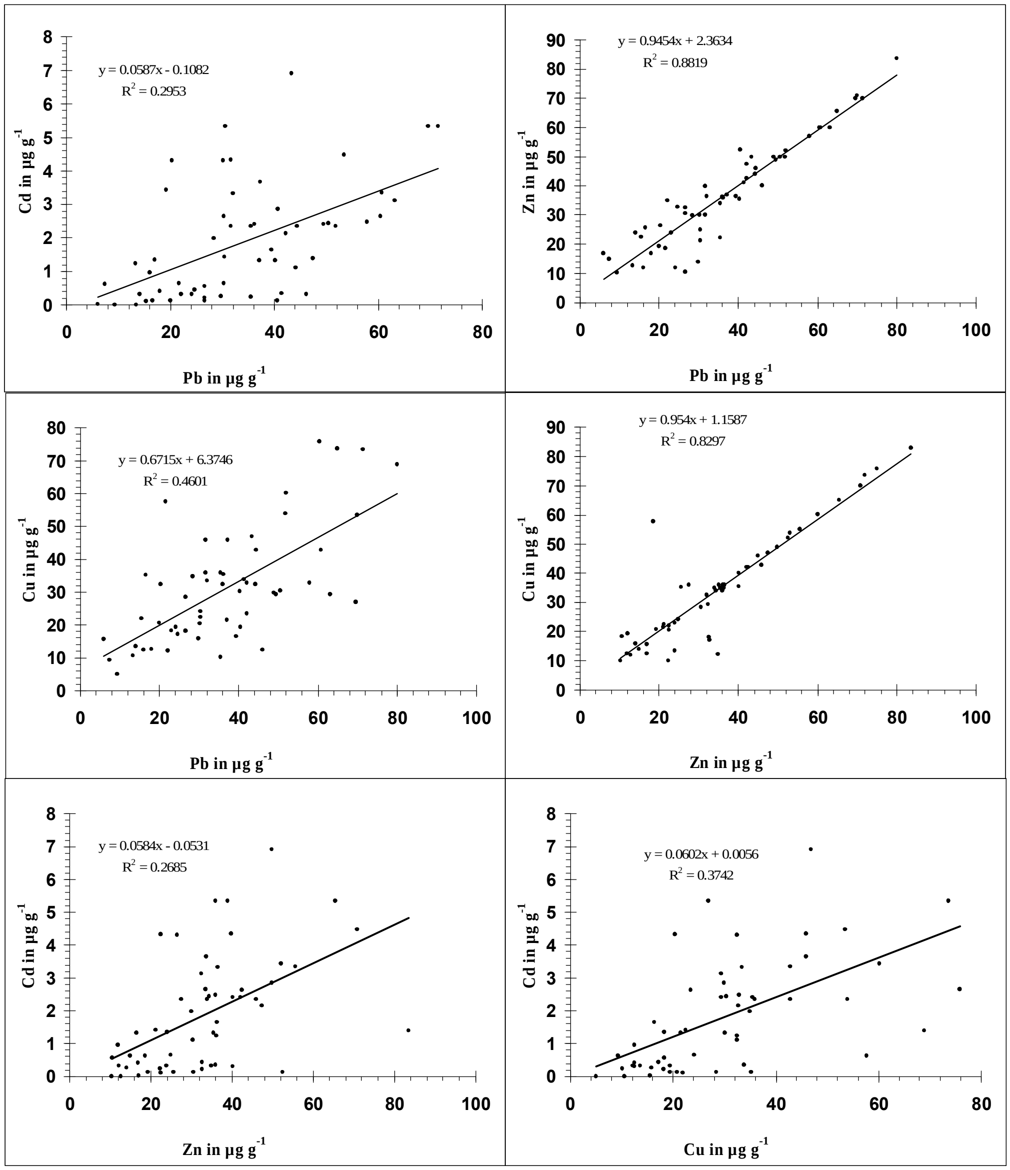

Fig 13: Correlation coefficients amongst metal concentration in Kumaon Hill (India)through active monitoring in year 2004 using *R. crispulum* as a biomonitoring species.

areas. An increase in the Cd on such places could be from abrasion of clutch and breaks of vehicles. Batteries could also be one of the sources for high level of Cd in urban areas. Higher concentration in agricultural land might be due to the use of phosphatic fertilizers (Otvos 2003). Perhaps Cd is also present in petrol as mining impurities. Service shops of batteries, bicycles, automobiles and metal utensils are the other factors for this elevation.

