Biomonitoring of Trace Element Air Pollution Using Mosses

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Abstract

This paper discusses biomonitoring using mosses as a technique used in programs for investigating trace-element air pollution. Emphasis is given to mosses as the dominant plant species used in biomonitoring surveys. Biomonitoring is regarded as a means to assess trace-element concentrations in aerosols. Current literature is reviewed for moss biomonitors and their physiological process responsible for accumulation of element concentration. Attention is given to the principles, selection of the moss species as biomonitors for air pollution and the possibilities for employing biomonitoring techniques in future studies. Combining the integrating property of moss biomonitors with availability and uniform matrix characteristics of air particulates as a prerequisite for monitoring of air pollution is discussed.

Keywords: Trace element; Biomonitor; Moss; Air pollution.

INTRODUCTION

Air quality research using biomonitors

Monitoring of air pollution using bioindicators is emerging as a potentially effective and more economical alternative performing by direct ambient air measurements. This is especially relevant for monitoring large areas (Rühling and Tyler, 1968). The usefulness of mosses in determining trace- and heavy-metal concentrations in different geographical areas has been discussed and demonstrated in several studies (Gjengedal and Steinnes, 1990; Markert *et al.*, 2003). Many European countries have used mosses since the beginning of 1960s in national and multinational surveys of atmospheric-metal deposition (Rühling, 1994). In practice, controlling anthropogenic air pollutants is a very complex problem where sources and emissions have to be managed and monitored, and economic aspects have to be integrated (Sloof, 1993). Most air

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pollution studies in India are based on atmospheric aerosols collected on particulate matter filters. This is an active method that gives an idea of trace-element atmospheric pollution only during the sampling time. It requires long-term sampling at a large number of sampling sites. The measurements require sophisticated technical equipment which is expensive.

There is lack of sufficiently sensitive and inexpensive techniques that permit the simultaneous measurement of many air contaminants (Puckett, 1988). In India, it is difficult to use air samplers in remote areas due to lack of electricity. Biomonitoring is the one and only solution. In this paper, we discuss the usefulness of biomonitoring as a technique used to investigate trace-element atmospheric pollution. It includes the principles, possibilities, and suggested strategies to develop such approaches.

The biomonitoring concept

Biomonitoring is defined generally as the use of bio-organisms to obtain information on certain characteristics of the biosphere. The organism used is called a bioindicator or biological monitor (Markert *et al*., 1997). There is considerable variation between the term bioindicator or biological monitor. Bioindicator generally refers to all organisms that provide information on the environment or the quality of environmental changes, and biomonitors are the organisms that provide quantitative information on the quality of the environment (Markert *et al.,* 2003). Thus, with proper selection of organisms, air-pollution monitoring is possible even in remote areas as samples can be collected and measurements of pollutants done at laboratories miles away from the site. Even though there might be deposition of pollutants on the surface, these can be taken care of by proper washing of samples before measurements. Biomonitoring is a passive method and provides a measure of integrated exposure over a period of time. The major advantages are:

a) No long term use of expensive sampling equipment is required.

b) Sampling of organisms used as biological monitor is generally easier.

c) The concentrations in the monitor organisms are higher than the system to be monitored. This improves the accuracy of measurements.

d) Most organisms reflect external conditions averaged over certain periods of time. This becomes important when monitoring levels change rapidly with time.

Selection criteria of the biomonitors

Theoretically, biomonitoring species for trace-element air pollution are selected on the basis of specificity (accumulation is considered to occur from the atmosphere only) (Rühling, 1994). In practice, to be a suitable biomonitor some specific requirements have to be met.

- The organism has to be common in the area of interest.

- It has to be available for sampling during all seasons; if not, then some simple special devices have to be developed to grow it in all seasons.
- The monitor should be tolerant of pollutants at the relevant levels.

Besides these necessary criteria, other additional requirements are:

- Element uptake should be independent of local conditions.
- The biological variation of the organism should be limited.
- The accumulated concentration levels must be measurable by routine analytical techniques.
- Absence of appreciable amounts of element uptake from sources other than the atmosphere.
- Physiological mechanisms for uptake of elements should be known to facilitate interpretation of the results.
- The biomonitor should average the elemental concentrations over a period of time as a result of integrated exposure.
- The organism should have low background concentrations of these elements.
- The sampling method and protocol for sample preparation for measurement should be simple and quick.

