Detection of air quality improvement within a suburban district (southern Italy) by means of lichen biomonitoring

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ABSTRACT

The present work compared both the bioaccumulation of trace elements and the values of ecophysiological parameters measured in thalli of the lichen Pseudevernia furfuracea (L.) Zopf in two monitoring campaigns performed before and after improvement measures put in place by a 15 MW biomass power plant (BPP): the activation of a concentrated solar thermodynamic plant and the increasing percentage of exhausted olive pomace used as fuel. The cases of no enrichment and moderate enrichment change from 49 and 17% in 2013 to 68 and 4.2% in 2019, respectively. Several metals in 2019, show a Delta (difference between exposed and not exposed lichen thalli concentration) that is significantly lower than in 2013. The spatial pattern of contamination is comparable between the two years. However, the BPP affects the spatial variation of Ti, Al, V and Co in both 2019 and 2013, but only in the latter year also that of Cu, Cr and As which, in some monitoring sites, developed extremely high levels of enrichment. Traffic, whose rate increased over time, constantly influences the bioaccumulation of Cu, Sb and Mo. In 2019, the lichen oxidative stress is significantly reduced as well as the number of correlations between malondialdehyde levels and those of trace elements. Pigment values never differ (p > 0.05) from pre-exposure levels. Our results suggest that the development of hybrid plants, as well as a better fuel selection can reduce the environmental impact due to the combustion of biomass contributing to make this type of energy source more sustainable.

1. Introduction

Electricity production from combustion processes is still one of the main sources of air pollution, especially when it comes from coal- or heavy-oil-fired power plants (Gune et al., 2019; Wang et al., 2010; Zhang et al., 2020). The persistence of several harmful substances in the atmosphere, as well as the contamination of water and soil, poses a threat to the environment and exposes human populations to severe risks due to the development of multiple diseases (Khilare et al., 2012; Li et al., 2019), especially in the early age groups (Amster and Lew Levy, 2019). This issue has reached overwhelming dimensions. In a recent study based on the application of the GEOS-Chem chemical transport model (Vohra et al., 2021), air pollution due to fossil fuels was estimated to have caused 8.7 million premature deaths worldwide in the year 2018 alone. Several strategies can be implemented to reduce emissions related to this industrial sector. First of all, the use of abatement systems for coarse (cyclone systems) and fine/ultrafine (fabric filter and electrostatic precipitator) particulate matter levels, which have an efficiency ranging from 94 to 99.9% (TFTEI, 2020). On the other hand, combined heat and power plants, where heat generated in combustion processes is reused for thermal purposes or to generate electricity (EPA, 2014), result in a reduction, on average, of around 480 g/kWh of CO₂, 1.8 g/kWh of NOₓ and 8.8 g/kWh of SO₂ (OGL, 2021). This leads to significant savings in production costs as well. The implementation of renewable energies seems to be the best practice to obtain the maximum benefits for air quality (National Academic of Sciences, 2010). In this perspective, it is worth mentioning the development of many types of hybrid power plants where the combustion of natural gas is flanked by photovoltaic solar panel systems or wind turbines with the ability to store the electricity produced in batteries and release it into the grid in accordance with the energy demands (Wind Europe, 2019; Gorman et al., 2020). Biomass is also counted as a renewable source. This definition turns out to be erroneous since from an application of life cycle assessment it is evident that the CO₂ footprint associated with them is not neutral, but rather positive (Varun et al., 2009; Yi et al., 2018). In addition, emissions from such power plants may be even more polluting than those...
2. Materials and methods

2.1. Study area

The investigated territorial district, the urban-industrial area of Rende (Calabria Region), has an extent of 25 km² and includes a large urban area, with a total population of 24,000 inhabitants, and several industrial activities. The main one is a biomass power plant with a power of 15 MWh, currently fuelled by wood (60%) and exhausted pomace (40%). Since 2014, a concentrated solar thermodynamic plant has also contributed to energy production. Horticulture and arboriculture prevail on the northern side of the study area. The road network includes segments of the A2 motorway and a few provincial (SP241 and SP234) and state (SS107) roads. From the geological point of view, the study area is characterized by a succession of pleiocenic sediments such as light brown and red sands and gravels, blue gray silty clays, and silt interlayers (Dell‘Anna et al., 1981), and a paleozoic intrusive-metamorphic complex (Crittelli et al., 1990; Le Pera and Sorriso-Valvo, 2000; Le Pera et al., 2001). The investigated area has an altitude between 143 and 315 m a.s.l., while the average values of the main yearly weather parameters are: rainfall of 1247 mm, temperature of 16.4 °C and relative humidity of 65%.

