

A BIOMONITORING STUDY ON *Xanthoria parietina* (L.) Th.Fr IN ISPARTA, TURKEY**YAVUZ Mustafa, ÇOBANOĞLU Gülşah**

Abstract. This study aims at determining air quality and potential pollutant sources in Isparta, as well as Gölcük Nature Park, through lichen biomonitoring. The specimens of cosmopolite epiphytic foliose lichen *Xanthoria parietina* (L.) Th.Fr were sampled from 8 localities in the study area and analysed in an ICP – MS device with reference material, in order to detect Al, As, Cd, Cr, Cu, Fe, Mn, Ni, Pb, V, and Zn elements. The distribution of these airborne elements in the study area was mapped spatially. The results indicate that the elemental sequence of average concentrations measured is $Ni > V > Cr > Fe > As > Cu > Al > Zn > Mn > Cd > Pb$. Cluster analysis results of the elemental data indicate a strong correlation of the major 3 contaminants in the urban area, which are Ni, V, and Cr. The lichen specimens accumulated maximal concentrations of Cd, Cu, Mn, Ni, Zn within the Nature Park, the rest of the elements in northern and the eastern localities surrounding the city. The elemental pollution in Isparta is higher than expected due to extensive use of coal as fossil fuel in the city, and it seems to be associated with topographic and climatic characteristics of the city.

Keywords: airborne elements, lichen, biomonitoring, Isparta.

Rezumat. Studiu de biomonitorizare al speciei *Xanthoria parietina* (L.) Th.Fr în Isparta, Turcia. Acest studiu are ca scop determinarea calității aerului și posibilele surse poluante în Isparta ca de asemenea și în parcul Natural Gölcük, prin biomonitorizarea lichenilor. Speciile de foliole ale epifitelor cosmopolite *Xanthoria parietina* (L.) Th.Fr au fost luate din 8 localități ale zonei de studiu și au fost analizate cu aparatul ICP – MS pentru a detecta elementele: Al, As, Cd, Cr, Cu, Fe, Mn, Ni, Pb, V și Zn. Distribuția acestor elemente transmise prin aer în zona de studiu, a fost cartografiată păstrând anumite limite. Rezultatele indică, că există o legătură elementară între concentrațiile măsurate și aceasta este $Ni > V > Cr > Fe > As > Cu > Al > Zn > Mn > Cd > Pb$. Analizele rezultate indică o strânsă legătură a celor 3 contaminanți majori în zona urbană, aceștia fiind Ni, V și Cr. Speciile de licheni au acumulat o concentrație maximă de Cd, Cu, Mn, Ni, Zn în interiorul Parcului Natural, restul elementelor fiind acumulate în nordul și estul localităților din jurul orașului. Poluarea din Isparta este mai mare decât s-ar fi așteptat datorită folosirii excesive a cărbunelui ca și combustibil și pare a fi asociat cu caracteristicile climatice și topografice ale orașului.

Cuvinte cheie: elemente aeriene, licheni, biomonitorizare, Isparta.

INTRODUCTION

Lichens are symbiotic organisms, consisting of a mycobiont as the fungal partner, and one or more photobionts, like a green alga or cyanobacteria. Lichens are sensitive to environmental changes due to their biological peculiarities and symbiotic lifestyle (HAWKSWORTH & ROSE, 1976), thus lichens are sensitive bioindicators for air pollution, climatic changes, forest structures and dynamics regarding the quality of biodiversity (GIORDANI et al., 2012). Since lichens are slow-growing organisms and have neither roots nor a protective cuticle layer, they absorb air pollutants with moisture mainly through the thallus surface, hence they are open to the effects of any atmospheric contaminants (GARTY, 2001; WOLTERBEEK et al., 2003). Consequently, trace element concentrations in lichen thalli indicate the environmental levels for the elements (BARI et al., 2001). Using lichens as biomonitoring organisms facilitates many aspects such as sampling and cost, and makes them advantageous for the spatial and temporal assessment of pollutant levels in the environment (LOPPI et al., 2003; SCERBO et al., 2003).

The number of lichen biomonitoring studies in Turkey has been increasing during the last two decades (ASLAN et al. 2004; YENISOY-KARAKAŞ & TUNCEL 2004; MENDİL et al. 2005; CAYIR et al. 2007; YILDIZ et al. 2008; İÇEL & ÇOBANOĞLU, 2009; ŞENKARDEŞLER & AYSEL, 2010; ÖLGEN & GÜR, 2012; DOĞRUL-DEMIRAY et al., 2012; BOZKURT, 2017; KURNAZ & ÇOBANOĞLU, 2017). Isparta is a city located in the Mediterranean Region of Turkey and has undergone a dense and uncontrolled increase of population, followed by irregular urbanization at the end of 1990s. This increase in population and urbanization caused environmental problems in the Isparta province. Industrial zones are concentrated in the north-northeast direction of the city. As mentioned in the detailed report by DEMİR (2010), there are various workshops and some factories, focused on sectors of forestry and carpentry (51 facilities), food and beverage (46), metal equipment (34), cement and marble (28), textile and leather (25), petro-chemistry and paint (19), cosmetics (4), and fertilizers (1). For residential heating, wood, coal and fuel oil are widely used in the province, and 102,920,456 kg of coal are consumed between April 2008 – April 2009 in the centre. In addition to the industrial facilities, a major highway between Afyonkarahisar and Antalya pass through Isparta, which is another potential source of airborne pollutants. Air quality assessment done by local authorities in Isparta is based on the monitoring of PM_{10} and SO_2 in air. In such cities with industrial pollution, biological monitoring through lichens is necessary to reach longer-term and broader perspective outcomes. The main objective of this study is assessment of the air quality in Isparta by lichen biomonitoring. In order to achieve this, the degree of metal deposition in Isparta is examined via quantitative analyses and the relationship between potential pollutant sources and levels of airborne metals are investigated. Considering that air pollutants can be transported to very long distances by environmental factors such as wind (ÇOBANOĞLU, 2015), Gölcük Nature Park (GNP) within the provincial borders was also included in this study. This work also aims to establish a baseline for future biomonitoring studies in Isparta.