Suitable biomonitors, which meet the requirements, make continuous monitoring and even retrospective monitoring of air pollution possible at relatively low cost. When information on time-averaged trace-element concentrations at specific sites in the environment is the aim, the use of such non-mobile monitors is preferred.

Mosses as biomonitor

Mosses are cryptogams that thrive in a humid climate. Ectohydric mosses have been used as biomonitors, in most cases, of trace-element atmospheric pollution. They possess many properties that make them suitable biomonitors for air pollutants (Onianwa, 2001; Zeichmeister *et al.,* 2003a)

- They obtain nutrients from wet and dry deposition.
- They do not have real roots. So, they cannot take their nutrient from soil.
- Nutrient uptake from the atmosphere is promoted by their weakly developed cuticle.
- Large surface-to-weight ratio improves adsorption.

Other advantages are:

- Slow growth rate lets them accumulate pollutants over a larger time period.
- Undeveloped vascular bundles allow better adsorption than vascular plants.

- Minimal morphological changes during moss lifetime.
- Perenniality.
- Wide distribution.
- Ease of sampling.
- Possibility to determine concentrations in the annual growth segments.

Moss or lichen?

Mosses and lichens are the plants which readily accumulate air pollutants (Rühling and Tyler, 1970; Bargagli, 1998). The results obtained from the differences between mosses and lichens as accumulator indicators depend on: 1) the species used in the studies, and 2) the type of emissions and environment in the studied area.

The factors affecting the heavy-metal concentration in ground mosses and epiphytic lichens growing in the same place are canopy throughfall and stem flow. Throughfall can have an especially strong effect on element concentration in epiphytic lichens (Barkman, 1958). The effect of throughfall on ground mosses varies and depends on whether the moss is growing under or between the crowns.

In biomonitoring studies, moss samples are collected in open areas between the crown canopies in order to minimize the effect of throughfall. The amount of throughfall and stemflow varies according to the type of tree crown (Rasmussen, 1978). The crown canopy retains a part of the elements transported in free precipitation, but precipitation also leaches and washes off; e.g., nutrients (Ca, K, Mg, Mn) from the canopy, which subsequently are absorbed by stemflow by epiphytic lichens.

There is also variation in the element concentrations in mosses and lichens, especially in arid areas where precipitation is concentrated in the winter period. The differences between the concentrations in mosses and epiphytic lichens in areas in northern Europe may be due to the fact that the epiphytic lichens are exposed to air pollutants throughout the year, while mosses are protected by snow cover for almost half a year.

The different morphological and physiological properties of mosses and lichens account partly for the differences in metal-uptake efficiency. Since the surface structure of mosses is different than that of lichens, they have a larger surface-area-to-weight ratio. The surface of lichens in most cases is rougher and more porous than that of mosses.

Many studies, such as Finnish surveys, show that epiphytic lichens accumulate more heavy metals per dry weight than in mosses. Similar results have been reported in other studies (Kansanen and Venetvaara, 1991). The reason for the difference may be because of the variation in uptake efficiency in different deposition conditions, or due to the effect of throughfall on epiphytic lichens (Steinnes, 1993). Lichens also accumulate volatile heavy metals (Hg, Pb), which are continuously re-circulated back into the atmosphere more readily than in mosses (Evans and Hutchinson, 1996).

Many studies have shown that moss accumulates dust more easily than lichens (Steinnes, 1995). In Finland, the metal concentration in mosses were usually higher close to the emission sources, and lower in background areas, than the corresponding values in epiphytic lichens. It is also reported that the relative contribution of particulate material to the total concentration in mosses increases in places near emission sources, in arid areas and in agricultural areas. Especially in arid regions and those with sparse vegetation cover, metals that originate from the soil (Al, Cr, Fe, Ti) accumulate more readily in mosses than in lichens (Loppi and Bonini, 2000). However, there is no evidence of this phenomenon in the Finnish surveys, because the amount of dry deposition in Finland is relatively small (Berg *et al.,* 2002), and the soil normally has a fully developed vegetation cover that effectively prevents the dispersion of soil dust.

Mosses and lichens seem to depict wet deposition in different ways. Laboratory tests have shown that cation exchange is a very fast process in *Hylocomium splendens*, and that led to the conclusion that the concentration measured in this species of moss reflects the effects of the composition of rainwater prior to the sampling, rather than the effects of long-term accumulation (Brown and Brûmelis, 1996). Reimann *et al*. (1999) reported in their studies carried out in the Kola Peninsula in northwest Russia that the concentrations of many of the elements in mosses were more closely related to the chemical composition of rainwater than to the annual deposition levels as reflected by terricolous lichens (Bargagli, 1998).