2.2. Lichen transplants

Due to the scarce lichen colonization in the study area (Fig. 1), biomonitoring was carried out by means of transplantation technique. Thalli of the epiphytic lichen Pseudevernia furfuracea (L.) Zopf transplanted in an urban-industrial district. In this area, the main sources of pollutants are a 15 MWh biomass power plant and vehicular traffic (Lucadamo et al., 2016). In 2013, diffuse contamination due primarily to Ti, Al and Mn and secondarily to Co and V was highlighted, as well as some criticalities in terms of several peaks in bioaccumulation of Cu, Cr and As and high levels of oxidative stress. In 2019, as the biomass power plant has undertaken a series of actions aimed at reducing its environmental impact (installation of a concentrated thermodynamic photovoltaic system of 1 MWh which integrates replaces the energy produced by the combustion of biomass and an increase in the use of exhausted olive pomace, compared to woody material, as fuel), the same parameters measured previously were again monitored to verify: a) the persistence or not of the contamination phenomena detected in 2013, b) the temporal trend of the trace element bioaccumulation spatial pattern, c) the different contribution to this pattern by the main local anthropogenic sources, d) the physiological status of lichen transplants at the end of the exposure period and its association with the spatial variation of trace elements concentration.

2.3. Local scale wind monitoring

In order to use the wind through quantitative relationships to track the contribution of trace element sources in the study area, a meteorological station (Vantage Vue, Davis Instruments) was placed as close as possible to the biomass power plant at a controlled site (S13: CREA-OLI, Research Centre for Olive and Oil Industry) to protect the instrumentation from possible vandalism, and with a location that was not influenced by potential disturbances due to adjacent buildings. Wind speed and direction were measured and recorded every 15 min, resulting in a final database of 8640 records which were then used for further statistical processing.

2.4. Analysis of contaminants

2.4.1. Trace elements

Trace element determination was performed using 3 sub-samples per site, including lichen origin area. Aliquots of 100 mg of lichen thallus were pulsed by immersion in liquid nitrogen and macerated with a ceramic mortar and pestle. Subsequently, the samples were mineralised using 12 mL of ultrapure nitric acid in a microwave oven (Milestone Ethos 900) and Al, Ti, V, Cr, Mn, Co, Ni, Cu, Zn, As, Mo, Cd, Sn, Sb were measured for each sample by inductively coupled plasma mass spectrometry (Elan DRC PerkinElmer SCIEX). The accuracy of the analysis was evaluated by certified reference material BCR482 (Lichen Pseudevernia furfuracea (L.) Zopf). The analysed elements were selected because of their representation in the atmospheric emissions from the main anthropogenic sources of the study area, such as Mn, Cu, Sb, Mo typically associated to traffic emissions, and Ti, Al, Co, Cu, Mn, and V prevalently related to biomass combustion (Eastern Research Group, 2001; Demirbas, 2005; Thy et al., 2008), while Co, Cr, Fe, Mg, As and V are of geogenic origin considering the geological characteristics of our study area (Guagliardi et al., 2012).

2.5. Ecophysiological parameters

2.5.1. TBArs test

Total peroxidation levels were assessed according to the procedure of Huang et al. (2004). Firstly, 50 mg of lichen thallus are washed with distilled water and, after drying, homogenised in 2.5 mL of 0.1% trichloroacetic acid using a T25 digital ultra-turrax (Ika, Staufen, Germany). After centrifugation at 12,000g for 20 min, 0.5 mL of the
supernatant is transferred to a glass tube containing a solution of 0.6% 2-thiobarbituric acid (TBA) and 10% trichloroacetic acid and incubated at 95 °C for 30 min. After that time, the samples are allowed to cool and then centrifuged again at 12,000 g for 10 min. Peroxidation levels are quantified measuring the supernatant absorbance at 532 nm using the molar extinction coefficient of the TBA-MDA (malondialdehyde) adduct (155 mM⁻¹ cm⁻¹) by means of a PerkinElmer λ-40 spectrophotometer. The results are expressed as μMolg⁻¹ dry weight.

