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ORIGINAL ARTICLE

A Comparison of Bird Feathers and Plant Leaves as a Biological Tool for Assessing Air Pollution by some Heavy Elements

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INTRODUCTION

Air pollution in urban cities is one of the biggest problems worldwide in both developed and emerging nations [1, 2]. Vehicle traffic, wood burning, power stations, smoke, steel factories, vegetal parts and cooking exhausts are considered sources of air pollution[3]. In urban areas, vehicular activity is the main cause of pollutants and traffic operations produce aerosols, particulate matter and heavy elements in the urban atmosphere [4, 5]. In recent decades, the reliance has increased on the use of living organisms as biomonitors because they provide more clear pictures of the level of pollutants in the environment because they are in direct contact with these pollutants and their continuous interaction with the them [6]. Biomonitors are a free alternative for monitoring air quality and collecting information about the sensitivity of the populations to air pollution, allowing several different sampling areas to be concurrently analyzed [7, 8] . Many organisms have been tested for biological monitoring of pollutants such as heavy elements. However, to be accepted as a biological monitor for air quality, an organism must meet certain general and

specific requirements. It is also important that the organism cannot absorb appreciable amounts of elements from sources other than atmospheric^[5, 8]. Therefore, studies have been conducted on many organism's species for selection of appropriate species for monitoring environmental pollution in urban environments [9]. Most researches have focused on plant species for monitoring air pollution [6, 10]especially deciduous and evergreen plants [9, 11] and very few of them on bird species [12-14]. Heavy elements contamination is a major ecological and public health hazard around the world [15]. The danger of heavy elements lies in their resistance and tendency to accumulate in ecosystems, especially biological tissues, as well as biomagnification in food webs that lead to stress and poisoning of living organisms [16, 17]. Heavy elements can cause chronic or acute human diseases, most notably osteoporosis, kidney failure, lung cancer, and heart failure. Heavy elements also affect the nervous system, causing headaches, epilepsy, and sometimes coma. These elements inhibit some important reactions or stimulate harmful reactions within the body [18, 19]. A recent study indicated a significant correlation between the concentration of heavy elements in the bark of plants and data on morbidity and mortality related to the respiratory system and heart disease [20]. One of the toxins in the air of big cities are heavy

elements. Due to their accumulative property, it may reach the human body system via the inhalation of contaminated air and trigger many health problems [21]. Despite the importance of periodic monitoring to evaluate the level of heavy elements, local studies are still very few and insufficient. Most of them have been interested in studying the distribution and concentration of heavy elements in river water and soil surrounding industrial sites. Therefore, the current study aims to evaluate the concentration of heavy elements in the air of Kerbala governorate and to find the most effective biological tool for monitoring it through a comparison between the use of bird feathers and plant leaves.

MATERIALS AND METHODS

Study area

The holy governorate of Kerbala is the most important tourist city in Iraq, as nearly 20 million visitors flock to it annually. It has an area of 5034 km^2 , and it is 106 km away from the capital, Baghdad. Five different and distinct areas have been selected within the Kerbala governorate, as shown in Table 1. These areas differ in terms of human and industrial activities and the number of vehicles and the rural area was considered as a reference area.

Total suspended particulate (TSP) collection and analysis

In order to estimate the element of lead, cadmium, chromium, and nickel in the air, total suspended particulate (TSP) samples were collected during May, June and July (summer season) of the year 2019 at a rate of once per week in all areas using Portable Dust Sampler (Type L60 240V) at a height of 1.5 m from the ground with airflow rate of 60 L/min. The filters were then digested with a mixture of nitric acid and perchloric acid in a ratio of 4: 1 respectively. The heavy elements concentration was measured and calculated in μ g.m⁻³following the steps described by CPCB

[22]. The concentrations of total suspended particulates (TSP) were calculated according to the following equation:

Where:

$$
TSP\ Con. = \frac{(W2 - W1)}{TVA} \tag{1}
$$

 $W2$ = weight of the filter after suction (μ g) $W1$ = weight of the filter before suction (μ g) TVA = total volume of air flow (m^3)

As for the concentrations of heavy elements in a cubic meter, they were calculated according to the following equation:

Heavy Element Con. (µg. m $-$ 3) $=$ $\frac{(1-\epsilon)}{2\epsilon}$ V *(2)*

Where:

C = element concentration (μ g m⁻³). $Ms = element concentration (\mu g mL^{-1})$ $Mb = concentration of blank (µg mL⁻¹)$ $Vs = total volume of extraction (mL)$ Fa = total area of exposed filter $(cm²)$ V = volume of air sampled (m^3) Ft = filter area taken for digestion $(cm²)$

Collection and analysis of bird feather

Thirty-five, adult pigeons (*Columba livia*) were captured using nets during the summer season at five areas. Ten feathers were separated from different parts of the bird's body. Then they were placed in polyethylene containers and transferred carefully to the laboratory. About one gram of each sample was extracted using a mixture of nitric acid and hydrogen peroxide 1:1 and heated to a temperature of 200°C and the resulting solution was filtered and diluted to 25 mL with deionized water [23].

Collection and analysis of plant leaves

The leaves of mature plants of *Conocarpus lancifolius* were collected during the same season at a height of 1.5 to 2 m carefully cut and preserved in containers of polyethene. Then it was transferred to the laboratory and dried at a temperature of 60 ° C. One gram of each sample was taken

and placed in 10 ml of nitric acid for a whole night, and then 4 ml of perchloric acid were added to it and heated until the digestion process was complete $[1, 2]$. Unwashed leaf and feather samples were selected based on the results of some studies to monitor air pollution [25-27]. Heavy elements concentration in suspended particulate, feathers, and leaves samples were measured by used atomic absorption spectroscopy (AAS). The samples were analyzed using the statistical software SPSS (version 24.0). One-way ANOVA followed by a Tukey's posthoc HSD test was used to compare study areas while a T-test was used to compare between bird feathers and plant leaves. Pearson correlation coefficient test was applied to find the relationship between heavy elements concentration in total suspended particulate (TSP) and both feathers and leaves.

RESULTS AND DISCUSSION

The concentration of heavy elements in TSP

The average concentrations of lead, cadmium, chromium, and nickel associated with total suspended particulates (TSP) with some descriptive statistics are shown in Table 2. The elements at the studying areas can be rated based on averages as lead > cadmium> nickel > chromium. However, the level of pollution differs in each of the five areas. The study recorded an increase in the concentrations of elemental lead in all areas and the highest average was 3.69 μ g.m⁻³, while chromium element was the least concentrated element with an average of $0.02 \mu g.m^{-3}$. The results showed the effect of the study areas on the level of concentrations of heavy elements, as the concentrations of all elements increased in industrial and urban areas with high human activity and decreased in rural areas, with statistically significant differences at a significant level of *P* ≤0.01 (Table 2). Many researches indicated that the main reason for the increase in concentrations of heavy elements in urban environments is industrial activities, especially transportation and factories [4, 28]. Also, the reason for the high concentration of lead in the air is due to the increase in the number of vehicles that still use leaded gasoline. While the reason for the low concentrations of elements in the rural area is due to the lack of industrial activities, in addition to that, plants play an important role in the deposition and absorption of air pollutants[9, 29]. The correlation coefficient showed a positive significant correlation between some elements in total suspended particulate (Table 6). Some researchers have indicated that the association between heavy elements is often linked to their sources [30, 31]. So, when the correlation between them is strong, they often originated from the same source.

Area type	Descriptive statistics	P _b	C _d	Cr	Ni	
	average	0.77a	0.33a	0.02a	0.05a	
Area 1	SD	0.24	0.19	0.03	0.03	
	Minimum	0.39	0.11	0.00	0.00	
	Maximum	1.12	0.71	0.09	0.10	
	average	1.03a	0.56 ab	0.04a	0.06a	
Area 2	SD	0.42	0.22	0.04	0.03	
	Minimum	0.29	0.21	0.00	0.01	
	Maximum	1.65	0.96	0.11	0.10	
	average	1.80a	0.62 ab	0.09 ab	0.10a	
Area 3	${\rm SD}$	0.76	0.29	0.06	0.04	
	Minimum	0.63	0.33	0.02	0.05	
	Maximum	2.93	1.18	0.22	0.18	
	average	3.69 _b	0.98c	0.12bc	0.38 ab	
Area 4	SD	0.95	0.33	0.06	0.43	
	Minimum	1.84	0.48	0.05	0.05	
	Maximum	5.00	1.41	0.22	1.08	
	average	3.18 b	0.79 _{bc}	0.19c	0.66 _b	
	SD	1.28	0.24	0.08	0.51	
Area 5	Minimum	0.87	0.44	0.08	0.11	
	Maximum	5.04	1.30	0.32	1.56	
	P value	$0.000*$	0.000	0.000	0.000	
	(ANOVA)					
	Overall Average	2.09	0.66	0.09	0.25	

Table 2. Descriptive statistics of heavy element concentrations μ g.m⁻³in suspended particulate (TSP) from the five collection areas.

