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water

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# Analysis of Heavy Metal Contamination in Macroalgae from Surface Waters in Djelfa, Algeria

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**Abstract:** The heavy metals concentration in algae can be used as a bioindicator of the pollution of an ecosystem. In the present study, we determined the levels of the elements Fe, Pb, Cu, and Zn in chlorophyta of the species *Cladophora, Spirogyra, Chara, and Zygnema*. The samples were collected in February of 2021 in three different locations in Algeria (Dar El Chioukh lake, Oued Mellah, and Oued Boucedira). Our results showed that all four algae species accumulated high levels of heavy metals in surface water, with concentrations ranging from 3.14 to 5600 mg/kg. The Pb levels in all four species exceeded the recommended standard set by the International Atomic Energy Agency (IAEA) of 0.574 mg/kg, with the highest concentration of 50.85 mg/kg recorded in *Cladophora* at the first location. Similarly, Fe and Cu levels were found to be above the recommended standard, with maximum concentrations of 5600 mg/kg and 82.5 mg/kg, respectively. On the other hand, Zn levels were found to be lower than the standard of 128 mg/kg, with the highest concentration of 47.5 mg/kg recorded in *Spirogyra* at the third location. The results of the principal component analysis (PCA) confirmed that the heavy metal pollution of these waters was of anthropogenic origin, likely stemming from urban, industrial, and agricultural activities.

**Keywords:** algae; heavy metals; pollution; water

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## 1. Introduction

Lakes and rivers serve as the principal sources of water for household, industrial, and agricultural operations as a consequence of rapid urbanization and population growth. In many nations, the industrial, manufacturing, and agricultural sectors are the primary contributors to economic growth. These industries' products emit tons of waste containing metals into the ecosystem [1]. Chromium, cadmium, lead, copper, nickel, and zinc are the most frequent metals detected [2]. This has become a severe environmental issue, causing harmful impacts on flora and animals and even endangering human life. It should also be mentioned that some studies prefer the phrase "potentially toxic elements" to "heavy metals" [3] or propose developing a new categorization for them. We utilized the terminology "heavy metals" or trace metals (TMEs) in our study.

There are heavy metals, such as Fe, Cu, Zn, and Pb, that are essential for the metabolism of living organisms. Their detrimental impact is only detected when the quantity necessary

for metabolism implementation is surpassed. Furthermore, among the heavy metals, there are some that are very poisonous and have a harmful impact on living beings even in trace amounts [4].

Different countries are adopting their own limits for heavy metals to prevent the gradual poisoning of land and water, in accordance with World Health Organization recommendations [5]. In order to evaluate the toxicity of metals, the repercussions of their accumulation in ecosystems, and the nature of their influence on living creatures, bioindication and biotesting techniques have become more popular [6]. Bioindication and biotesting involve the use microorganisms, higher plants, animals, and algae [7].

Multiple species coexisting in an ecosystem may give resilience to environmental fluctuation, species stability, the capacity to share metabolites and weather periods of nutrient constraint, and resistance to invasion by other organisms. As a result, photosynthetic microorganisms that are prevalent in the environment and are sensitive to environmental changes may be useful indicators of heavy metals by giving biological reactions when these heavy metals come into contact with them. Algae are a vital component of both aquatic and terrestrial ecosystems and react rapidly, often preceding higher plants, to external factors, while exhibiting a distinct response to a given anthropogenic cause [8]. It is well known that the responsiveness of algae to metal action is determined by the specific properties of algae species as well as the metals themselves. Heavy metals' harmful impacts on algae are linked to the development of oxidative stress [9]. It was reported that copper, cadmium, and lead cause lipid peroxidation in cell membranes by attaching to the sulfhydryl of membrane proteins; metal ions such as Zn, Cd, Cu, Hg, Pb, and Ni may substitute the Mg ion in the chlorophyll molecule, hence inhibiting photosynthesis [10]. A change in metabolic pathways is found as a consequence of stress in the cells of algae. An increase in the synthesis of lipids, carbohydrates, and carotenoids by algal cells is indicative of defensive mechanisms that minimize the damage caused by heavy metals [9]. The diverse reaction of algae to the actions of various metals offers several options for their use in assessing the ecological status of ecosystems and determining the toxicity of metals [11].

According to Ramade [12], the development of bio-indication opens the door for broader ecological monitoring that incorporates the impacts on the environment via the use of sentinel species. Moreover, investigations performed directly on water for these kinds of pollutants have a high cost for a random efficacy, but analyzing such pollutants through the bioindicators of accumulation in plants might offer more relevant information at a lower cost.

Our research focuses primarily on the presence of some of the most harmful heavy metals (Cu, Fe, Pb, and Zn) in four types of chlorophyta (*Chara*, *Cladophora*, *Zygnema,* and *Spirogyra*). Three aquatic habitats were studied in February 2021: lake Dar El Chioukh, Oued Mellah, and Oued Boucedira in the district of Djelfa city, approximately 300 km south of Algiers in the highlands (Algeria).This analysis would then allow us to assess the degree of accumulation of the chosen TMEs in the tissues of the chlorophyta studied and established in two freshwater ecosystems in the Djelfa region, with the goal of finding adequate interventions to slow the deterioration of these environments and minimize the impact of metallic pollution.