2.5.2. Lichen vitality

Cell respiration was determined by the reduction of triphenyl tetrazolium chloride to triphenyl formazan due to dehydrogenase activity (Backor and Fahselt, 2005). Aliquots of the thalli (15 mg) are incubated for 20 h at 15 °C in 2 mL of 50 mM phosphate buffer (pH 6.8) containing 0.6% triphenyl tetrazolium chloride and 0.005% Triton X100. After washing the thalli with distilled water, a second incubation at 65 °C for 2 h in 2 mL of dimethyl sulfoxide follows. The triphenyl formazan formed is then extracted using n-hexane and vortexed for 1 min. Finally, centrifugation at 4000 rpm for 15 min is carried out. The supernatant is read at 492 nm to assess triphenyl formazan levels using a PerkinElmer λ-40 spectrophotometer.

2.5.3. Photosynthetic pigments

Prior to analysis, the thalli are repeatedly treated with an acetone solution saturated with calcium carbonate to remove lichenic acids that could cause pigment denaturation. Each sample (60 mg of thallus) is homogenised in 3 mL of dimethyl sulfoxide and 3 mg of polyvinylpyrrolidone and then incubated at dark for 18 h in a final volume of 7 mL of dimethyl sulfoxide. In order to make pigment extraction more efficient, centrifugation is performed and the pellet is resuspended with 3 mL of dimethyl sulfoxide and incubated a second time in the dark for 6 h. Both aliquots are then combined, centrifuged and subjected to spectrophotometric analysis (PerkinElmer λ-40) at wavelengths of 665, 649 and 480 nm. The concentrations of chlorophyll a, chlorophyll b, and xanthophylls + carotenoids were assessed by applying the equations of Wellburn (1994).

2.5.4. Photosynthetic efficiency

The quantum potential product is a measure of the conversion of photosynthetic energy into chemical energy at the level of the photosystem II. Lichen thalli are hydrated for 1 h at room temperature. Afterwards, the lacinae are initially kept in the dark by means of leaf clips for 10 min and then subjected to a saturating light beam (2400 μmol s⁻¹ m⁻²) using the Handy PEA fluorimeter (Hansatech Instruments Ltd). The photosystem II efficiency is expressed as Fv/Fm ratio (Ronen and Galun, 1984).

2.6. Statistical analysis

The comparison of bioaccumulation levels between the two different years was performed in two ways. First of all, we converted the concentrations measured in the exposed lichen thalli into enrichment levels using two scales for interpreting metal concentrations in lichens in terms of environmental alteration (deviation from natural backgrounds): the Bargagli-Nimis scale (2002) and a scale developed by the authors (Corapi, 2011). The Bargagli-Nimis scale is based on the statistical analysis of thousands of measurements referring to Italy, while the second scale is focused on several international references concerning foliose/fruticose lichens used in transplants monitoring in areas characterized by strong anthropization for a maximum exposure period ranging from 3 to 6 months. Secondly, we compared the percentage of sites falling into the different categories. In order to validate the results of this ranking, the statistical significant differences between lichen origin area and study area central tendency values were also verified through the Mann Whitney test, to check the consistency of the enrichment process at the level of the entire study area. Secondly, by converting the absolute concentrations into Delta (Delta = Concentration after Exposure – Concentration before Exposure) to obtain the net bioaccumulation of trace element levels in the two different years. The significance of differences between Delta 2013 and Delta 2019 were also evaluated using the Mann Whitney test. The subsequent treatment of the data was limited only to those trace elements showing at least one case of enrichment, since the aim of the work was to analyse the development of contamination processes. The spatial variation in distribution patterns of such elements was evaluated by means of a combined multivariate-univariate analysis approach. The application of a range of multivariate analysis techniques to the Site x Trace Elements dataset showed that in 2013, Non-Metric Multidimensional Scaling was the most effective technique in interpreting the spatial variation of bioaccumulation levels, while in 2019 Bray Curtis Analysis was the best performing one. These were followed by the Multi Response Permutational Procedure (McCune and Grace, 2002) and Analysis of Variance (Anova) performed for each trace element, using in both cases the factor Groups resulting from the application of the two multivariate analysis techniques. In 2013, a parametric Anova was performed with Tukey test for post-hoc comparisons, while in 2019 a non-parametric Anova (Kruskal-Wallis) was applied with Dunn test to multi-comparisons. To trace the anthropogenic sources responsible for the bioaccumulation levels, firstly a Cluster Analysis was applied in both years to the Trace Elements ranks x Sites dataset. Ranks were used to avoid prevalent associations occurring between macroelements on the one hand, and trace elements on the other hand. The covariance of trace elements was further assessed by means of non-parametric correlation analysis (Spearman). The two outcomes were compared in order to obtain the best evidence to support the hypothesis that elements whose spatial variation is associated with each other most likely derive from the identical emission source. In the case of the biomass power plant, the potential role as a source of trace elements at the level of the whole study area was assessed by means of a parameter widely used by the authors for such purposes (Lucadamo et al., 2016, 2018, 2021a, 2021b). It consists of the frequency with which the wind passing through a potential site reaches all the others located in the study area during the entire period of exposure of the thalli (PNTWRS = Potential Number Times Winds, passing through the site where an anthropogenic source is located, Reaches the other Sites in the study area). The significance of the covariance between this parameter and the lichen bioaccumulation values, tested by means of correlation analysis (Spearman), was used to verify the contribution of the biomass power plant emissions to the spatial variation of trace element concentrations. With regard to traffic, the measurements were conducted using the same method as 2013. Specifically, hourly vehicle transit (counting the number of vehicles passing during the same time interval, 9 a.m.-2 p.m.) was recorded at each of the 41 monitoring sites and, once again, the non-parametric correlation coefficient relating to the spatial variation of the vehicle rate and bioaccumulation levels was calculated. The comparison of the ecophysiological parameters between the two years was carried out in analogy to that of trace elements. The significance of the differences between the lichen origin area and study area central tendency values (medians) for the two years separately, and between the Delta 2013 and Delta 2019, was evaluated. Finally, the possible contribution of trace elements to the spatial variation of ecophysiological parameters was tested by means of non-parametric correlation analysis (Spearman) in both years.

