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Mosses as Bioindicators of Heavy Metal Air Pollution in the Lockdown Period Adopted to Cope with the COVID-19 Pandemic

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Abstract: The coronavirus disease, COVID-19, has had a great negative impact on human health and economies all over the world. To prevent the spread of infection in many countries, including the Russian Federation, public life was restricted. To assess the impact of the taken actions on air quality in the Moscow region, in June 2020, mosses Pleurozium shruberti were collected at 19 sites considered as polluted in the territory of the region based on the results of the previous moss surveys. The content of Cd, Cr, Cu, Fe, Ni, and Pb in the moss samples was determined using atomic absorption spectrometry. The obtained values were compared with the data from the moss survey performed in June 2019 at the same sampling sites. Compared to 2019 data, the Cd content in moss samples decreased by 2–46%, while the iron content increased by 3–127%. The content of Cu, Ni, and Pb in mosses decreased at most sampling sites, except for the eastern part of the Moscow region, where a considerable number of engineering and metal processing plants operate. The stay-at-home order issued in the Moscow region resulted in a reduction of vehicle emissions affecting air quality, while the negative impact of the industrial sector remained at the level of 2019 or even increased.

Keywords: COVID-19; air pollution; metals; industry; moss survey; biomonitoring

1. Introduction

On March 11, 2020, the World Health Organization declared COVID-19 as a global pandemic [1]. On March 12, based on a decree of the Government of the Moscow region, a self-isolation regime was introduced. According to the official decision, only essential services such as healthcare, logistics, food supply, public transport, and industrial enterprises due to technical reasons did not cease to operate. A significant part of the regular national and international flights and train services was cancelled. Since the majority of the population switched to remote working, the number of vehicles has dropped significantly.

New rules adopted in many countries to help slow the spread of COVID-19 resulted in a decrease in the negative impact on the environment in some regions of the world [2]. Nadzir et al. [3] measured the concentrations of CO, PM2.5, and PM10 in Malaysia and observed that the concentration of pollutants declined significantly, by ≈20 to 60%, during the Control Order in Malaysia (MCO) days at most studied locations. At the same time, in Kota Damansara, the level of pollutants significantly increased due to local anthropogenic activity. COVID-19 resulted in a worldwide decrease in the concentration of CO2.
in March 2020 by 7% in comparison with the monthly concentration in 2019 [4]. In three Chinese cities (Chongqing, Luzhou, and Chengdu) concentrations of PM$_{2.5}$, PM$_{10}$, SO$_2$, CO, and NO$_2$ in February 2020 were lower by 17.9%–62.1% than the values determined in February 2017–2019 [5]. In three other cities in China (Wuhan, Jingmen, and Enshi) concentrations of the abovementioned pollutants measured in February 2020 declined by 27.9%–61.4% compared to February 2017–2019. At the same time, in the period of January–March 2020, an increase of O$_3$ concentration of up to 15% was noticed [6]. In Turkey, the concentrations of PM$_{10}$ and NO$_2$ in 2016 and 2020 were compared, and no significant difference in PM$_{10}$ concentrations was observed. In the after-lockdown period, the NO$_2$ concentrations were lowered by 11.8% [7]. A decrease in the PM$_{2.5}$ concentration in Wuhan, Daegu, and Tokyo by 29.9%, 20.9%, and 3.6%, respectively, took place after one month of COVID-related restrictions. The concentration of NO$_2$ also declined in all three cities, with the most pronounced decrease being by 53.2% in Wuhan [2]. In Brazil, a remarkable decrease in CO and NO$_2$ concentrations was observed during the lockdown period [8].

PM contains various metals, including some elements recognized as human carcinogens. Sources of metals in the atmosphere can be natural: soil dust, volcano eruptions, and forest fires, or they can be anthropogenic, which include road traffic, industry, and thermal power plants [9]. It is often difficult to conduct measurements of concentrations of atmospheric metals over large territories. Passive moss biomonitoring proved to be a suitable technique to study the spatial distribution of metals over the large territories [10]. This technique is well recognized and widely applied in many European countries [11]. Although data obtained using moss biomonitors do not correspond to the direct quantitative measurement of metal deposition [12], this information is very useful for regions where it is difficult to obtain official data related to air pollution. In the Moscow region, moss survey studies were performed in 2004 [13], 2014 [14], and 2019 (data not yet published).

The aim of the present study was to find out whether moss can be used as a tool to assess the impact of the restrictions due to the lockdown imposed to face the pandemic emergency on air quality in a relatively short time (2.5 mounts). For this purpose, the data obtained for moss samples collected in the Moscow region in June 2020 were compared with the data obtained in 2019 for the same collection sites.