No clear-cut evidence exists showing which could be a better heavy-metal biomonitor for regional surveys, because the results appear to vary from one place to another. However, the mechanisms through which mosses and lichens accumulate heavy metal are so different that they can not be used to replace each another in national surveys. In Finnish conditions, mosses appear to be more suitable for regional surveys than epiphytic lichens. The differences between different parts of the country and the location of emission sources were expressed more clearly on the mosses than on lichens. Wolterbeek *et al*. (1996) recommended the use of mosses because they more readily reflect local changes in heavy-metal deposition. However, lichens may be better accumulation indicators than mosses in arid conditions.

Biomonitor Behavior

Mechanism of entrapment of air particles and heavy metals

Air pollutants are deposited on mosses in three forms as aqueous solution, gaseous form or attached particles. The accumulation of pollutants in mosses occurs through a number of different mechanisms.

- As layers of particles.

- Entrapment on the surface of the cells.
- Incorporation into the outer wall of cells through ion exchange processes.
- Metabolically controlled passage into the cells (Brown and Bates, 1990).

The attachment of the particle is affected by the size of the particle and the surface structure of the mosses. Ion exchange is a fast physiological-chemical process that is affected by the number and type of free cation exchange sites, the age of the cells, their reaction to desiccation, growing condition, temperature, precipitation, pH, composition of the pollutants, and leaching (Tyler, 1990). In the ion exchange process, cations and anions become attached to the functional organic groups in the cell wall primarily through chelation (Rao, 1984).

The chemical composition of deposition has a large effect on the accumulation of pollutants, because the uptake efficiency of the mosses for individual elements varies considerably (Berg *et al*., 1995). The uptake efficiency of most common heavy metals follows mostly the order Pb > Co, Cr > Cu, Cd, Mo, Ni, V > Zn > As (Ziechmeister *et al*., 2003b). A high proportion of the pollutant load accumulates in mosses through wet deposition. The amount, duration and intensity of the precipitation affect accumulation and leaching (Berg *et al.,* 1995). The contribution of dry deposition increases on moving from humid to arid climates (Couto *et al*., 2004). There are considerable differences in the leaching of elements depending on whether they are bound to the cell wall, or accumulated on the surface of the mosses (Čeburnis and Valiulis, 1999). Uptake efficiency is also affected by competition for free cation exchange sites; for instance, the presence of the sea salts and acidic deposition has been found to have an effect on the absorption of metals by mosses (Gjengedal and Steinnes, 1990). The type of vegetation and soil dust have also have been reported to cause regional differences in uptake efficiency (Čeburnis *et al*., 1999). In general, the best correlation between the concentrations in mosses and in wet deposition has been found for elements that have a high uptake efficiency from wet deposition (e.g., Pb, Cd, Co, Cu) (Ross, 1990).

Factors affecting the concentrations of trace metals in moss

Epiphytic mosses may be considered for common use as biomonitor organisms. This is largely based on their lack of roots when compared with higher plants. Thus, they obtain their mineral supplies only from aerial sources and not from the substratum (Martin *et al.,* 1982).

In addition to air pollutants that originate from the anthropologic sources, the concentrations in mosses are affected by many "natural" factors associated with: 1) morphological and physiological properties of the mosses, and 2) the site where the mosses are growing and their immediate environment.

There are natural differences in chemical composition between individual species with different growths and conditions, and between separate parts of the individual moss. There are natural differences in chemical composition between individual species and even among populations of the same species, between individuals with different growth and conditions, and between the separate parts of the individual moss (Thöni *et al.,* 1996). Small amounts of nutrients may pass into the mosses from the substrate (Økland *et al.,* 1999) and nutrients can also be translocated from one part of the moss to another (Brŭmelis and Brown, 1997). Mineral particles originating from soil and bedrock also increase Fe, Cr, Al and Ti concentrations in areas which have a sparse vegetation, an arid climate, or exposed mineral soil (Mäkinen, 1994). Other factors affecting the concentrations include:

- Stand throughfall.
- Leaching from vegetation layers located above the mosses (Steinnes, 1993).
- The nutrient status of the site.
- Snowmelt water (Ford *et al*., 1995).
- Vegetation zone (DeCaritat *et al*., 2001).
- Altitude has an effect (Zechmeister, 1995; Gerdol *et al.,* 2002) due to changes in amount of precipitation, dust or biomass production.
- The sampling and measuring methods employed also have a considerable influence on the analytical results in biomonitoring studies (Markert and Weckert, 1989).
- Age of the moss. The finding that older moss parts have higher metal concentration has led to the assumption that the plants provide a historical and interactive recording of the metal supply in the environment.