3. Results

3.1. Meteorological conditions

During the two monitoring years, the prevailing winds blow from the south, with a quite modest speed (Fig. 2). Indeed, the series of high speed median values are within the range of “light breeze” according to the Beaufort scale, suggesting a very slow diffusion of contaminants
emitted by the potential local sources (Lucadamo et al., 2021a; NOAA, 2017).

Meteorological data collected in 2013 and 2019 are reported in Table 1. The only parameter showing a difference during the two monitoring campaigns is the value of total rain, which is higher in 2013 than in 2019. It is worth to analyse this result to demonstrate its not relevant influence in the bioaccumulation different trend in the two years. The rainy days were 33 in 2013 (234 mm) and 30 in 2019 (134 mm). In particular during the first monitoring campaign there were four days of moderate-high rain rate events totalling 112 mm (Llasat, 2001; Mukherjee et al., 2016; Wang et al., 2021), highly promoting the trace elements leaching from the lichen thallus surface (Malaspina et al., 2014). Moreover, in both monitoring campaigns there were ca. 130 mm of rainfall in the exposure period, classifiable as light-moderate events (Wang et al., 2021), resulting in both uptake and leaching of trace elements (Dron et al., 2021; Gallo et al., 2017). Therefore, rainfall did not influence the different bioaccumulation values detected in the two different biomonitoring campaigns.

3.2. Trace elements

Fig. 3 shows the classes of enrichment and the relative cases for the 2 years as the average value of the cumulative percentage of all elements. In 2019, the level of diffuse enrichment has significantly decreased. In fact, the average value of cases of no enrichment is 49.6% in 2013 and 68.5% in 2019, while that of moderate enrichment is reduced from 17% in 2013 to 4.2% in 2019. Cases of low enrichment fall, on average, from 28.9% to 24.3%. On the contrary, cases of high or extremely high enrichment are 22 in 2013 and 21 in 2019, although the elements involved change. The elements that contribute most to the variation are expressed as total mm.

![Fig. 2. Wind Direction (a) and wind speed (b) values collected during the two biomonitoring campaigns.](image)

![Fig. 3. Cumulative percentage distribution of the enrichment cases compared to a condition of no land use per element and per year according to the Bargagli and Nimis (2002) Naturality-Alteration Scale and to an enrichment scale developed by the authors (Corapi, 2011). N.E. = No Enrichment, L.E. = Low Enrichment, M.E. = Moderate Enrichment, H.E. = High Enrichment, V.H.E. = Very High Enrichment.](image)

### Table 1

Meteorological data collected in the study area in 2013 and 2019 monitoring campaigns by means the Vantage Vue meteorological station. The values of temperature (°C) and relative humidity (%) are expressed as mean, while rain is expressed as total mm.