MATERIALS AND METHODS

Isparta is located in the North-Western Mediterranean Region, Turkey (Fig. 1). The southern part of the city is mountainous while the western part is sporadically hilly and due to these geographical peculiarities, industrial zones are located in eastern and northern parts of the urban area. The northern and eastern parts of the suburban area are surrounded by agricultural fields as well. The Gölcük Nature Park (GNP) is located in the South, South-western part of Isparta city centre. Isparta has a Semi-Arid Mediterranean climate with a mean annual rainfall of 506 mm and a mean annual temperature of 12 °C (Fig. 2). The prevalent winds in the region are from the South-west (9 m/s), South (8.1 m/s), South-east (6 m/s), and West (1.6 m/s) directions (IMIM, 2010).

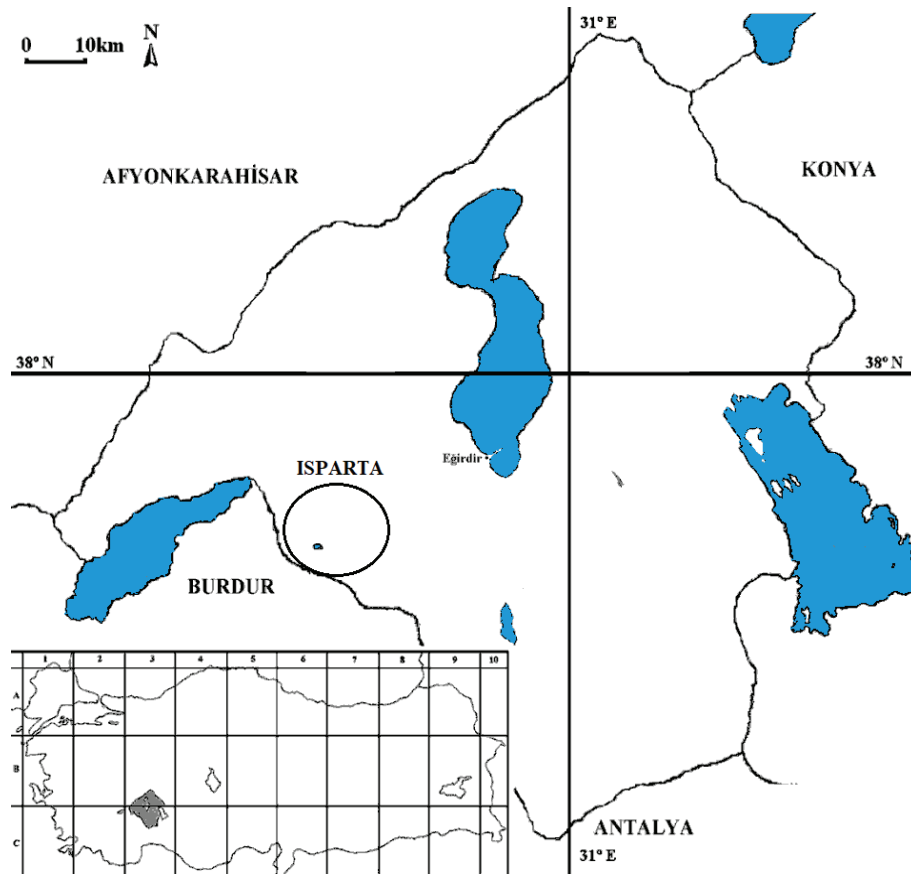


Figure 1. Isparta and the study area (original).

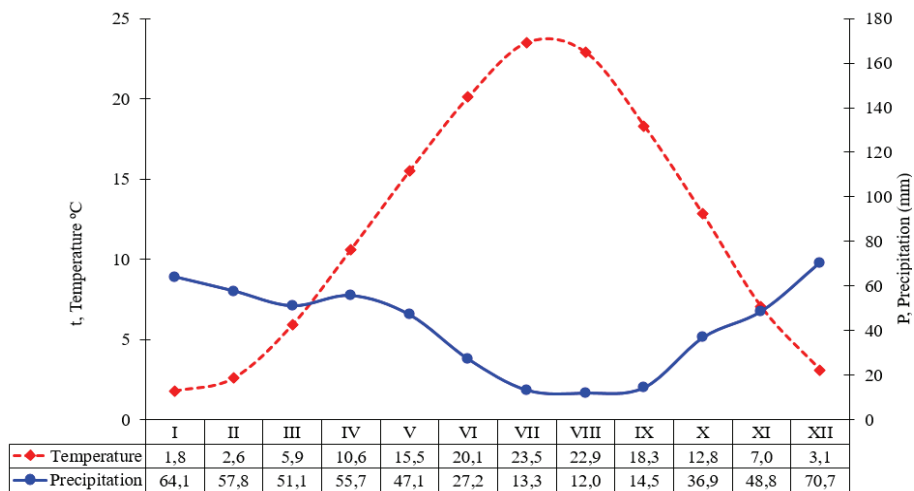


Figure 2. Ombrothermic diagram of Isparta (original).