## 2. Materials and Methods

### *2.1. Study Area*

Djelfa is situated in middle northern Algeria, beyond the southern foothills of the Atlas Tellien from the north, with the chief town of Wilaya 300 km south of the capital (Figure 1). It is situated between 2° and 5° east longitude and 33° and 35° north latitude, and has a total area of 32,256.35 km$^2$ (1.36% of the country's total area). It is now mainly composed of 36 municipalities divided into 12 Dairates [13].

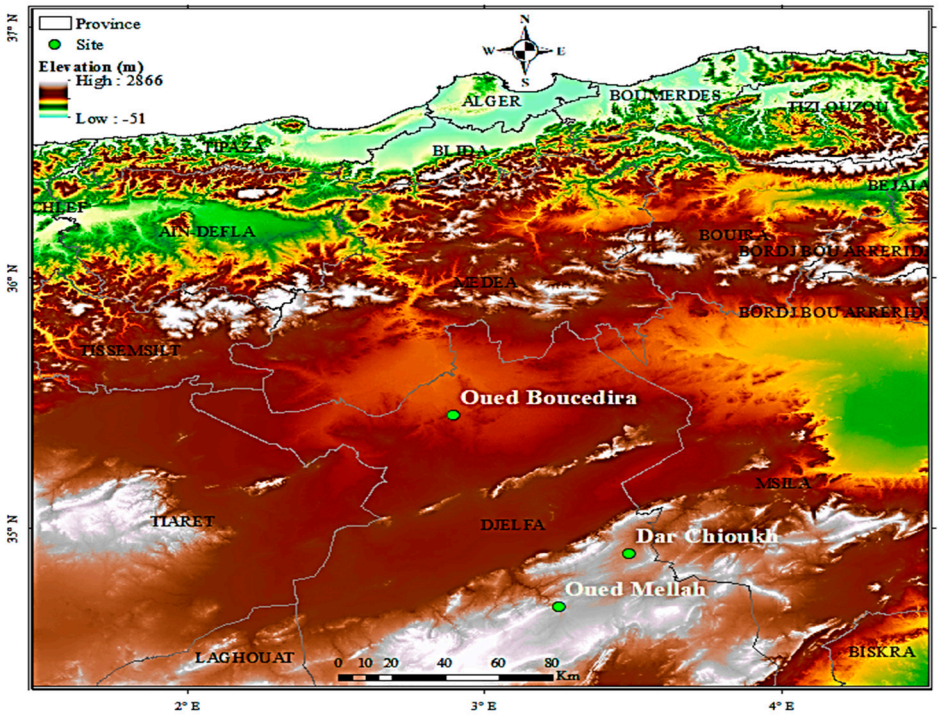



**Figure 1.** The research region and chosen sites including Oued Boucedira, Dar El Chioukh, and Oued Mellah.

### *2.2. Hydrology*

The hydrographic network in this area is quite dense, extending over all reliefs. The direction of the major rivers varies depending on the terrain, although it is commonly perpendicular to them, running north–south to north-west–south-east (for example, the wadis M'zi, Messad, El Djorf, and so on). It may, however, be in accordance with these reliefs since affluent valleys formed inside depressions are caused by relief inversion, as in the case of the combes of Djebels Lazreg and Tebag in the Southwest of Fernane and the perched syn-clinals of Bou Kahil of Djebel Zerga and Djelfa (Figure 1). Furthermore, the majority of the wadis in this dry to semi-arid environment flow only when it rains [14].

### *2.3. Climate*

The region of Djelfa is characterized by a semi-arid to arid climate with a continental nuance, with an average annual temperature of 14.32 °C. It is characterized by the existence of two seasons: a dry and hot summer season, and a cold winter season (Figure 1).

The three locations were selected for the sampling were:

- Site 01: the lake of Dar El Chioukh.
- Site 02: Oued Mellah.
- Site 03: Oued Boucedira (Figure 1).

The selection of these locations was based on the abundance of algae and their closeness to various wastewater discharges and agricultural and household waste releases.

### *2.4. Algae Sample Collection and Identification*

Samples were collected in February 2021. The hand-driven algae were cleaned of all epiphytes and detritus associated with their tillers, washed in situ with water, and then transferred to plastic bags. After reception at the laboratory, they were sorted by species. Specimen identification and description specimen identification began in the field and continued in the laboratory. The description was essentially based on observation with the naked eye, with a magnifying glass, or with an optical microscope (MOTIC-AE2000AE). Observation with the naked eye made it possible to identify all the most "accessible"

morphological characteristics, such as the color (pigment), the nature of the thallus, the shape and amplitude of the ramification of the thallus, and the size and the consistency of the thallus. Observation with a magnifying glass or under a microscope made it possible to see the fine characters, such as the sexual and asexual organs, the rhizines, and the structure of the thallus. In this work, the identification was based on the observation of the morphological characteristics by comparing the data obtained with other works, while being based on the following criteria: the type of ramification, the color, the consistency, the attachment organs, and the aspect of the thallus. Consultation of the WoRMS (World Register Marine Species) and Algea Base databases also facilitated the identification of taxa. All the species collected are described morphologically, photographed to build a photo library, and establish identification keys.