<table>
<thead>
<tr>
<th>Monitoring campaign</th>
<th>High temperature</th>
<th>Low Temperature</th>
<th>Humidity</th>
<th>Rain</th>
</tr>
</thead>
<tbody>
<tr>
<td>2013</td>
<td>14.1</td>
<td>8.5</td>
<td>68.6</td>
<td>234</td>
</tr>
<tr>
<td>2019</td>
<td>12.4</td>
<td>9.5</td>
<td>66.1</td>
<td>134</td>
</tr>
</tbody>
</table>
(C1–C5) are also evident, again appearing reducible to 3 zones, due to the absence of statistically significant differences between elements: zone B (Clusters C1 and C3), including 18 sites arranged 44% in the north and 66% in the south, zone A (Cluster C2) of only 4 sites, 3 in the south and 1 in the north, and zone C (Clusters C4 and C5) composed of 19 sites, 11 of which are in the north (57%) and 8 in the south (43%).

The segregation between the three zones is once more attributable to the higher values of Al, Ti, V and Co, on average, in zone C, which differ in a statistically significant manner from those of zone A and lichen origin area. The central tendency values of these four elements in zone B are halfway between those of zone A and zone C, as well as not differing from the lichen origin area. Zone A can therefore be considered, in analogy to what was found in 2013, as a “local scale internal control” group.

Fig. 4. Absolute Delta values for 2013 and 2019 expressed as Log (Delta+1). (*) Statistically significant differences (p < 0.05).

Fig. 5. Distribution, in the study area, of the clusters derived from the application of the Non-Metric Multidimensional Scaling to the Sites x Trace Elements dataset related to the monitoring performed in 2013 (C1–C2, Zone A = green; C3, Zone B = yellow; C4–C5, Zone C = red).

In 2013 the spatial variation of the PNTWRS parameter, calculated for the site corresponding to the biomass power plant, showed a statistically significant association (p < 0.05) with Ti (r = 0.38) and Co (r = 0.36) (Lucadamo et al., 2016), while in 2019 this correlation persists (p < 0.05) only in the case of Ti (r = 0.32). On the other hand, in the first monitoring study of the urban-industrial district of Rende there were 4 elements that covaried significantly (p < 0.05) with the traffic rate (Sb: r = 0.56, Sn: r = 0.66, Mo: r = 0.62, Cu: r = 0.38), whereas in 2019 the association with tin is absent (Sb: r = 0.43, Mo: r = 0.34, Cu: r = 0.31).

Regarding the ecophysiological parameters in Table S4 is reported the comparison between the central tendency values (medians) measured in the lichen origin area and in the study area at the end of the thalli exposure performed in 2013 and 2019. Fig. 8 shows the absolute Delta
relative to the pre- and post-exposure conditions of lichen thalli in both years.

In both years, the quarterly exposure to the conditions of the study area determines a relevant increase in the values of TBA reactive substances equal, respectively, to 337% of the values measured in the lichen origin area in 2013 and 203% in 2019 ($p < 0.05$). The difference between the two Deltas (30.6%) also results in a statistically significant reduction of oxidative stress levels from 2013 to 2019. The differences between the 2013 and 2019 absolute Delta appear statistically significant with those related to pigments showing the highest % deviations in 2019 compared to the lichen origin area (Chl $a = 24\%$, Chl $b = 21\%$ and $X + C = 16\%$). Table 2 reports the correlation coefficients (Spearman) and relative significance for the association between the spatial variation of trace elements bioaccumulation in thalli transplanted in the study area and the corresponding spatial variation of ecophysiological parameters values for both years. Two parameters are associated with the spatial variation of trace elements, namely peroxidation levels (TBArs) and photosynthetic efficiency ($Fv/Fm$). In 2013, TBA reactive substances show 11 cases of statistically significant association with bioaccumulation levels. Of these 6 are also confirmed in 2019 with no new correlations appearing. On the other hand, in this year, 4 statistically significant correlations (of negative sign) between photosynthetic efficiency and V, Mn, Co and Ni, absent in 2013, can be highlighted.

4. Discussion

Comparison of metal bioaccumulation developed by thalli of the lichen *Pseudevernia furfuracea* transplanted in the same area and period has highlighted that in 2019, 6 years after the first monitoring campaign, the levels of environmental exposure to some of the trace elements examined have been significantly reduced. In 2013, the cases of no enrichment and the sum of those of slight and moderate enrichment are almost equivalent (49.6% and 45.8% respectively). The latter are essentially attributable to Al, Ti, V, Co, Mn, Mo, Sn and Sb, while 22 cases of high or extremely high enrichment due to Cu, Cr, As, Zn and Sb are evident (Lucadamo et al., 2016), which constitute the main environmental criticalities. In 2019, the cases of no enrichment increase significantly (68.5%), while the total of those of slight and moderate
Table 2