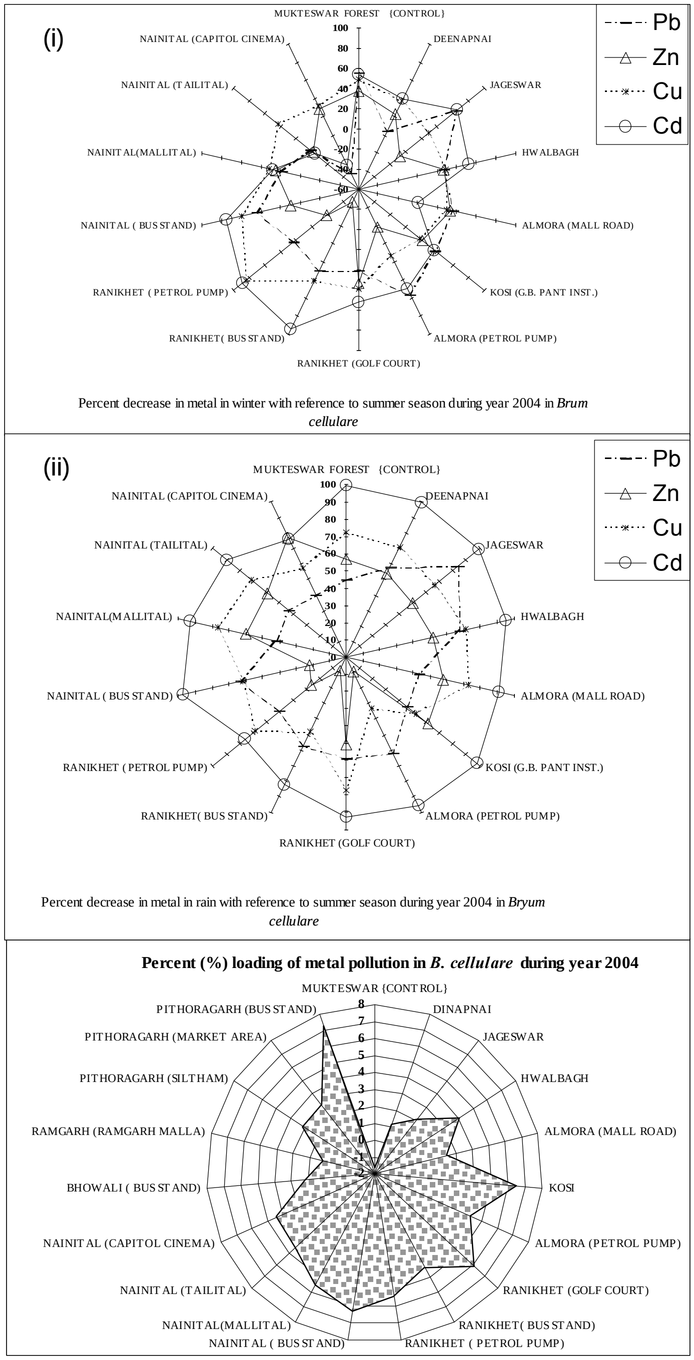

Fig. 14: Percent decrease in metal in (i) winter and (ii) monsoon with reference to summer season during year 2004. (iii) percent metal load (%) by using moss *Bryum cellulare* as a biomonitoring species exposed in Kumaon hills.

Percent metal load and pollution index value: The pollution index (PI) values at Nainital bus stand, Almora Mall Road, Kosi, Almora petrol pump, Ranikhet petrol pump and Pithoragarh market area were above one in positive sign representing the pollution input.

Continuous increase in metal load at Nainital (Tallital) during experimental year 2004 was a significant result, which could be explained by taking into consideration the internal environmental factors like meta-data influencing the metal deposition (Gjengedal & Steinnes 1990, Ceburnis & Valiulis 1999). In addition, converting the values measured in biomonitor into deposition value is problematic because there is not enough information available about the factors affecting the concentration in mosses (Wolterbeek et al. 2003, Berg et al. 2003).

The pollution index (PI) value was highly positive at the areas near bus stand and at petrol pumps. This was further supported by metal loading in respective locations during these years. The maximum positive values were measured in proximity to the city area. Negative value at Mukteshwar forest reveals that it is a pollution free site. The effect of intensity of traffic compared to the influence of other factors like farming are much higher as the sampling point were nearer to the roads except the control site. Besides this, contamination was high and reduces with distance (Tuba et al. 1994).

In conclusion, the present research gives the supportive evidence of global warming in Himalaya which is in consequence to metal pollution in that area. Elemental concentration in *B. cellulare* was in order Zn > Pb ~ Cu > Cd in summer while, same was Zn > Cu > Pb > Cd in winter season, and Zn > Cu > Pb > Cd in rains indeed reflects atmospheric trace elemental load. Amongst seasons summer deployed maximum metal load followed by winter and monsoon. Present study describes that Nainital is most polluted for heavy metals (Pb, Zn, Cu and Cd) followed by Almora and Pithoragarh. Present study is very useful for policy makers to take necessary steps to take control measures in Himalayan belt, which directly reflect the weather condition of India.

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