Among these harvested and identified microalgae, four algae were selected on the basis of the dominant species. All of these algae belong to chlorophycophytes (Figure 2). The four chlorophyta are filamentous taxa *Chara (Charophyceae), Cladophora (Cladophoraceae), Spirogyra*, and *Zygnema*, which constitute the group Zygnemataceae.

The four selected algae were washed again with tap water, then with distilled water, dried in the open air for 24 h, and finally dried in the oven at 40 °C. for 24 h. These algae were then ground into a fine powder which was later used.

### *2.5. Mineralization*

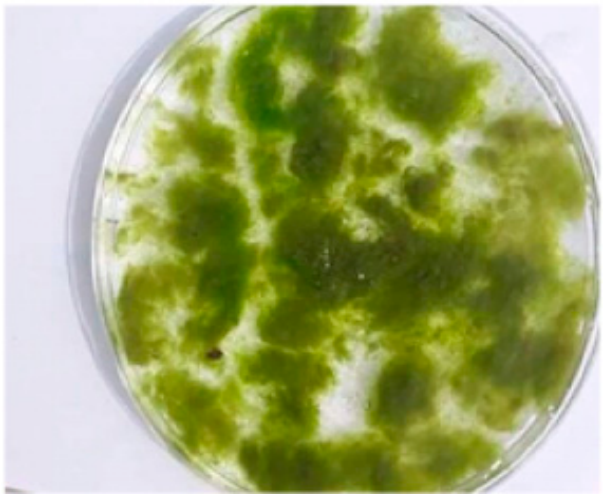
This stage was accomplished using the aqua regia technique, which is a combination of 3:1 hydrochloric acid and nitric acid that is often employed in mineral and organic chemistry. It permits metal dissolution and the attack of the remaining organic molecules [15]. The steps of mineralization are as follows: 1 g of the sample (powder) is taken and added to 1 mL of Nitric acid ($HNO_3$ 65%) and 3 mL of HCl (35–38%); the mixture is then heated under stirring to complete homogenization. This operation is repeated twice. A volume of 50 mL of distilled water is then added to each beaker. The resulting mineralization is filtered through a filter paper and atomic absorption spectrophotometry (UNICAM PYE UNICAM 969) is used to analyze the samples.

(a)

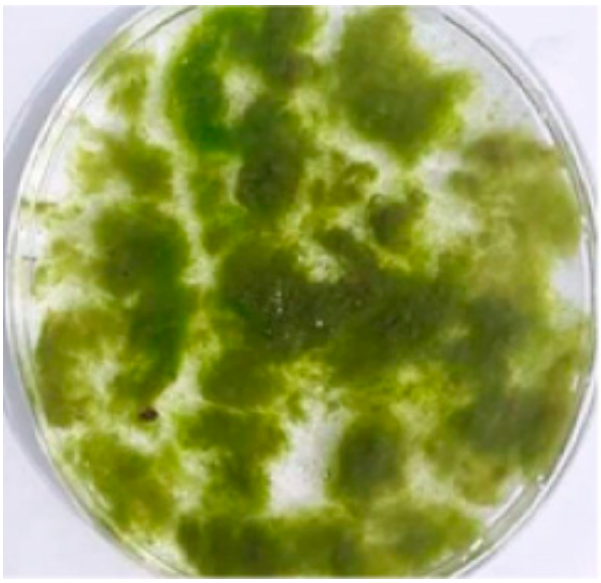


**Figure 2.** *Cont.*

(b)



(c)



(d)

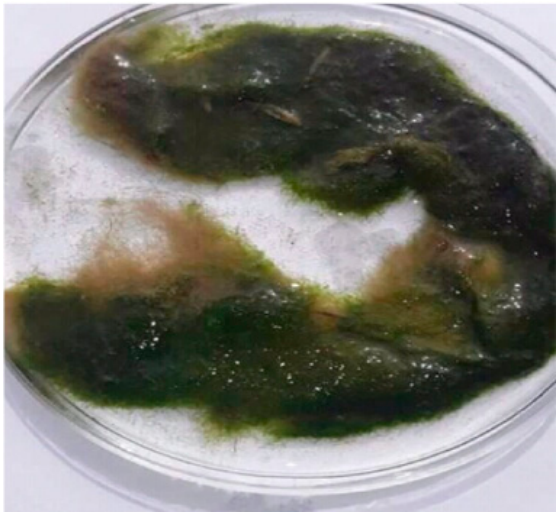


**Figure 2.** Chlorophyta: (**a**) *Chara*, (**b**) *Zygnema*, (**c**) *Cladophora*, (**d**) *Spirogyra*.

### *2.6. Selection of Contaminants*

Although heavy metals may have a natural origin (subsoil rocks, ores), they are mostly caused by the contamination of water by industrial discharges. They have the particularity of accumulating in living organisms and in the trophic chain [16]. These metals may accumulate in living species' tissues, causing significant harm to the whole food chain [17]. In addition, the accumulation of metals in algal cells can influence the formation of reactive oxygen species. At normal levels, they act as essential molecules to control algal metabolisms such as growth actions, cell death, and defense against pathogenic organisms. When the amount of metals is excessive, it can cause high oxidative potential and contribute to cell damage, which directly interferes with their cellular composition, altering protein and lipid levels and, in many cases, leading to cell death.