2. Experiments

2.1. Study Area and Sampling

The Moscow region, which is a subject of the Russian Federation, is one of the most densely populated and industrially developed regions of the country [14]. The Moscow region can be considered the economic core of the country, with about 70% of Russia’s financial wealth concentrated here. It is located in the center of railway and air networks, reaching Transcaucasia, Central Asia, and the Pacific, and it is connected by rivers and canals to the Baltic, White, Azov, Black, and Caspian Seas [15]. In the center of the region is Moscow, the capital of the Russian Federation, and the most important industrial city, transportation hub, educational and cultural center of the region [13,15]. The industrial sector of the Moscow region includes metallurgical, oil refining, mechanical engineering, food, energy, and chemical enterprises [14]. The main industrial centers in the Moscow region are: Krasnogorsk, Lyubertsy, Mytishchi, Klin, Noginsk, Pavlovsky Posad, Voskresensk, Kolomna, Dmitrov, Klin, Elektrostal, Balashikha, and Sergiyev Posad.

In 2019 and 2020, moss Pleurozium schreberi was collected on the sheared tree bark. Moss sampling was performed in June 2019 and June 2020 at 19 sampling sites (Figure 1) in accordance with [16]. The selection of the sampling sites in 2020 was based on the moss survey data obtained in 2019 (analyzed but not yet published). Moss samples were collected at sites with the highest metal concentrations in 2019. After collection, samples were cleaned of soil particles and other contaminants. The upper 3–4 cm of green and green-brown shoots from the top of the moss were separated and dried at 105 °C to constant weight. For analysis, approximately 0.2 g of moss was placed in a Teflon vessel and treated
with 2 mL of concentrated nitric acid and 1 mL of hydrogen peroxide. The Teflon vessels were put into a microwave digestion system (Mars; CEM, USA) for complete digestion. Digestion was performed in two steps: (1) ramp: temperature 180 °C, time 15 min, power 400 W, and pressure 20 bar; (2) hold: temperature 160 °C, time 10 min, power 400 W, and pressure 20 bar. Digests were quantitatively transferred to 100-mL calibrated flasks and made up to the volume with bidistilled water.

Figure 1. Sampling map (in 2019 and 2020 samples were collected in the same sampling sites).

2.2. Chemical Analysis

The content of Cd, Cu, Pb, Cr, Ni, Fe, V, and Sb in the moss samples was determined by means of atomic absorption spectrometry using a Thermo Scientific™ iCE™ 3400 AA spectrometer (Thermo Scientific, Waltham, MA, USA) with electrothermal (graphite furnace) atomization. Stock solutions (AAS standard solution; Merck, Germany) with metal concentrations of 1 g/L were used to prepare calibration solutions. The National Institute of Standards and Technology (NIST) reference materials 1570a (Trace Elements in Spinach Leaves) and 1575a (Pine Needles) were used to ensure the quality control of measurements. The difference between determined and certified values did not exceed 5%.

2.3. Data Processing

Statistical analysis of the data was done using Excel 2016 (Microsoft, Redmond, Washington, USA) and IBM SPSS software (IBM, Armonk, New York, USA). The Wilcoxon signed-rank test [17] was applied to define differences between the values obtained in 2019 and 2020. The ArcGis 10.6 software (Esri, Redlands, California, USA) was used to build maps showing the spatial distribution of elements.

To quantify the anthropogenic influence on the environment, several indices were calculated, such as the contamination factor (CF), the Geo-accumulation Index (Igeo), and the pollution load index (PLI).

The contamination factor CF is defined as:

\[
CF = \frac{C_m}{C_b}
\]  

(1)
where $C_m$ is the measured content of the metal at any given site and $C_b$ is the background level for that metal [18].

CF < 1 no contamination; 1–2 suspected; 2–3.5 slight; 3.5–8 moderate; 8–27 severe; and >27 extreme [19].

The index of geo-accumulation, $I_{geo}$, was calculated using the following formula:

$$ CF = \frac{C_m}{C_b^{1.5}} $$

where $C_F$ is the contamination factor. The factor of 1.5 is introduced to minimize the effect of possible variations in the background [20].

$I_{geo}$ <0 no contamination; 0–1 slightly polluted; 1–2 moderately polluted; 2–3 moderately to severely polluted; 3–4 severely polluted; 4–5 severely to extremely polluted; and $I_{geo}$ > 5 extremely polluted [21].

The PLI represents the nth order geometric mean of the entire set of CF regarding the contaminating elements as follows:

$$ PLI = \sqrt[n]{\prod_{i=1}^{n} CF_i} $$

where $n$ is the total number of contaminating elements.