<table>
<thead>
<tr>
<th>Elements</th>
<th>2013 Correlations</th>
<th>2019 Correlations</th>
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<tbody>
<tr>
<td></td>
<td>r</td>
<td>p</td>
</tr>
<tr>
<td>Mn-TBArs</td>
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</tr>
<tr>
<td>Sn-TBArs</td>
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</tr>
<tr>
<td>Sb-TBArs</td>
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<td>Cr-TBArs</td>
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</tr>
<tr>
<td>Ni-TBArs</td>
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<tr>
<td>Cu-TBArs</td>
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<tr>
<td>Mo-TBArs</td>
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<td>Zn-TBArs</td>
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<tr>
<td>As-TBArs</td>
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<td>0.001</td>
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</tr>
<tr>
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<td>N.S.</td>
<td>N.S.</td>
</tr>
<tr>
<td>Co-Fv/Fm</td>
<td>N.S.</td>
<td>N.S.</td>
</tr>
<tr>
<td>Ni-Fv/Fm</td>
<td>N.S.</td>
<td>N.S.</td>
</tr>
</tbody>
</table>

Fig. 8. Absolute Delta between (a) the values of TBA reactive substances (p < 0.05, Mann Whitney test), (b) the pigments values (p < 0.01, Mann Whitney test), (c) the values of viability (A492) and PSII efficiency (p < 0.01, Mann Whitney test, except for A492 which is not statistically significant) measured before and at the end of the exposure period in the two monitoring campaigns. Chl a = Chlorophyll a, Chl b = Chlorophyll b, X + C = Xanthophylls and Carotenoids.

Enrichment fall to 28.5%. This result is primarily attributable to a significant reduction in the Delta of Al, Ti, V, Co, Cr, Mo, Sn and Pb. The cases of high or extremely high enrichment of Cu, Cr, As, Zn and Sb are drastically reduced (from 22 to 8), but there is an increase in those of Mn (from 0 to 13). Following this result, it should be noted that the patterns of spatial contamination are quite similar. In fact, in both years they are characterised by a prevalence of higher levels of Al, V, Ti and Co bio-accumulation in the northern side, consistent with the trend of prevailing winds blowing from the south for 56% of cases in 2013 and 62% in 2019. Most of the wood biomasses used as fuels by the biomass power plant are conifers coming from the Sila massif. From a geological point of view, this massif is characterised by the prevalence of intrusive rocks (granites, granodiorites, quartz diorites) (Ogniben, 1973), i.e. substrates with an acid pH that tend to release aluminium. It is known that needle-bearing trees can accumulate this element without negative consequences, either as a result of co-deposition mechanisms with silicon (Hodson and Sangster, 1999) or through the synthesis of proteins that allow the phloematic transport of aluminium (Brunner and Sperisen, 2013). On the other hand, literature data indicate that Ti, Co and V are found, associated with particulate matter, in emissions from wood combustion with emission factors of $10^{-3}$, $10^{-4}$ and $10^{-7}$ lb/MBtu respectively (EPA, 1999; California Air Resources Board, 2011). It should be added that the presence of titanium in some cases could also suggest phenomena of chemical contamination of the wood employed since titanium dioxide nanoparticles are used as an antifungal agent (De Filipo et al., 2013). In both years, some peaks in the concentration of these 4 elements, were detected in the first kilometre away from the biomass power plant. However, these do not determine statistically significant differences (Kruskal Wallis, p > 0.05) between the central tendency values calculated for kilometric ranges (0–1 Km, 1–2 Km, 2–3 Km). This could be the result of the particle size with which these elements are associated, as well as the height of the chimney (42 m) and the wind speed (on average between 2 and 3 ms$^{-1}$ in both years). Wood combustion generates a broad spectrum of particle sizes ranging from 0.05 to 10 μm (Pagels et al., 2002). Moreover, when wood is burned without debarking process, the particles show a bimodal distribution of 0.1 μm and 4.5 μm (Hasler and Nussbaumer, 1998), which is the practice followed by the Actelios power plant. Ti is mainly associated with fine-ultrafine particles (Kleeman et al., 1999), while Co and V are also found in coarse particles (Nzihou and Stanmore, 2013). Based on our results, it can be assumed that the above-mentioned elements were mainly emitted in association with finer particles, resulting in a low deposition rate that significantly limited their accumulation close to the biomass power plant. It is worth noting that in 2013 the use of the PNTWRS parameter made it possible to associate (p < 0.05) the contribution of the biomass power plant to the spatial variation of Ti and Co and, given their covariance with V and Al, also that of these latter in the hypothesis of being emitted less frequently (Lucadamo et al., 2016). In 2019, the PNTWRS parameter only associates Ti with the biomass power plant. Considering the lower bioaccumulation values (compared to 2013) not only of this element but also for Al, V and Co, it can be assumed that their emission frequency/intensity has been further reduced. However, as the covariance of the spatial variation of these elements in 2013 appears to be lower than in 2019, it is possible that in the second case if the absolute number of events in which Al, V, Ti and Co emitted from the stack of the biomass power plant has decreased, the number of cases in which they were emitted together has increased. This would suggest a greater homogeneity in the composition of the wood materials burned. A confirmation of this could come from the lack of formation of the Cr, As, Cu and Zn cluster in 2013, ascribed to an infrequent blending of wood from forests with demolition wood treated with preservatives such as chromated copper arsenate (Wasson et al., 2005; Morais et al., 2021). Considering that the use of chromated copper
arsenate was banned, except for the industrial sector, in 2003 (CSTE, 2003) it is likely that the quantities accumulated over time are being depleted or at least significantly reduced, thus decreasing the need to dispose of it in power plants fuelled by biomass combustion. Undoubtedly, two important factors contributing to the reduction of trace element emissions from the Actelios power plant are the actions taken to diversify the way electricity is produced. Firstly, the installation of a concentrating solar thermodynamic plant in 2014, which allows the integration/replacement of biomass combustion as a source of electricity generation. It has led to a reduction in CO₂ emissions of 1250 tonnes per year (Rinnovabili, 2021), as well as in atmospheric particulate matter and associated substances. There are several literature studies suggesting the economic and energy convenience in the development of hybrid combined heat power plants (Servert et al., 2011; Srinivas and Reddy, 2014; Soares et al., 2018; Hartl et al., 2012). However, there is a strong scarcity of studies on the related environmental impacts. The present work represents one of the few studies on this important issue, associating the development of a hybrid power plant with positive effects on air quality. A second factor that may have contributed to the improved emission characteristics of the Actelios biomass power plant is the increased use of exhausted olive pomace as fuel (Falck, 2009; Silateam, 2021). In fact, it contains very low levels of trace elements (Medouni-Haroune et al., 2018; Intini et al., 2011; Leite et al., 2016), especially when compared to woody biomass, so its combustion contributes only marginally to metals atmospheric contamination.