NOTE * *P* value is significant. Similar letters in columns indicate the significant differences between the study areas, while the different letters indicate significant differences using the Tukey test at level P≤0.05.

The concentration of heavy elements in bird feathers

Table 3 represents the concentrations of heavy elements in pigeon feathers. The current study recorded an increase in the concentration of the element of lead in the bird feathers and the highest average of it was 6.39 μ g g⁻¹ in the urban area, while nickel was the least accumulated element and the lowest concentration of it was 0.27 μ g g⁻¹ in the rural area. The order of the elements based on their concentration in the bird's feathers was as follows: lead $(5.00 \text{ µg g}^{-1}) >$ chromium (0.34 µg g^{-1}) = nickel (0.34 µg g^{-1}) > cadmium $(0.33 \mu g g^{-1})$. (Table 3). The results show that the concentration of lead is several times higher in bird feathers compared to other elements. The main source of lead can be what is deposited and incorporated into the feathers from outside, such as air and soil (external source). While the source of other elements is the result of internal absorption (internal source), birds remove harmful concentrations of toxic elements that enter with water and food and isolate them in the feather tissue during its formation [32]. This is reinforced by the presence of a moderate positive correlation ($r = 0.404$; $P < 0.01$) between the concentration

of lead in suspended particulates and feathers. While there was no significant correlation between the concentration of cadmium, chromium, and nickel in suspended particulates and bird feathers. Despite the high concentrations of the studied elements in the urban and industrial area and the decrease in the rural area, the results of the statistical analysis did not show any significant differences between the study areas for all elements except for the lead element $(P \leq 0.01)$. The reason for this could be the movement of birds and that their habitat and daily activity are wider than the study areas. In other words, all study areas constitute an ecological habitat that birds use to carry out their daily activity, and therefore no significant differences appeared between the study areas. (Table 5) Some research has indicated that the pigeon travels distances of up to 25 km per day[33], and these distances vary according to the availability of food and weather condition [34].

Concentration of heavy elements in leaves of plants

In contrast, the results showed an increase in the concentrations of all studied elements in plant leaves by several times compared to bird feathers, with significant differences between the concentration of elements in bird feathers compared to plant leaves for each area using the Ttest. (Table 5). The order of the elements based on their concentration in the leaves of the plant was as follows: lead

(67.66 μ g g⁻¹) > nickel (10.55 μ g g⁻¹) > chromium (4.39 μ g g^{-1}) > cadmium (3.66 µg g^{-1}). The highest recorded average of lead was 89.36 μ g g⁻¹ in the urban area, while the lowest average of cadmium was 1.71 μ g g⁻¹ (Table 4), with statistically significant differences ($P \le 0.01$) between most of the study areas for all elements using the ANOVA test. The higher concentrations of these elements in plant leaves

compared to bird feathers can be attributed to the ability of plant leaves to retain and accumulate these elements from the atmosphere. Based on the results of the current study, unwashed plant leaves can be considered a more efficient and effective biological tool in biomonitoring of air pollution with heavy elements from bird feathers. What confirms this is what was shown by the analysis of the correlation coefficient, as it found a strong positive significant correlation between the concentration of both lead ($r = 0.764$; $P \le 0.01$) and chromium ($r = 0.702$; $P \le$ 0.01) in suspended particulates and their concentrations in plant leaves and a moderate correlation of cadmium. (*r* = 0.676, $P \le 0.01$), as shown in Table 6. This averages that the increase in the concentration of these elements in the air, followed by the increase in their concentrations in plant leaves. While there was no significant correlation between the concentration of elements in bird feathers and their concentration in suspended particulate (TSP) except for element lead ($r = 0.404$; $P \le 0.01$) (Table 6). Therefore, can be believed that pollution from endogenous sources (food