PLI < 1 (non polluted); 1 ≤ PLI < 2 (slight polluted); 2 ≤ PLI < 3 (moderately polluted); PLI < 3 (highly polluted) [22].

3. Results and Discussion

Eight elements were determined in the analyzed moss samples using atomic absorption spectrometry (AAS). Since the concentrations of V and Sb were below the detection limits, these elements were excluded from further discussion. The results of the statistical analysis for the analyzed elements in 2019 and 2020 are presented in Table 1.

Table 1. Descriptive statistics of results for moss samples collected in 2019 and 2020 (in mg/kg dry weight (d.w.)).

| Element | Year | Range     | Md     | Mean ± St. Dev | Q1   | Q3   | CV (%) | p         |
|---------|------|-----------|--------|----------------|------|------|--------|-----------|
| Cd      | 2019 | 0.11–0.64 | 0.30   | 0.34 ± 0.14    | 0.23 | 0.41 | 41.9   | <0.05     |
|         | 2020 | 0.14–0.52 | 0.24   | 0.26 ± 0.09    | 0.22 | 0.30 | 33.9   |           |
| Pb      | 2019 | 1.71–17.2 | 4.41   | 5.62 ± 3.45    | 3.80 | 7.02 | 61.4   | >0.05     |
|         | 2020 | 1.79–13.6 | 4.60   | 5.31 ± 3.14    | 3.21 | 6.26 | 59.2   |           |
| Cu      | 2019 | 6.38–21   | 8.98   | 10.3 ± 3.93    | 7.29 | 12.9 | 38.3   | <0.05     |
|         | 2020 | 4.72–15.8 | 7.76   | 8.8 ± 3.18     | 6.64 | 9.36 | 36.3   |           |
| Cr      | 2019 | 1.1–3.09  | 1.98   | 1.87 ± 0.58    | 1.32 | 2.16 | 30.8   | >0.05     |
|         | 2020 | 1.01–4.29 | 1.98   | 2 ± 0.75       | 1.46 | 2.18 | 37.6   |           |
| Ni      | 2019 | 1.53–5.86 | 2.91   | 3.36 ± 1.17    | 2.67 | 4.27 | 34.9   | <0.05     |
|         | 2020 | 2.59–7.35 | 3.85   | 4.27 ± 1.27    | 3.37 | 4.61 | 29.8   |           |
| Fe      | 2019 | 343–1175  | 579    | 607 ± 237      | 446  | 682  | 39.1   | <0.05     |
|         | 2020 | 309–2551  | 693    | 846 ± 476      | 594  | 933  | 56.2   |           |

Md: median; P90–90 percentile; St. Dev.: standard deviation; CV: coefficient of variance, $p$-values for differences were obtained from Wilcoxon signed-rank test.

According to the Wilcoxon test, no significant differences ($p > 0.05$) were found for Pb and Cr, while for Cu, Cd, Fe, and Ni, significant differences ($p < 0.05$) between the median concentrations were revealed. The CV values for all elements in both years were less than 75%, which points to the main influence of the regional source of pollution [14].
The median values determined in the present study were compared with the data obtained for the previous moss surveys performed in the Moscow region in 2004 and 2014 (Table 2). The content of Cd, Cu, and Ni in moss samples was almost at the same level in the period 2004–2020. The higher values of the mean Pb content in 2019–2020 in comparison with 2014 can be explained by the fact that in 2019–2020 and 2014, the moss samples were collected at different sampling sites: in 2014—in the north-eastern part of Moscow, whereas in 2019–2020—in places considered to be potentially highly polluted. The iron content was the highest in 2014, followed by 2004, 2020, and 2019, while the content of Cr in mosses collected in 2019 and 2020 was significantly lower than in 2014 and 2004.

Table 2. Comparison between the median values obtained in the present study and data reported for previous surveys (in mg/kg d.w.).

| Element | Moscow Region 2020 | Moscow Region 2019 | Moscow Region 2014 [14] | Moscow Region 2004 [13] |
|---------|--------------------|--------------------|-------------------------|-------------------------|
| Cd      | 0.24               | 0.30               | 0.3                     | x                       |
| Pb      | 4.60               | 4.41               | 0.67                    | x                       |
| Cu      | 7.76               | 8.98               | 7.1                     | x                       |
| Cr      | 1.98               | 1.98               | 3.2                     | 3.1                     |
| Ni      | 3.85               | 2.91               | 3.2                     | 2.4                     |
| Fe      | 693                | 579                | 1050                    | 800                     |