The epiphytic foliose lichen *Xanthoria parietina* was selected as biomonitoring organism due to its common presence in the urban regions and GNP, besides high tolerance to atmospheric pollution. Lichen samples were collected from 8 stations distributed around the major industrialized and suburban areas of Isparta as well as GNP between June 2009-July 2010 (Fig. 3).

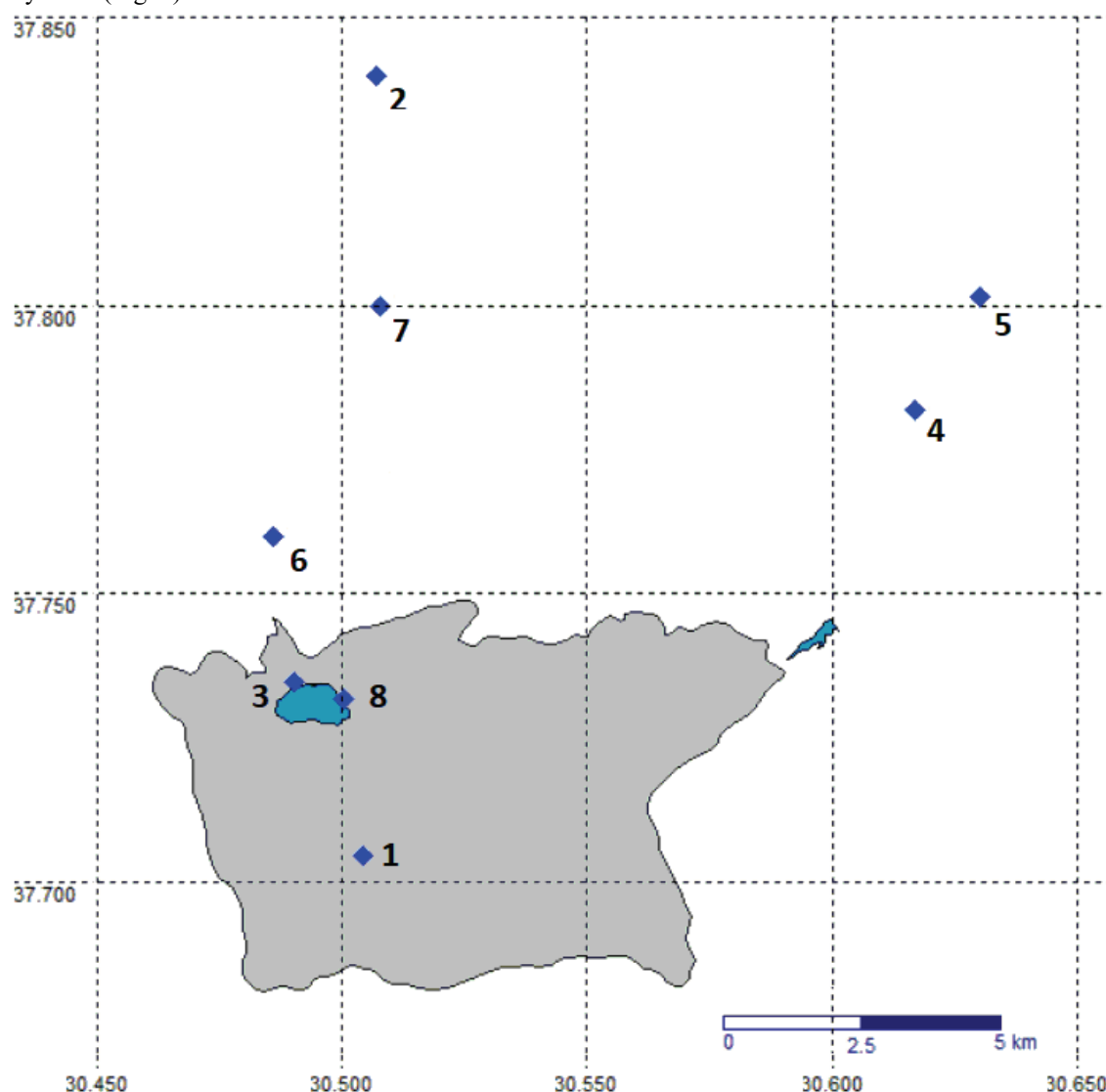


Figure 3. The sampling sites of lichens in the study area.

A composite lichen sample was randomly collected in each station by exploring an area of 50×50 m. Lichen rosettes ≥ 2 cm in diameter were selected as samples of biomonitoring and were taken from the bark of *Amygdalus* sp., *Pinus nigra*, *Populus alba*, *Populus nigra*, *Quercus* sp., and *Robinia pseudacacia* at a height of at least 120 cm above the ground in order to avoid terrestrial contamination. Lichen samples were air-dried and stored in polyethylene bags in the laboratory, until chemical analysis. GPS and altitude information of the localities recorded by a *Garmin e-trex summit* device are given in Table 1.

Table 1. Description of the Localities.