or water) play a more important role in the accumulation of heavy elements in feathers than pollution from exogenous sources (air). The efficiency of plant leaves in biomonitoring compared to bird feathers can be attributed to the fact that plants are fixed, non-moving creatures, unlike birds that can move and fly away from sources of pollution when exposed to it. In addition, the leaves have a distinctive surface coating that is able to capture and retain pollutants [9, 35]. The stomata on the surface of the leaf play an important role by the processes of gas exchange and transpiration in the accumulation of pollutants on/or in the leaves [36-39]. And this is what bird feathers lack. Furthermore the bird alternates bouts of washing with those of strongly shaking the body to allow the water out of the feathers [40]. Therefore, dust and some contaminants attached with feathers will be removed. Some studies indicated that separating between external (dust, air) and internal (food) pollution is difficulty when bird feathers are using as bioindicators [41].

-1

	Elements											
Type area	Pb			C _d		Cr			Ni			
	Birds	Plants	P value $(T - test)$	Birds	Plants	P value $(T-test)$	Birds	Plants	P value $(T-test)$	Birds	Plants	P value $(T-test)$
Area 1	3.67a	42.03a	$0.000*$	0.29a	1.71a	$0.000*$	0.28a	2.45a	$0.000*$	0.27a	8.15a	$0.000*$
Area 2	4.60ab	54.28b	$0.000*$	0.32a	3.07ab	$0.000*$	0.31a	3.72ab	$0.000*$	0.31a	8.81a	$0.000*$
Area 3	4.98ab	69.47c	$0.000*$	0.35a	4.07bc	$0.000*$	0.33a	4.26ab	$0.000*$	0.31a	9.83ab	$0.000*$
Area 4	6.39b	89.16d	$0.000*$	0.34a	4.86bc	$0.000*$	0.39a	5.09bc	$0.000*$	0.37a	12.22 bc	$0.000*$
Area 5	5.38ab	83.36d	$0.000*$	0.36a	4.59c	$0.000*$	0.40a	6.40c	$0.000*$	0.44a	13.75c	$0.000*$
P value (ANOVA)	$0.020*$	$0.000*$		0.953	$0.000*$		0.662	$0.000*$		0.245	$0.000*$	

Table 5. Average dry weight and element concentrations µg g⁻¹ of unwashed feathers and leaves from the five collection areas based on T-TEST and ANOVA.

NOTE * *P* value is significant. Similar letters in columns indicate the significant differences between the study areas, while the different letters indicate significant differences using the Tukey test at level P≤0.05.

Table 6. Correlation coefficient between concentrations of heavy elements in suspended particulates (TSP), plant leaves and bird feathers.

** Correlation is significant at the 0.01 level; * Correlation is significant at the 0.05 level

CONCLUSIONS

The results of the research indicate a high level of lead, cadmium, chromium, and nickel concentrations in the total suspended particles, especially lead, in urban and industrial areas compared to the reference area. The current study revealed the source of pollution with heavy elements are human and industrial activities and vehicles. The presence of these elements in high concentrations in the air portends

great environmental and health risks, so the government should take strict measures to reduce the level of pollutants, prevent the addition of lead to fuel, and monitor air pollution periodically. Research investigations showed that the use of plant leaves in assessing and monitoring air pollution with heavy elements was more efficient and capable of detecting pollution status and intensity in the

environment compared to bird feathers. This belief is reinforced by the presence of significant differences between the study areas for all elements. The correlation coefficient also showed a significant correlation between the concentration of each of lead, cadmium, and chromium in the air and their concentrations in the leaves of plants. The efficiency of plants is due to the fact that they are stable organisms in their place that are directly and continuously exposed to the pollutants of each area, as well as their interaction with the air environment through the processes of respiration and transpiration. The leaves also have a distinctive surface coating that has the ability to retain and accumulate high amounts of pollutants. The birds did not show a high sensitivity to heavy elements, except for lead, as there were no significant differences between the study areas. The reason could be the movement of birds and their distance from sources of pollution and the inability of the feathers to accumulate and trap air pollutants compared to the leaves of plants, as well as the birds doing the washing and cleaning operations that cause the removal of pollutants suspended in the feathers. The current study recommends that the use of unwashed leaves of plants is preferred in monitoring air pollution by heavy elements and that the use of bird feathers is not preferred.

Conflicts of Interest

In this study, there are no conflicts of interest to declare.

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