In order to reveal the differences in metal uptake by moss in 2019 and 2020, the element distribution maps of are given in Figure 2. Natural sources of Cd emissions are volcanic activity and release by vegetation [23]. The main anthropogenic sources are non-ferrous metal production, waste incineration, dust generated during the operation of vehicles, and resuspension of road dust [23,24]. Hjortenkrans et al. [25] showed that emissions from brake linings/tire tread rubber contain a wide range of elements, including Cd, Cu, Pb, Sb, and Zn. Comparing the data obtained in 1998 and 2005, the authors showed that Cu and Zn emissions remained unchanged in the studied period, suggesting that brake lining is one of the main sources of these metals. On the other hand, a pronounced decrease in the content of Pb and Cd in the studied period was observed. The input of anthropogenic sources of Cd emission significantly exceeded the release from natural resources. According to the data obtained in 2020, the content of Cd in the Moscow region diminished by 2–46%. The most pronounced decrease was noticed near the cities of Sergeyev Posad, Solnechnogorsk, and Domodedovo. According to the reports by the national authorities, during the period of self-isolation, traffic in the Moscow region declined by 50%.

Figure 2. Cont.
The increase of Cd concentrations near Klin and Stupino can be explained by the impact of metallurgical and engineering complexes.

The major sources of Cr in the environment are metal processing, coal burning, and vehicles. Chromium is one of the most abundant metals in diesel particles [26]. Compared to 2019, in 2020, the content of Cr in moss samples decreased by 7–35%, mainly in the north-eastern part of the Moscow region. In Sergeyev Posad, the increase in Cr content by 59% may be related to the activities of an engineering plant and the production of paint and coatings. The rise of Cr content in satellite cities near Moscow by 42–100% may be associated with the industrial activity of machine-building, metallurgical, and chemical plants, which were operating at full capacity during the self-isolation period.

Brake wear emissions account for up to 75% of Cu emissions into the air [27]. A decrease in the Cu content in moss samples collected in 2020 by 8–50% compared to the data for 2019 confirms this hypothesis. At the same time, an increase in its content by 38% in satellite cities near Moscow points to the dominant contribution of industrial activity to Cu emissions.

The main sources of Fe in the atmosphere are industrial and metallurgical processes, combustion of fossil fuels, transport, as well as resuspension of crustal materials and road dust [28]. In contrast to the previously discussed element, the content of Fe in the moss samples collected in 2020 increased significantly in comparison with the data for 2019. This increase of 3–217% can be attributed mainly to the resuspension of crustal materials.

Figure 2. Maps of element content in moss samples collected in 2019 and 2020 in the Moscow region (in mg/kg d.w.).
significantly in comparison with the data for 2019. This increase of 3–217% can be attributed mainly to the resuspension of crustal materials.

Nickel can be released into the atmosphere from natural and anthropogenic sources. The main anthropogenic sources are fossil fuel combustion, smelting of ferrous and non-ferrous metals, waste incineration, and other various sources [29]. The increase in Ni content in satellite cities around Moscow in 2020 may be associated with industrial activities of metallurgical plants in Electrostal, Shchyolkovo, and Podolsk, and engineering plants in Electrostal and Podolsk. The rise of Ni content at other sampling sites is mainly due to vehicles and resuspension of dust particles. In spite of the fact that Pb content has been declining in many countries due to the introduction of unleaded fuels, vehicles continue to be one of the main sources of Pb emissions. On a regional scale, industrial activities may contribute to emissions of lead into the air [30]. A significant decrease in Pb content was noticed in 2020 in comparison with 2019 (up to 48%), indicating a decline in traffic flow. A considerable increase in Pb content by 30–65% in the western part of the Moscow region indicates the dominant role of industrial activity in Pb emission in this area.

The strength of association of the chemical elements in the moss samples collected in 2019 and 2020 can be seen in Table 3. The Spearman correlation coefficient between 0.5 and 0.7 indicated a good association between the elements, whereas r in the range of 0.7–1.0 shows strong association of elements [31].