### *2.7. Statistical Data Processing Techniques*

The findings were statistically processed using the Excel stat program, version 2007. A Principal Component Analysis (PCA) on reduced centered variables is used in the descriptive statistics technique. The goal of this study is to examine the link between metal concentration and algae observed in surface waters. The use of multivariate exploratory methods (factorial analysis and ascending hierarchical classification) as reported by Belkhiri et al. [18] and Bencer et al. [19] allows for the identification of the most relevant parameters that describe wastewater quality and demonstrate their variability.

## 3. Results and Discussion

Our research contributes to the study of metal contamination by four heavy metals (Cu, Fe, Pb, Zn) in four chlorophyta (*Chara, Cladophora, Spirogyra*, and *Zygnema*), which were collected in the lakes of Dar El Chioukh, Oued Mellah, and Oued Boucedira, three aquatic environments in the city of Djelfa, in February 2021.

The average metal contents are expressed in mg/kg of the four metals (Cu, Zn, Fe, Pb) in algae (*Chara*, *Cladophora*, *Spirogyra,* and *Zygnema*,), in the three sites (Lake Dar El Chioukh, Oued Mellah, and Oued Boucedira) in Djelfa are reported in Table 1.

**Table 1.** Comparison of the values obtained with the limit values (International standards accepted by (IAEA, 2005)).

| | | Cu (mg/kg) | Zn (mg/kg) | Fe (mg/kg) | Pb (mg/kg) |
|---|---|---|---|---|---|
| **Values obtained in site 1** | *Chara* | 26 (±0.6) | 40 (±0.8) | 5600 (±3.7) | 48.1 |
| | *Zygnema* | 30.5 (±0.3) | 39 (±0.5) | 1485 (±2.8) | 30.9 |
| | *Cladophora* | 82.5 (±0.4) | 44.5 (±0.9) | 2051 (±4.4) | 50.85 |
| | *Spirogyra* | 32.5 (±0.8) | 39 (±0.4) | 772.5 (±3.2) | 4.65 |
| **Values obtained in site 2** | *Chara* | 28.5 (±0.4) | 38.5 (±0.8) | 918 (±2.7) | 45.25 |
| | *Zygnema* | 38.5 (±0.1) | 17.5 (±0.3) | 1490 (±3.9) | 34.75 |
| | *Cladophora* | 14.5 (±0.8) | 30 (±0.7) | 3030 (±3.4) | 40.55 |
| | *Spirogyra* | 13 (±0.7) | 36 (±0.7) | 1475 (±4.2) | 4.95 |
| **Norm Values obtained in site 3** | *Chara* | 27 (±0.2) | 39 (±0.9) | 1500 (±2.6) | 45.1 |
| | *Zygnema* | 19.5 (±0.3) | 21 (±0.5) | 1335 (±3.4) | 16,4 |
| | *Cladophora* | 19 (±0.7) | 35.5 (±0.4) | 1565 (±5.3) | 3.15 |
| | *Spirogyra* | 52.5 (±0.6) | 47.5 (±0.3) | 2295 (±2.9) | 50.35 |
| **NOR Standards (AIEA, 2005)** | *Algae* | 23.2 (±0.2) | 128 (±1.4) | 497 (±1.7) | 0.574 |

The bioaccumulation of the investigated metals seems to be different in the four species of algae tested from one location to another in the city of Djelfa, according to the findings. Metal deposition occurs primarily in algae in proportion to metal availability, organism size, ecology, and morphology, and the time of immersion and exposure of algae [20]. Because of anthropogenic pressures and the establishment of various industrial, economic, and tourist activities, the Oued Mellah, the region's main watercourse, has undergone and continues to undergo significant degradation as a result of industrial discharges and direct release of domestic waste water. The distribution of metallic elements in the algae investigated

reflected these degradations. Indeed, according to the findings of this study, the affinity for certain trace metals differs at the algal level. The accumulation also varies from one location to the other.

According to the various analyses, the amounts of Fe and Pb observed are rather significant in comparison to the other metals analyzed and exceed the requirements (Table 1). Road traffic, raw industrial discharge, and direct release of household wastewater may all explain these amounts [21]. The data demonstrate that Fe is the most abundant element in all of the species tested, with concentrations that exceed the permissible limits (IAEA, 2005). Its presence in all algae species investigated indicates that all three locations have iron contamination. Comparing the metal composition of the four species from the three locations, we found that the highest concentrations of the three metals (Fe, Pb, and Cu) are found in *Chara* for iron and in *Spirogyra* for Pb and Cu, with the exception of iron, which exhibits very high levels and surpasses IAEA standards in all four species, most notably in "*Chara*" at "site 1". The quantities of metals in various species of algae from the same biotope are observed to vary. In addition, the presence of trace amounts of heavy metals in large concentrations inside living species reveals the contamination of the aquatic environment.

In general, these data demonstrate that metal bioaccumulation varies across species and locations, as is the case with *Chara* and *Cladophora* [22]. The trace elements chosen for this study are among the most frequently encountered metal pollutants in soil: Cu, Zn, Fe, and Pb. However, Pb is toxic at high concentrations [23], despite being an element with low mobilization but high toxicity when assimilated by plants [15].