The evaluation of the two years vehicular traffic measurements showed a mean value of 606 and 741 veh/h respectively in 2013 and 2019, with an increase of about 22% in 2019 compared to 2013 for the same period of the year and stint of measurements. In Table S5 are reported traffic measurements in each site of biomonitoring area in 2013 and 2019. Cu, Sb and Mo are frequently used as tracers of brake bearing deterioration (Dongarrà et al., 2009; Bukowiecki et al., 2009; Song and Gao, 2011). This is in line with the results of the 2019 monitoring, while in 2013 Sn, likewise produced by bearing wear (Amato et al., 2009), was also associated with vehicular traffic. On the other hand, in a previous work carried out by the authors, it was demonstrated that biomass power plants may also contribute to the emission of Sb (Lucadamo et al., 2018) as this element, together with halogens, is used as a retardant in wood combustion, an effect that is however masked by the main traffic contribution. If this was also the case for the Actelios biomass power plant, a reduction in Cu and Sb emissions from the latter would have been offset by an increase in those from traffic, resulting in a substantial lack of difference in bioaccumulation levels between the two years. The interpretation of Sn and Mo results is not immediate, as it is evident that the Delta value in 2019 is lower than in 2013. A partial explanation could be that in the last 10 years there has been a change in brake bearing materials from metallic to semi-metallic or ceramic-based (Bridgestone, 2021), i.e. a reduction in the former components. However, while Sn and Mo are mainly found as friction modifying agents, Cu and Sb are also used as pad filling and reinforcing elements (Sinha et al., 2020; Vijay et al., 2020), so their representation in the percentage composition of new brakes would still remain appreciable in contrast to that of Sn and Mo as well as their contribution to environmental pollution. The only element bucking the general trend is Mn. In both years, the exposure to this element leads to a statistically significant increase compared to the lichen origin area levels. The Delta in 2019 is not significantly higher than in 2013, but Mn shows 13 cases of high enrichment, which were not found in the previous monitoring. If that circumstance it appeared reasonable to assume a contribution of both the power plant and traffic to the spatial variation of this element (Lucadamo et al., 2016) being either environmental sources of manganese (Sippula et al., 2007; Swietlik et al., 2013). However, given the reduction, in 2019, of trace element emissions by the biomass power plant and, conversely, the relevant increase in the car-vehicle rate, it can be assumed that at least some of the cases of high enrichment can be ascribed to vehicular traffic, although a statistically significant correlation between the spatial variation of the two parameters is not detectable. Such an outcome is likely due to the confounding action of other unidentified sources acting at the local scale.