No	Locality	GPS Coordinates	Altitude (m)	Date
1	Gölcük Nature Park, Southern Border	37°42'16.00"N - 30°30'15.50"E	1620	20.06.2009
2	Behind Campus, Koçtepe Village	37°50'23.00"N - 30°30'26.00"E	1118	04.04.2010
3	Gölcük Nature Park, Picnic Area	37°44'04.70"N - 30°29'25.40"E	1395	01.05.2010
4	Sav Village	37°46'54.40"N - 30°37'02.20"E	980	18.04.2010
5	Küçük Hacılar Village	37°48'05.60"N - 30°37'49.40"E	972	18.04.2010
6	Gülbirlik Rose-oil Factory	37°45'34.80"N - 30°29'09.90"E	1215	24.04.2010
7	Kayı Village	37°47'59.70"N - 30°30'28.50"E	1069	24.04.2010
8	Gölcük Nature Park, North-Eastern Part	37°43'53.30"N - 30°30'02.50"E	1390	22.07.2010

Before the analyses, lichen samples were cleaned with plastic tweezers and pieces of bark or extraneous materials were removed. Then samples were stirred in deionized water for thirty seconds to remove the dust over the thalline surface. Cleaned samples were dried at room temperature for 24 h and then at 105 °C of incubation for 18 hours. The dried samples were ground and homogenized with an agate mortar and pestle. 200 mg of dry powdered lichen sample were digested in a mixture of 10 mL 2:8:2 HCl:HNO₃:H₂O₂ at 180 psi pressure, between 0-95 °C for 15 minutes, at 95 °C for 1 minute and finally between 95-200 °C for 15 minutes in CEM - Mars Xpress Microwave device. The digestion solution was finally diluted to 20 mL with ultra-pure water and analysed for Al, As, Cd, Cu, Fe, Mn, Ni, Pb, V and Zn with ICP-MS (ACME Analytical Labs., Canada). Analytes were selected mainly based on the potential pollutant sources in Isparta. Efficiency and quality of analytical results were evaluated by analysing a certified reference material, IAEA-336 Lichen (International Atomic Energy Agency, Vienna), with the same procedures adopted for the samples. The precision of analyses was found to be less than 5% for all elements analysed (Table 2).

The Surfer® 15 software package was used to draw a contour map for each analysed element. The Kriging algorithm with gridding method was selected to create the contour maps representing the aerial distribution of metal concentrations in Isparta.

Correlation and cluster analyses were performed on the analytical data to evaluate the relationship between the pollutants and possible pollutant sources. Cluster analyses of all elemental data from the urban and the industrial districts of Isparta were conducted separately using the PAST® 3.18 program package. Complete linkage clustering method and squared Euclidean distance metric were used in the cluster analysis.

RESULTS AND DISCUSSION

The analysis of certified reference material showed that the concentrations of almost all analysed elements were found within the limits of their certified values. The IAEA reference sheet does not include a recommended value for Ni element concentration. The element concentrations detected in *Xanthoria parietina* samples collected from localities in the study area are given in Table 2.

Table 2. Concentrations (µg/g) of elements in lichens collected from 8 localities in the Isparta province.

Locality	Al	As	Cd	Cr	Cu	Fe	Mn	Ni	Pb	V	Zn
1	5,697.53	6.48	0.23	18.29	14.14	5,315.73	279.10	20.74	8.93	27.74	264.42
2	11,294.94	7.76	0.24	33.81	19.16	8,733.10	373.10	32.66	12.99	62.31	212.42
3	5,155.99	5.17	0.46	19.97	65.80	5,013.40	442.70	169.13	8.50	23.23	420.12
4	3,231.03	4.30	0.16	14.44	10.43	3,279.33	124.60	13.27	10.04	15.84	234.82
5	6,275.47	7.54	0.13	20.26	26.10	6,017.91	278.90	42.23	13.48	31.16	219.12
6	993.30	0.84	0.55	3.61	25.96	750.40	47.17	7.01	7.63	2.41	15.35
7	572.81	0.43	0.42	0.89	23.80	436.59	0.20	32.02	6.82	1.32	41.76
8	5,583.76	5.24	0.10	16.92	13.71	4,878.21	146.00	12.49	10.65	24.56	243.72
Mean	4,850.60	4.72	0.29	16.02	24.89	4,303.08	211.47	41.19	9.88	23.57	206.47
IAEA (Certified)	680.00	0.63	0.12	1.06	3.60	430.00	63.00	**	4.90	1.47	30.40
IAEA (Measured)	661.20	0.62	0.11	1.11	3.55	413.44	60.60	2.45	4.75	1.41	28.91
Confidence %	97.24	97.95	95.73	95.22	98.53	96.15	96.19	**	96.89	95.81	95.10

** No certified value

The order of the average element concentrations in the study area is **Ni > V > Cr > Fe > As > Cu > Al > Zn > Mn > Cd > Pb** according to data of multi-element analysis. The major 3 contaminants in the research area are Ni, V, and Cr respectively probably due to the heavy use of coal in heating facilities and gasoline in vehicles. *X. parietina* specimens tend to maximal degrees of Al and As concentrations in the northern localities while those of As and Pb in eastern localities and interestingly Cd, Cu, Mn, Ni and Zn in the localities of Gölcük Nature Park. The heavy metal pollution in Isparta city is higher than expected due to the extensive use of coal as fossil fuel, and seems to be associated with topographic and climatic characteristics.

In Isparta, **Al** is consumed or used in cars, trucks, doors and windows as a construction material. **As** is consumed or used in dyeing industry, in some agricultural pesticides and it is emitted by coal-fired heating. **Cd** is consumed or used in phosphate-based fertilizers in agriculture and it is emitted by coal-fired heating. **Cr** is consumed in textile and refrigerant industries. **Cu** is consumed or used in dyeing industry, and it is emitted by coal-fired heating. **Fe** is consumed or used in dyeing industry, constructions and metal works, it is emitted from soil and by coal-fired heating. **Mn** is consumed or used in ceramic, tile and brick production as well as dyeing industry. **Ni** is emitted by coal-fired heating and through exhaust gas from cars. **Pb** is emitted by coal-fired heating and through exhaust gas from cars. **V** is consumed or used in dyeing industry, and it is emitted by coal-fired heating and through exhaust gas from cars. **Zn** is consumed or used in dye, textile, plastic industries and cosmetics. It is emitted by coal-fired heating and through exhaust gas from cars.