➢ ***Case of Cu***

The greatest value of 82.5 mg/kg was obtained at "site 1" of "*Cladophora*", above the IAEA-accepted limit of 23.2 mg/kg, while the lowest value was 13 mg/kg at "site 2" of "*Spirogyra*". For *Chara*: 28.5 mg/kg (site1) > 27 mg/kg (site3) > 26 mg/kg (site1); the levels exceed the standard (23.2 mg/kg). For *Zygnema*: 38.5 mg/kg (site2 )> 30.5 mg/kg (site1) > 19.5 (site3); the levels exceed the standard for site 1 and 2. For *Cladophora*: 82.5 mg/kg (site1) > 19 mg/kg (site3) > 14.5 mg/kg (site2); the levels in site 1 exceed the standard (23.2 mg/kg), For *Spirogyra*: 52.5 mg/kg (site3) > 32.5 mg/kg (site1) > 13 mg/kg (site2); the levels exceed the standard for site 1 and 3 (Figure 3).

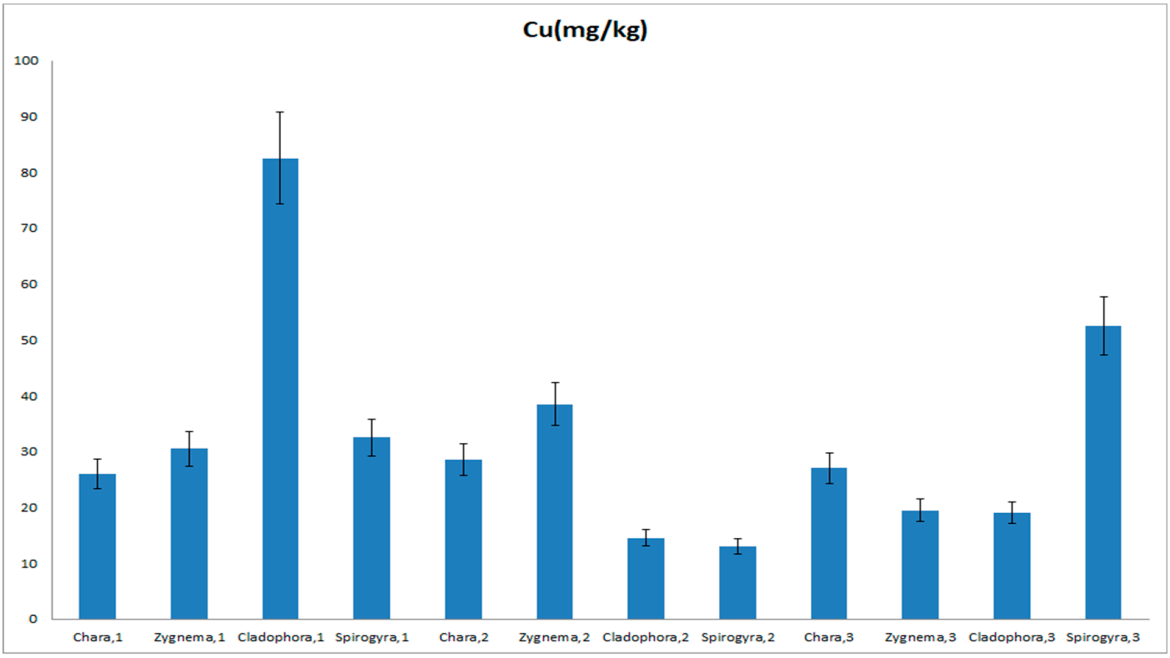



**Figure 3.** Cu contents of the four algae studied (for the three sites considered).

Cu data for site 1 indicated very high levels compared to the maximum dose authorized by the IAEA for all species investigated (Lake Dar El Chioukh), high values that exceed the limit in *Chara* and *Zygnema* for site 2 (Oued Mellah), and values significantly above the standard in *Chara* and *Spirogyra* for site 3 (Oued Boucedira). Some explanations have been proposed to explain these findings. First, since Cu is a necessary nutrient for organisms, it is naturally present in living organisms, and we have identified it in the species analyzed. Furthermore, it may be hazardous only at a certain concentration level [24,25].

➢ ***Case of Zn***

The greatest value is 47.5 mg/kg in "*Spirogyra*" at "site 3" (Figure 3), and the minimum value is 17.5 mg/kg in "*Zygnema*" at "site 2" (Figure 3); these reported values do not exceed the IAEA standard (128 mg/kg). For *Chara*:40 mg/kg (site1) > 39 mg/kg (site3) > 38.5 mg/kg (site2); levels do not exceed the standard (128 mg/kg) for the three sites. For *Zygnema*:39 mg/kg (site1) > 21 mg/kg (site3) > 17.5 mg/kg (site2); levels do not exceed the standard (128 mg/kg) for all three sites. For *Cladophora*: 44.5 mg/kg (site1) > 35.5 mg/kg (site3) > 130 mg/kg (site2); levels do not exceed the standard (128 mg/kg) for all three sites. For *Spirogyra*: 47.5 mg/kg (site3) > 39 mg/kg (site1) > 36 mg/kg (site2); levels do not exceed the standard (128 mg/kg) for all three sites (Figure 4).