Analysis of the results of the ecophysiological parameters shows, in both monitoring campaigns, that the only parameter that changes following exposure in the study area is the oxidative stress, with a reduction in the Delta of TBA reactive substance levels (p < 0.05) of approximately 30% in 2019 compared to that of 2013. This result could, at least in part, be associated with the decrease in trace element contamination. Literature data widely support the toxic effect of metals on plant organisms, determined, in several cases, by the activation of oxidative stress phenomena (Smeet et al., 2005; Rizvi et al., 2020; Mazhoudi et al., 1997). Moreover, in previous monitoring activities, where thalli of the same lichen species used in the present work were transplanted in urban-industrial areas, an association between the spatial variation of TBA reactive substances and that of several trace elements was found (Corapi et al., 2014; Lucadamo et al., 2016, 2018). Eleven metals-TBAr pairs showed a statistically significant correlation in 2013, a number that drops to six in 2019. Two of the five correlations not detected in 2019 are related to Ni and Mn. Of these elements, only Mn actually shows enrichment levels in both years that could lead to cellular peroxidation processes. However, the comparable mean Delta deviations between the two monitoring campaigns, suggests that the lack of significance of the coefficient (p > 0.05) in 2019, could be due to a concurrent action of other factors which, in 2013, were less effective than Mn, into promoting oxidative stress processes. The result of the other three missing correlations involving Mo, Cr and Co, is more interesting. Indeed, in 2019 both their bioaccumulation (Delta) is significantly lower than in 2013 and the difference between lichen origin area and study area, in the case of Cr and Mo, is not appreciable (p > 0.05). Since Cr and Co are attributable to emissions from the biomass power plant, this data supports the hypothesis that a reduction in the environmental impact of trace elements from the biomass power plant can contribute to an improvement in air quality. In the case of Mo, the interpretation given above regarding the change of materials for the production of vehicular brake pads applies. With regard to photosynthetic efficiency, the presence of statistically significant coefficients in 2019 for an inverse correlation with V, Mn, Ni and Co (absent in 2013) should be noted. However, both the difference with respect to LOA is not appreciable (p > 0.05) and the value measured does not denote any pathological state, as suggested in the literature (Backor and Loppi, 2009), so the relation detected does not appear to produce negative consequences on the physiological state of the thalli.

5. Conclusions

Energy production from biomass power plants is expected to increase in the forthcoming years due to rise of government subsidies and the expansion of the global pellet market, given its higher energy efficiency (Gielen et al., 2019; Aguilar et al., 2020; Zuidema, 2020; Yu et al., 2021). To make this energy market sector really sustainable, its environmental impacts need to be reduced. Our study confirms that conversion to hybrid plants and fuel diversification are among the various strategies that should be pursued. Two successive air quality monitoring studies of a 15 MWh biomass power plant have shown a significant improvement in air contamination levels and a reduction of ecophysiological stress measured in lichen thalli. The elements identified as tracers of emissions from the biomass power plant in 2013, namely Al, Ti, V, Co, Cr, Cu and As, by transplanted epiphytic lichens carried out in the winter-spring transition quarter, in 2019, following biomonitoring performed in the same way, show an average decrease in bioaccumulation, equal to respectively: 54%, 58%, 37%, 45%, 41%, 25% e 5%. In addition, the number of peaks in the concentration of Cr, Cu and As is reduced by more than 50%. The levels of oxidative stress measured in lichen transplants at the end of the exposure period are 30% lower in 2019 than those measured in 2013. At the same time the association
between the spatial variation of the bioaccumulation of Cr and Co and that of the values of TBA reactive substances is decreased, suggesting that the improvements carried out may also contribute to reduce the environmental pollution processes. The only element that manifests the development of contamination peaks in 2019, absent in 2013, is Mn. For this element it is hypothesized a contribution due to the significant increase in rates of vehicular traffic, a result suggesting the need to take more effective measures against this type of anthropogenic pressure in the monitored urban-industrial area.

Author statement

L. Lucadamo: Conceptualization, Supervision, Formal analysis, Writing – original draft. L. Gallo: Supervision, Writing – original draft, Project administration. A. Corapi: Writing – review & editing, Investigation, Data curation, Formal analysis, Visualization

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.apr.2021.101346.

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