Localities with the numbers 1, 3 and 8 are inside the borders of the Gölcük Nature Park while the rest of the localities are surrounding the urban area and are closer to possible sources of pollution. Spatial distributions of the element concentrations in the survey area are shown in Fig. 4a-b.

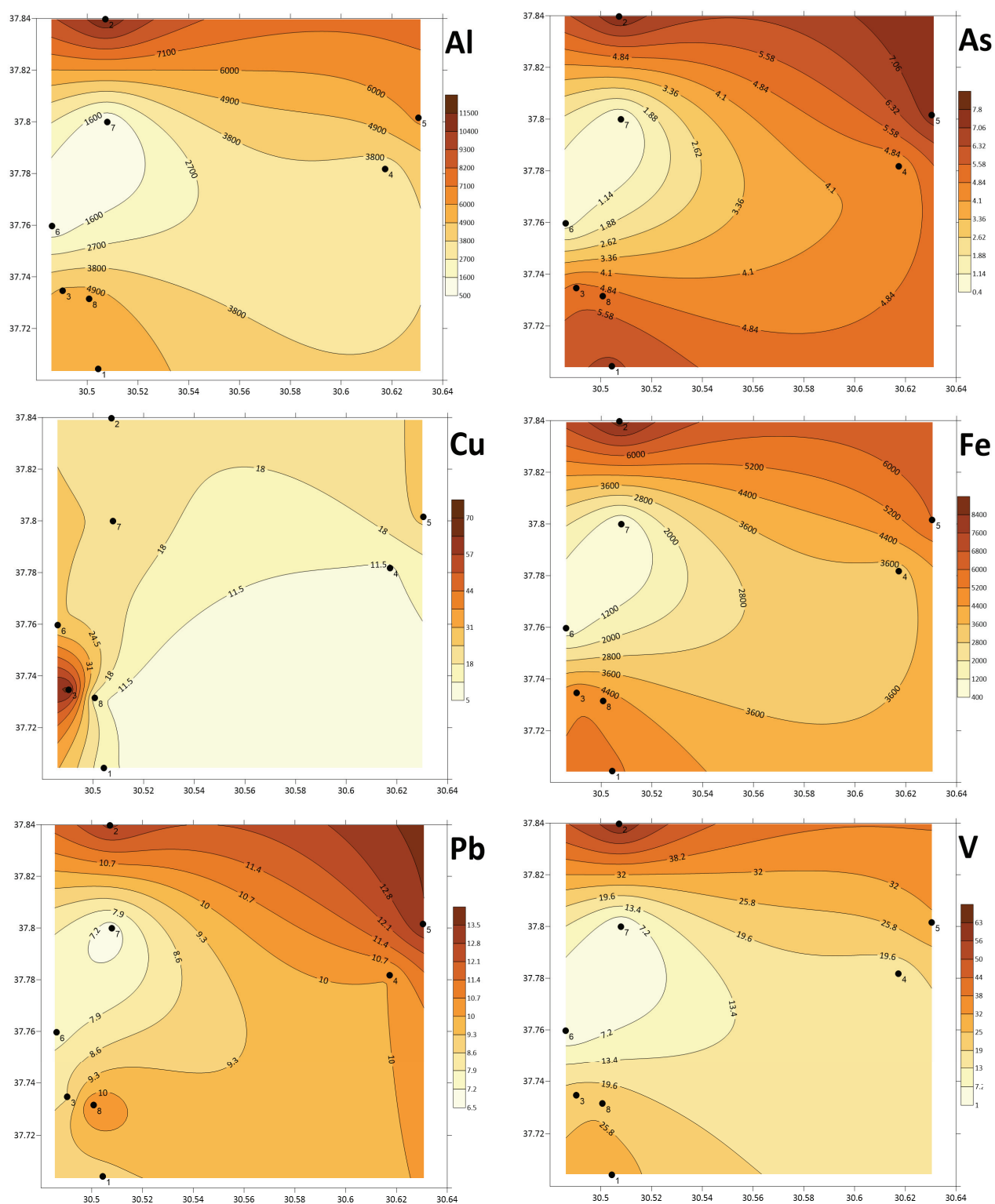


Figure 4a. Spatial Distribution of Al, As, Cu, Fe, Pb and V concentrations in studied locations.