These findings indicate that Zn is not a concern to the three locations. Different authors and species have already shown the same outcome. Zn accumulates faster than other heavy metals, such as Cu and Pb. These results are in agreement with those reported earlier [26,27]. Note that the algae "*Spirogyra*" accumulated more Zn than the algae *Chara, Zygnema* and *Cladophora*.

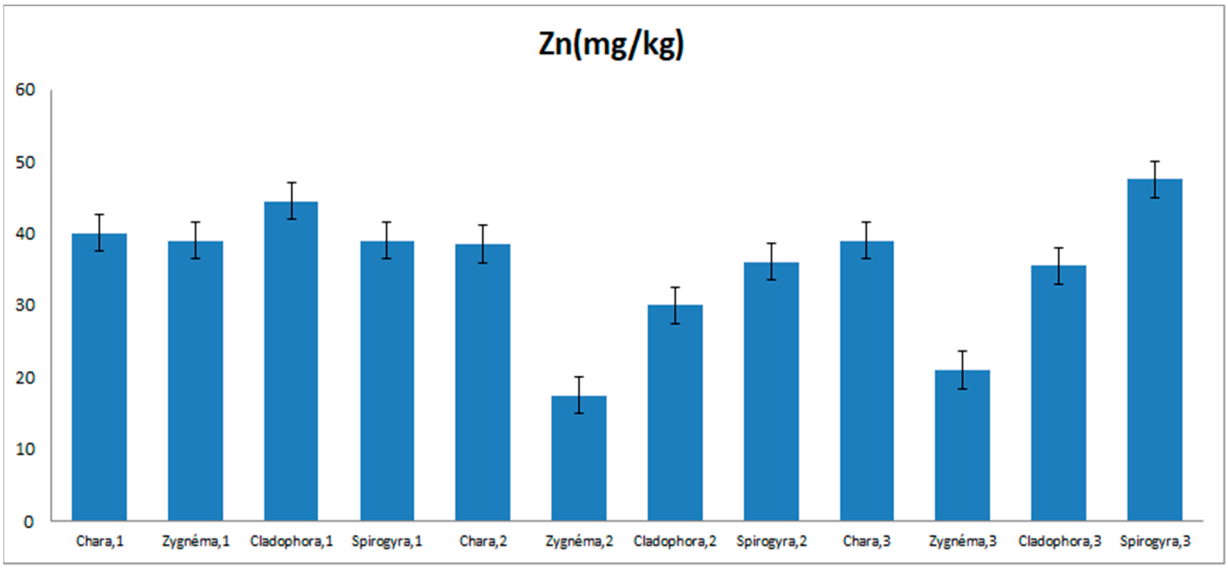



**Figure 4.** Zn contents of the four algae studied (for the three sites considered).

➢ ***Case of Fe***

We record a maximum value of 5600 mg/kg in "*Chara*" at "site 1" (Figure 5) and the lowest value of 918 mg/kg in "*Chara*" at "site 2"; these reported values far exceed the IAEA standard (497 mg/kg).

Our findings show that iron accumulates more than other heavy metals, including Zn, Cu, and Pb. It is worth noting that the algae "*Chara*" accumulated more Zn than the algae *Zygnema*, *Cladophora*, or *Spirogyra*. For *Chara*: 5600 mg/kg (site1) > 1500 mg/kg (site3) > 918 mg/kg (site2); levels exceed the norm (497 mg/kg) for the three sites. For *Zygnema*: 1490 mg/kg (site2) > 1485 mg/kg (site1) > 1335 mg/kg (site1); levels exceed the standard (497 mg/kg) for all three sites. For *Cladophora*: 3030 mg/kg (site2) > 2051 mg/kg (site1) > 1565 mg/kg (site3); levels exceed the standard (497 mg/kg) for all three sites. For Spirogyra:

2295 mg/kg (site3) > 1475 mg/kg (site2) > 772.5 mg/kg (site1); levels exceed the norm (497 mg/kg) for all three sites (Figure 5).