Table 3. Spearman correlation coefficient between element content in mosses collected in Moscow region in 2019 and 2020.

|       | Cd   | Pb   | Cu   | Cr   | Ni   | Fe   |
|-------|------|------|------|------|------|------|
| 2019  |      |      |      |      |      |      |
| Cd    | 1.00 |      |      |      |      |      |
| Pb    | 0.76 ** | 1.00 |      |      |      |      |
| Cu    | 0.25 | 0.38 | 1.00 |      |      |      |
| Cr    | 0.43 | 0.60 ** | 0.71 ** | 1.00 |      |      |
| Ni    | 0.28 | 0.14 | 0.28 | 0.55 * | 1.00 |      |
| Fe    | 0.56 * | 0.48 * | 0.46 * | 0.66 ** | 0.36 | 1.00 |
| 2020  |      |      |      |      |      |      |
| Cd    | 1.00 |      |      |      |      |      |
| Pb    | 0.55 * | 1.00 |      |      |      |      |
| Cu    | 0.36 | 0.74 ** | 1.00 |      |      |      |
| Cr    | 0.41 | 0.67 ** | 0.66 ** | 1.00 |      |      |
| Ni    | 0.42 | 0.39 | 0.61 ** | 0.73 ** | 1.00 |      |
| Fe    | 0.22 | 0.69 ** | 0.61 ** | 0.71 ** | 0.40 | 1.00 |

* Correlation is significant at the 0.05 level (2-tailed). ** Correlation is significant at the 0.01 level (2-tailed).

In 2019, a high positive correlation in Pb-Cd and Cr-Cu pairs was observed. Road traffic may be responsible for Cr, Cu, Pb, V, and Zn emissions, whereas fuel combustion is the major source of Cr, Cu, and V [29]. Good positive correlations were found between Fe-Cd, Cr-Pb, Ni-Cr, and Fe-Cr pairs of elements, and this may be related to industrial activities. In 2020, a high correlation between Pb and Cd was observed again. A high positive correlation was also determined in the Cu-Pb, Ni-Cr, and Fe-Cr pairs. Good correlations were obtained between the Cr-Pb, Fe-Pb, Ni-Cu, and Fe-Cu pairs of elements. The obtained associations indicate a possible anthropogenic influence in the study area; however, their source could be a resuspension of soil particles.

The use of geochemical indices is important for the assessment of the contamination status of the investigated territory [32]. In the present study, the contamination factor (CF), Geo-accumulation Index ($I_{\text{geo}}$), and pollution load index (PLI) were calculated [14,32] (Table 4). In 2019, no contamination with Ni and moderate contamination with Cr, Fe, and Cu were determined. The CF values for Cd and Pb were higher than 3.0 and indicated considerable contamination. The situation changed in 2020 when CF pointed to moderate pollution for all elements. The decrease of CF values for Pb and Cd may
be associated with the reduction of traffic flow, while the impact of the industrial activity remained at the same level. The I_{geo} results indicate that mosses were uncontaminated to moderately contaminated with the determined elements. In 2019, the moss samples were moderately contaminated with Cd, Pb, Cu, and Fe, and in 2020, with Cd and Pb.

Table 4. Mean values of the contamination factor CF and Geo-accumulation Index I_{geo} for the studied area.

| Element | Year | CF 2019 | CF 2020 | I_{geo} 2019 | I_{geo} 2020 |
|---------|------|---------|---------|--------------|--------------|
| Cd      | 2019 | 3.06    | 1.85    | 0.91         | 0.24         |
| Pb      | 2019 | 3.28    | 2.96    | 0.93         | 0.77         |
| Cu      | 2019 | 1.61    | 1.49    | -0.09        |              |
| Cr      | 2019 | 1.39    | 1.58    | -0.17        | -0.02        |
| Ni      | 2019 | 0.92    | 1.30    | -0.79        | -0.26        |
| Fe      | 2019 | 1.74    | 1.48    | 0.12         | -0.17        |

A PLI below 1.0 shows that elemental loads are approximately equal to the background level, and values above 1.0 indicate the degree of pollution [33]. As can be seen from the maps presented in Figure 3, in 2019, the entire territory under investigation belongs to the moderately polluted to unpolluted category (except for the sites near Troitsk and Domodedovo, which are moderately polluted). Similarly, in 2020, the sampling territory can be characterized as moderately polluted to unpolluted. The PLI values were higher than 2.0 near Domodedovo and Staraya Kupavna.

![Figure 3. The map of the distribution of PLI values in Moscow region.](image)

4. Conclusions

The results from two biomonitoring studies performed in the Moscow region in 2019 and 2020 were compared. Moss sampling has proven to be a suitable and low-cost indicator of heavy metal air pollution. The self-isolation period adopted to cope with the COVID-19 pandemic resulted in a decrease in Cd content in the Moscow region, while the content of other analyzed elements decreased in the north-eastern part of the Moscow region and remained the same or even increased in satellite cities near Moscow. Owing to the decline in the flow of traffic, stationary sources can be considered the primary source of metal emission into the atmosphere. In 2019 and 2020, according to the PLI values, the territory of the Moscow region was characterized as moderately polluted to unpolluted.
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