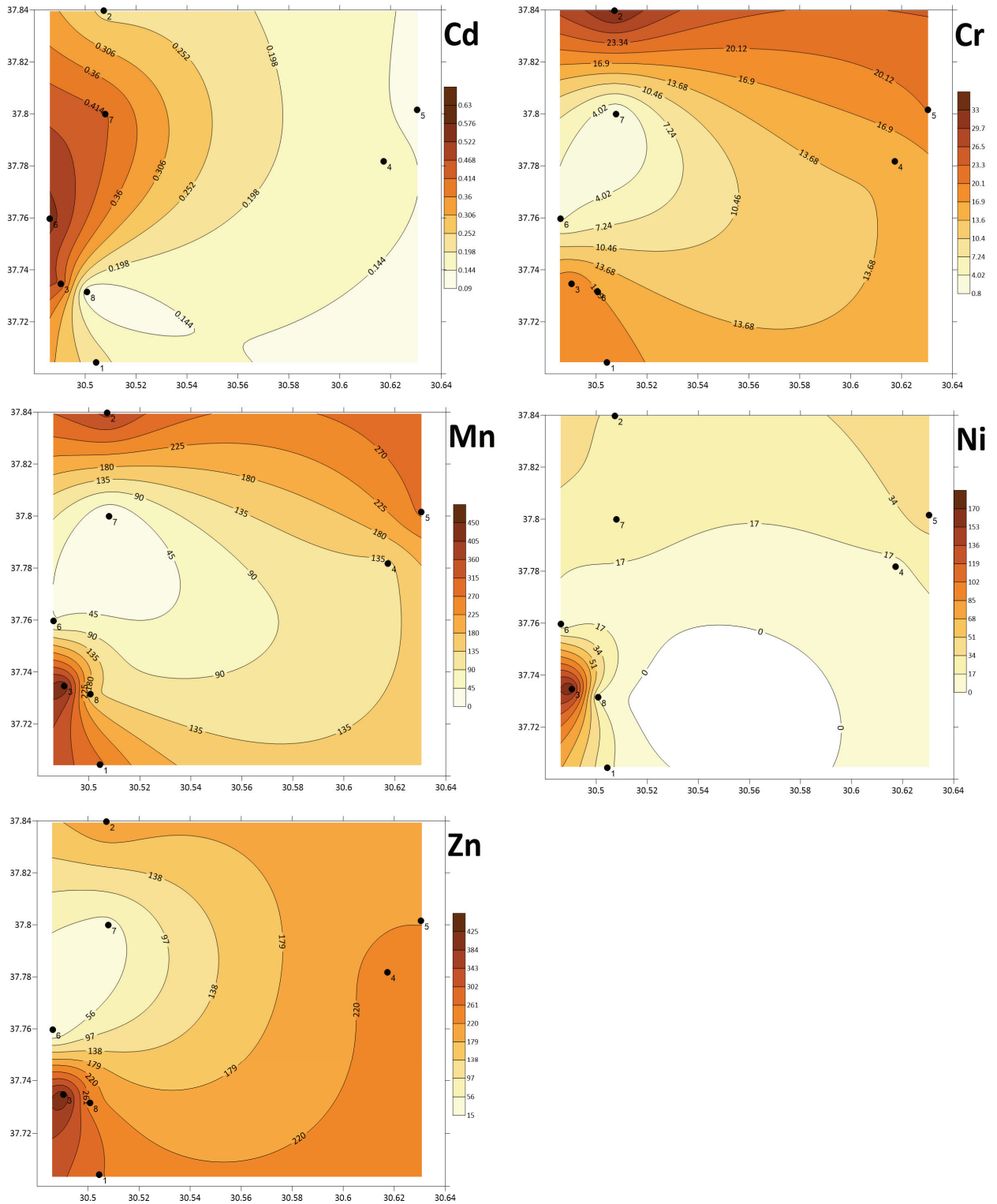


Figure 4. Spatial Distribution of Cd, Cr, Mn, Ni, and Zn concentrations in studied locations.

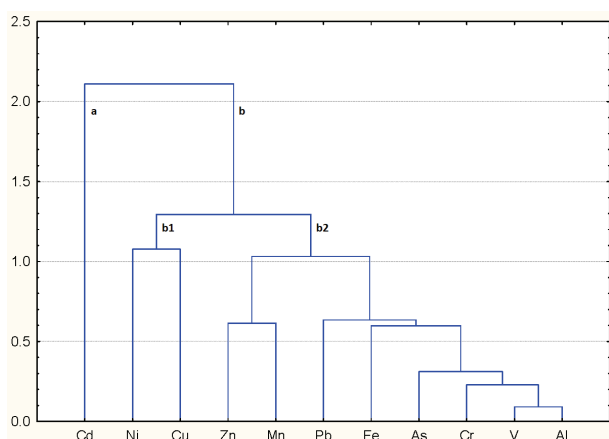


Figure 5. Dendrogram of elements in lichen samples.

evaluate air quality especially in the industrial centres and in the cities heated by coal combustion like Isparta, where heavy metal deposition can be severe.

Cluster analysis resulted in two main clusters of elements as given in Fig. 5. *Cluster a* consists of only Cd, while *cluster b* consists of the rest of the elements. The elements given in *cluster b1* show a strong correlation with each other (Ni-Cu: $r = 0.95$) as well as *cluster b2* consisting of correlated elements (Al – Cr: $r = 0.98$, Al – Fe: $r = 0.98$, Al – V: $r = 0.99$, As – Fe: $r = 0.96$, Cr – Fe: $r = 0.99$, Cr – V: $r = 0.97$, Cu – Ni: $r = 0.95$, Fe – V: $r = 0.97$, $p < 0.01$). Cluster analyses results of the elemental data indicate a strong correlation of the major 3 contaminants in the urban area, which are Ni, V and Cr.