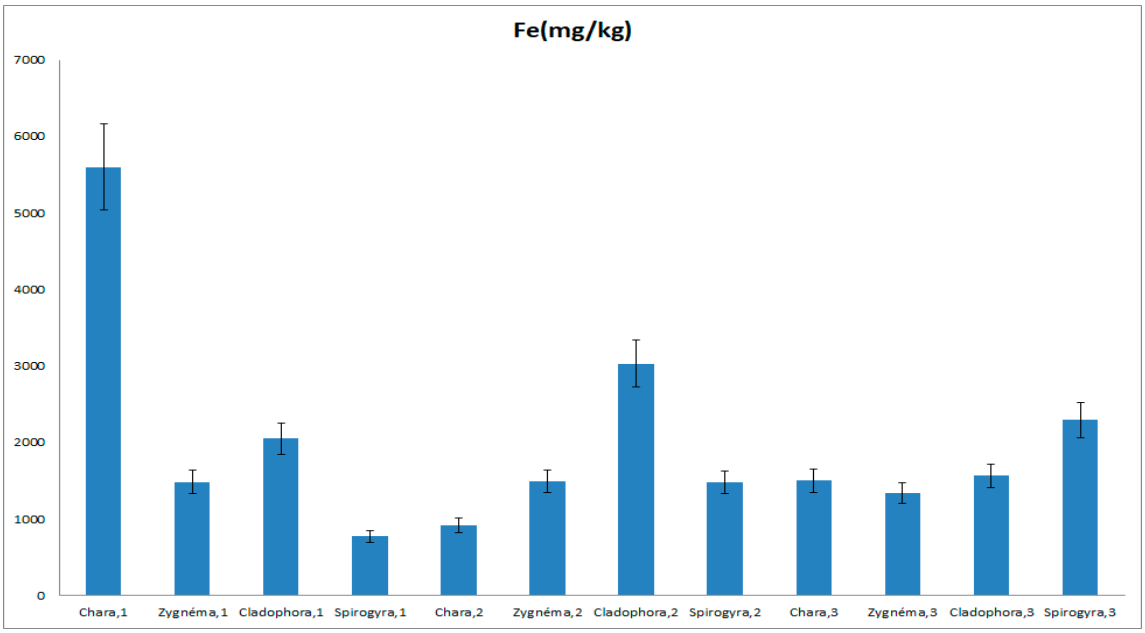



**Figure 5.** Fe contents of the four algae studied (for the three sites considered).

This metal is associated with the usage of fertilizers, which may explain its prevalence in algal tissues. We also observed that Fe is more easily assimilated by these algae than other metals (Cu, Zn, and Pb). We observed that all three locations are very iron-polluted as a result of fertilizers, raw industrial waste, and direct discharge of domestic wastewater.

➢ ***Case of Pb***

Figure 6 depicts the lead levels found in (Table 1) from samples collected at the three locations.

The greatest value is 50.85 mg/kg in "*Cladophora*" at "site 1", and the lowest value is 3.15 mg/kg in "*Cladophora*" at "site 3"; these reported values substantially exceed the IAEA limit (0.574 mg/kg).

Our findings show that Pb accumulates significantly in the four algae investigated. It is worth mentioning that the algae "*Cladophora*" has accumulated more Pb than the algae *Chara*, *Zygnéma*, or *Spirogyra*. For *Chara*: 48.1 mg/kg (site1) > 45.25 mg/kg (site2) > 45.1 mg/kg (site3); the levels exceed the standard (0.574 mg/kg) for the three sites. For *Zygnema*: 34.75 mg/kg (site2) > 30.9 mg/kg (site1) > 16.4 mg/kg (site3); the levels exceed the standard (0.574 mg/kg) for the three sites. For *Cladophora*: 50.85 mg/kg (site1) > 40.55 mg/kg (site2) > 3.15 mg/kg (site3); the levels exceed the standard (0.574 mg/kg) for the three sites. For *Spirogyra*: 50.35 mg/kg (site3) > 4.95 mg/kg (site2) > 4.65 mg/kg (site1); the levels exceed the standard (0.574 mg/kg) for the three sites (Figure 6).

This metal may be found in algal tissues. We also observed that *Cladophora* accumulates more Pb than other algae. We noticed that the three sites are significantly contaminated by Pb, which is caused by vehicle traffic and the release of fine metallic particles into the atmosphere, mostly in metropolitan areas, which are deposited on the ground and may reach the networks [26]. Indeed, metals generated by human sources are present in very reactive chemical forms, posing much greater dangers than metals derived from natural sources, which are often immobilized in relatively harmless forms [28]. The World Health Organization also emphasizes the substantial danger posed by inorganic Pb introduced into humans via drinking water [29]. Pb poisoning has multiple pathological consequences, which may be divided into two categories: physiological and neurological impacts [30]. The

former causes a rise in blood pressure, vascular and intestinal damage, and renal problems (Pb nephropathy). Pb may replace Ca in children's bones, which is readily detected using X-rays [31]. Finally, incidences of sterility have been reported after continuous Pb exposure [32].