Use of mosses as biomonitors

The use of mosses as biomonitors for atmospheric pollution has become common since suitable methods for sampling and analyzing them have been developed in Sweden (Tyler, 1970). Mosses have especially been used in regional heavy-metal surveys in Europe. The first extensive survey was carried out in Scandinavia in the end of 1960s, and then the first survey at a national level was carried out in Sweden, Norway and Denmark at the turn of the 1970s and 1980s (Steinnes, 1977) During the 1980s, the survey expanded to cover all over the Nordic countries, and in 1990s to most of the countries in Europe (Buse *et al*., 2003). The moss species were used to obtain information on the regional deposition of heavy metals, changes in the deposition patterns, the long-distance spread of emissions, and local emission sources.

A considerable number of other regional studies on heavy metal and other element concentrations have been carried out using mosses, primarily in North America (Pott and Turpin, 1996). In addition to national surveys, several regional surveys have also been carried out on other factors affecting concentration (Zeichmeister, 1998), including mosses as deposition accumulators in relation to other biomonitors, as well as absolute deposition values (Sucharová and Suchara, 2004). In Wisconsin USA moss-bag technique was used to monitor heavy metal, sulfur and nitrogen using mesh bags containing *Sphagnum russowii* (Makholm and Mladenoff, 2005). Similar studies were done in Romania, Russia and Bulgaria using *Sphagnum girgensohnii*; 36 elements were investigated (Culicov *et al.,* 2005).

In India, initial studies are being conducted to monitor trace element pollution using mosses as biomonitors (Chakrabortty *et al.,* 2004; Chakrabortty *et al.*, 2006). India has lot of geographic variation sustaining a wide variety of flora. Studies conducted in and around Mumbai, a major metropolis in western India, have identified *Pinnatella alopccuroides, Pterobryopsis flexiceps* and Bryum spp. as effective biomonitors for the region.

Perspectives

The relative ease of sampling, the absence of any need for complicated and expensive technical equipment, and the accumulative and time-integrative behavior of the moss biomonitor give biomonitoring of atmospheric trace elements advantages for their continue practice in the future, especially in larger-scaled surveys. Biomonitoring is a very important method for source identification.

One of the most important challenges in biomonitoring studies is to establish standardized protocol for sampling, sample preparation, elemental analysis in order to obtain comparable results on at least a regional scale. It is possible to collect mosses in selected areas ranging from pollution-free background regions to highly polluted regions. By collecting mosses in premonsoon, monsoon and post-monsoon seasons, it is possible to know the specific trace-element pollution area. One can also predict the suitable moss species that may be used as a biomonitor for a single trace element, or a group of trace elements. It is very difficult for any other approach to obtain such a detailed picture of variations in time and space at a reasonable cost. This can even be done using moss and lichen transplants (Giordano *et al.*, 2005).

Moreover the particular spatial structure of the pollution sources interacts with the spatial layout of the samples, resulting in data sets with distributions that are very different from the usually assumed normal distribution. The first step in analysis of this kind of data must be to check for the presence of spatial structure on scales larger than the sampling grid, to avoid mapping noise. Then the map generated must not contain information about pollution sources with a spatial scale smaller than the spatial scale of the sampling grid (Aboal *et al.,* 2006). The data can be aggregated by cluster analytical approach, which allows for an investigation of the emission structures that remain the same over time, or by percentile statistics that illustrate both spatial and temporal trends of metal accumulation (Pesch and Schroeder, 2006).

A detailed discussion on the feasibility of different analytical techniques to quantify the trace-element concentration is beyond the scope of this paper. Many multielemental techniques, such as EDXRF, INAA, and ICP-MS, can be used for determination of trace and toxic elements. Development of multidisciplinary programs that would collate data regarding trace element atmospheric pollution can make it possible to draw ecosystem-level models depicting the dispersal and effects of air pollutants.

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