As mentioned in the introduction of this paper, the results of air quality monitoring in Isparta between August 2006 and June 2010 by local authorities are given in terms of SO₂ (Fig. 6, İÇİM 2010) and PM₁₀ (Fig. 7, İÇİM 2010). Such monitoring studies can be used as references to

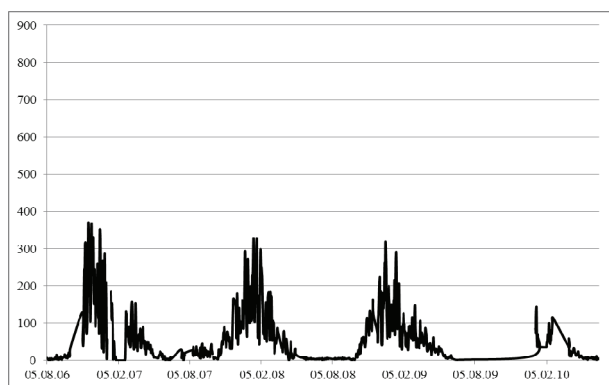


Figure 6. Annual SO₂ Data (2006-2010).

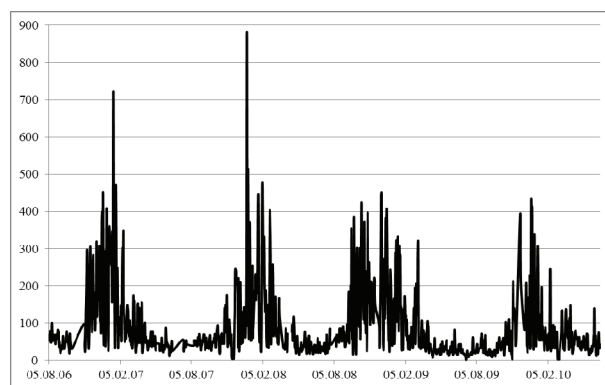


Figure 7. Annual PM₁₀ Data (2006-2010).

In the Isparta city centre, the mean concentration of SO₂ in winter seasons is 122 $\mu\text{g}/\text{m}^3$ while that of PM₁₀ is 91 $\mu\text{g}/\text{m}^3$. The levels of SO₂ concentration have been decreasing since the local authorities control the quality of coal combusted in the city. Both figures indicate that, though decreasing slightly from 2006 to 2010, there still is a considerable concentration of SO₂ and PM₁₀ in the atmosphere, which in the background corresponds to the outcome of analytes given in this study.

The most effective factors determining the extent of pollution include height, wind direction, humidity and temperature, and distance to the source of pollution (ÇOBANOĞLU, 2015). Due to the topographic peculiarities of the locations, particles may have been blown by the prevailing wind, in the study area. Airborne elemental pollution in Isparta, which is higher than expected, seems to be associated with the extensive use of coal as fossil fuel, besides the topographic and climatic characteristics of the city. Therefore, the geographical location, position, and other topographic features of the city and the localities are crucial when discussing the results of the survey. Considering Isparta, the localities in South-Southwestern parts have higher altitudes than those in North and East parts. The prevalent winds blowing in the region sweep airborne particles from SW (9 m/s), and from S (8.1 m/s) directions towards North-western, Northern and North-eastern parts of the city. Consequently, this wind-sweep causes the North-eastern part be contaminated more than the other parts of the city due altitude.

According to the data given in Table 1, in locality 2 (Behind Campus, Koçtepe Village, 1118 m) Al (11,294.94 $\mu\text{g}/\text{g}$), As (7.76 $\mu\text{g}/\text{g}$), Cr (33.81 $\mu\text{g}/\text{g}$), Fe (8,733.10 $\mu\text{g}/\text{g}$) and V (62.31 $\mu\text{g}/\text{g}$); in locality 3 (Gölcük Nature Park, Picnic Area, 1385 m), Cu (65.80 $\mu\text{g}/\text{g}$), Mn (442.70 $\mu\text{g}/\text{g}$), Ni (169.13 $\mu\text{g}/\text{g}$) and Zn (420.12 $\mu\text{g}/\text{g}$) have maximum concentrations. In locality 6 (Gülbirlik Rose-oil Factory, 1215 m) Cd (0.55 $\mu\text{g}/\text{g}$); and in locality 5 (Küçük Hacılar Village, 972 m) Pb (13.48 $\mu\text{g}/\text{g}$) has maximum concentration.

The lichen specimens accumulated maximal concentrations of Cd, Cu, Mn, Ni, Zn in locality III (inside Gölcük Nature Park) and next to GNP (locality VI), and the rest of the elements in northern and the eastern localities surrounding the city. The high concentration of these elements in the mentioned locations indicate that the *Xanthoria parietina* specimens collected from these locations may have been contaminated by charcoal or dust. Origin of the emissions may be charcoal used in barbecue, and cars driven on the unimproved road. The elemental concentration

detected in this locality is not affected directly from the urban area due the difference in the altitude and higher hills separating the urban area and the natural park, which means that localities III and VI have their local emission sources.

In locality II, airborne elements may origin from villages and exhaust gas from cars driven on the unimproved road, and also the airborne elements may have been blown by the prevailing wind, in the study area, from South-east to North-West, sweeping through the urban area. Since natural gas is not widely used in the centre, coal-fired heating is the dominant factor of emissions.

The comparison of average values obtained in this study with those of previous biomonitoring studies on *Xanthoria parietina*, in İzmir (YENISOY-KARAKAŞ & TUNCEL 2004), in İstanbul (İÇEL & ÇOBANOĞLU 2009) and in Kocaeli (DOĞRUL et al. 2012) to the reference material IAEA-336 ratios is given in Table 3. In the below table, the asterisk (*) indicates that since there is no data available in IAEA-336 certified list, results of Ni in previous studies have been compared to that of IAEA-336 reference material (*Evernia prunastri*) in this study.

Table 3. Comparison of *Xanthoria parietina* airborne elements' concentration to IAEA-336 ratios.

	İzmir (2004)		İstanbul (2009)		Kocaeli (2012)		Isparta present study		IAEA-336 Certified
	Average	Average to IAEA-336	Average	Average to IAEA-336	Average	Average to IAEA-336	Average	Average to IAEA-336	
Al	3,160.00	4.65	---	---	---	---	6,206.45	9.13	680.00
As	4.50	7.14	---	---	2.63	4.17	6.08	9.65	0.63
Cd	0.30	2.56	0.35	2.99	0.93	7.95	0.22	1.88	0.12
Cr	9.90	9.34	21.39	20.18	11.73	11.07	20.62	19.45	1.06
Cu	---	---	45.24	12.57	19.19	5.33	24.89	6.91	3.60
Fe	2,360.00	5.49	---	---	3,686.00	8.57	5,539.61	12.88	430.00
Mn	60.00	0.95	150.43	2.39	152.00	2.41	274.07	4.35	63.00
Ni	---	---	17.07	6.96	7.47	3.05	48.42	19.76	2.45*
Pb	---	---	71.35	14.56	70.20	14.33	10.77	2.20	4.90
V	6.40	4.35	---	---	10.67	7.26	30.81	20.96	1.47
Zn	100.00	3.29	194.87	6.41	280.10	9.21	265.77	8.74	30.40

Compared to the previous studies, the present study shows significantly greater degrees of contaminations by means of average to IAEA-336 ratios. For instance, Cr show 19.45 times, Fe 12.88 times, Ni 19.76 times and V 20.96 times magnitude of contamination when compared to the IAEA-336 reference material. Having in consideration that İzmir, Kocaeli and Istanbul are 3 major cities in Turkey by means of population, vehicles, accommodation and industry, Isparta shows a great magnitude of contamination of airborne elements (Fig. 8). This comparison of previous studies in bigger cities indicates that, the airborne elemental pollution in Isparta city is higher, due to the extensive use of coal as fossil fuel.

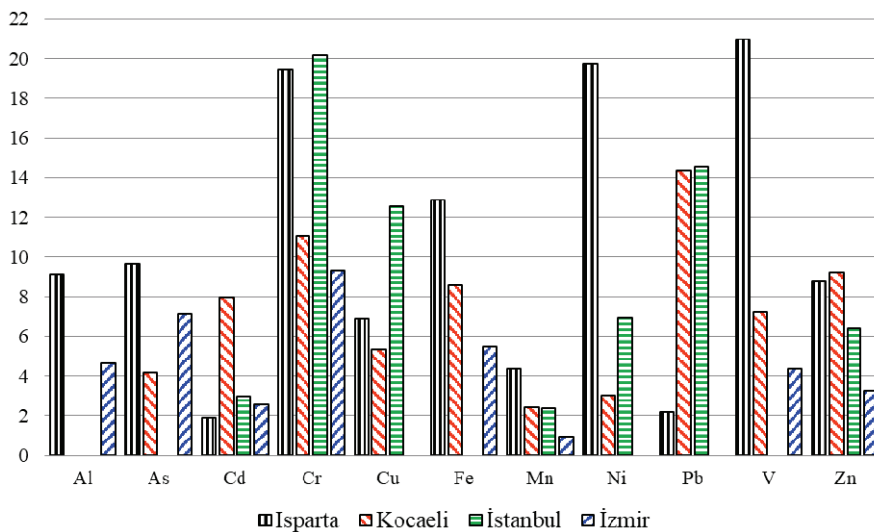


Figure 8. Comparison of average airborne elements' concentration to IAEA-336 ratios.

Compared to the concentrations of elements generally trapped (Ni > Cr > V > Fe > As > Al > Zn > Cu > Cd > Mn > Pb in descending order) by the lichen *Physcia aipolia* in the same region (YAVUZ & ÇOBANOĞLU, 2019), *X. parietina* accumulated more V than Cr; more Cu than Al and Zn; and more Mn than Cd in the present study (Ni > V > Cr > Fe > As >

Cu > Al > Zn > Mn > Cd > Pb in descending order). For differences in element contents, morphological differences of lichen species can be thought of as a reason, since *X. parietina* is larger lobed and less rough than *P. aipolia*.

CONCLUSIONS

In Isparta this is the first biomonitoring study using *Xanthoria parietina*. Thus, according to the YAVUZ & ÇOBANOĞLU (2019) study (based on *Physcia aipolia* (Erh. ex Humb.) Fűrnr.), this study lays a database for future biomonitoring studies in the region. The airborne element deposition in the urban vicinity of Isparta and Gölcük Nature Park investigated through a biomonitoring organism indicates that the degree of elemental deposition is severe in some locations of the city, especially in terms of Cr, Fe, Ni and V elements.

Considering the contamination levels in GNP, this study advises a precise and careful landscape use in the park, if necessary, a restriction or limitation of human activities to ensure minimum anthropogenic effects, for a sustainable environment. As mentioned by STEINHARDT et al. (1999), to prevent environment degradation, it is important to monitor the changes and to evaluate the impacts in a long-term investigation, as a future study. With regard to contamination levels in the urban area, this study indicates the necessity to use technology on renewable energy or natural gas for heating in order to decrease the concentration of airborne elements in the atmosphere.

Air quality in Isparta has been evaluated based on the levels of SO₂ and PM₁₀, as reported by the local authorities. However, in the regions with dense industrial activities and coal combustion, surveying only these two parameters to determine the air quality can result in the neglect of the *biomonitoring* part. A more rigorous approach - like biomonitoring of lichens - is required. In this view, further results can be achieved and long-term effects can be observed instead of instant evaluations.

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