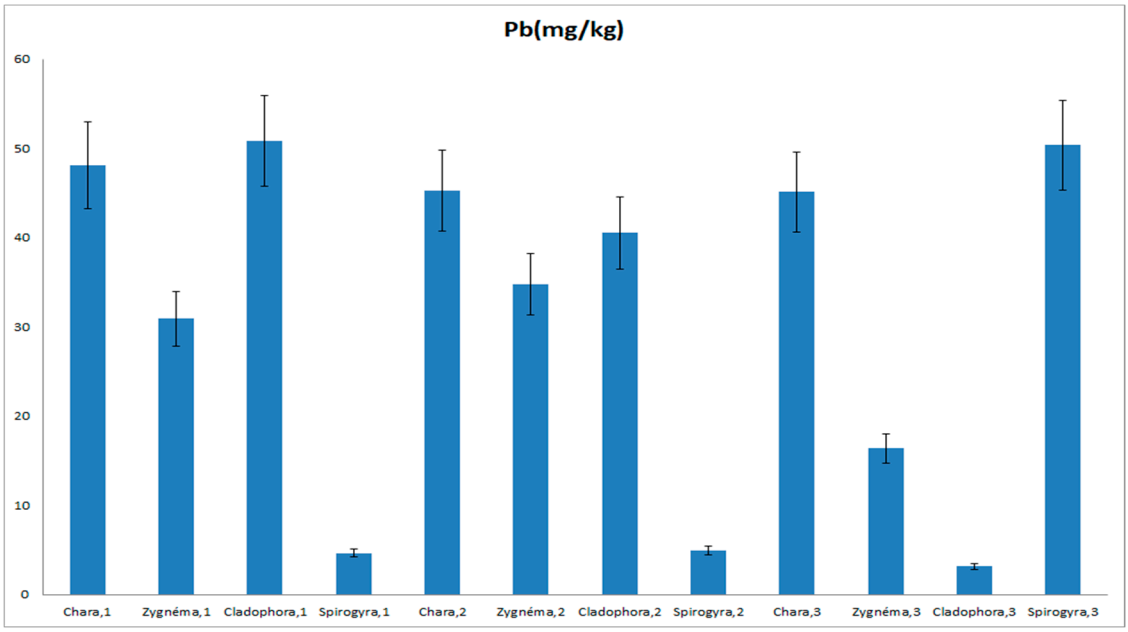



**Figure 6.** Pb contents of the four algae studied (for the three sites considered).

### *3.1. PCA Analysis*

Due to the fact that the first two eigenvalues account for 75.72 percent of the inertia, we will restrict ourselves to these two axes and summarize the data by the first two main components (Table 2).

**Table 2.** Eigenvalue matrix.

| | F1 | F2 | F3 | F4 |
|---|---|---|---|---|
| **Own Value** | 1.97 | 1.06 | 0.68 | 0.29 |
| **Variability (%)** | 49.26 | 26.46 | 17.02 | 7.26 |
| **% Cumulative** | 49.26 | 75.72 | 92.74 | 100.00 |

The degressive nature of the successive eigenvalues indicates a significant degree of data variability. Using the cosine square of the angle formed by the points with the projection plane to evaluate the quality of the points' depictions prevents misinterpretation. In addition, the relative contributions of the points to the development of the axes make it feasible to identify the points that contribute the most to the axes' meanings (Table 3).

This structure makes it easy to detect approximately X groups of individuals on the factorial plane generated by the first two principal components (Figure 7):

- Group 1: *Spirogyra* 1, *Spirogyra* 2, and *Cladophora* 3 with Fe
- Group 2: *Zygnema* 2, *Zygnema* 3 with Pb.
- Group 3: *Zygnema* 1, *Chara* 2, *Chara* 3, and *Cladophora* 2 with Zn.
- Group 4: *Cladophora* 1, *Spirogyra* 3, and *Chara* 1 with Cu.

The samples (those in the black circle with *Cladophora* 2, typically) positioned around the origin of the axis demonstrate random values for the factors under consideration (Figure 8).

**Table 3.** Explanatory points for axis one.

| Samples | Coordinates | Square Cosines | Contributions |
|---|---|---|---|
| ***Chara1*** | 1.695 | 0.291 | 12.147 |
| ***Chara2*** | 0.228 | 0.036 | 0.219 |
| ***Chara3*** | 0.378 | 0.149 | 0.603 |
| ***Zygnema1*** | 0.002 | 0.000 | 0.000 |
| ***Zygnema2*** | −0.877 | 0.156 | 3.250 |
| ***Zygnema3*** | −1.870 | 0.797 | 14.782 |
| ***Cladophora1*** | 2.623 | 0.704 | 29.105 |
| ***Cladophora2*** | −0.203 | 0.017 | 0.174 |
| ***Cladophora3*** | −1.423 | 0.673 | 8.566 |
| ***Spirogyra1*** | −1.018 | 0.320 | 4.382 |
| ***Spirogyra2*** | −1.532 | 0.705 | 9.921 |
| ***Spirogyra3*** | 1.996 | 0.905 | 16.852 |

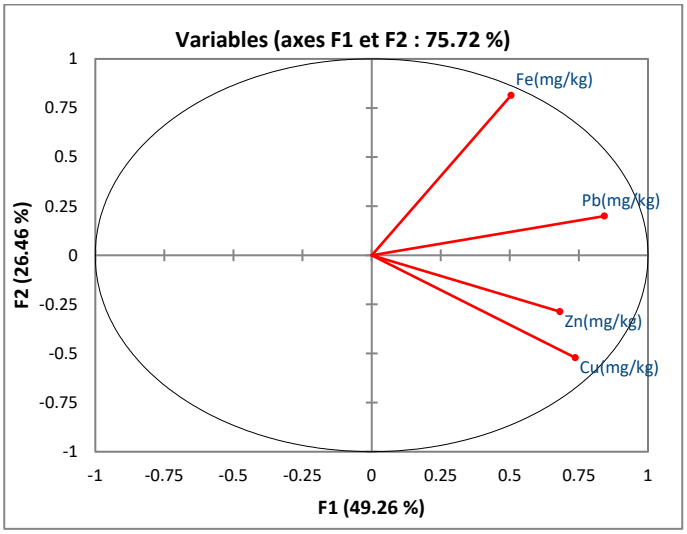



**Figure 7.** F1xF2 factorial design of variables.

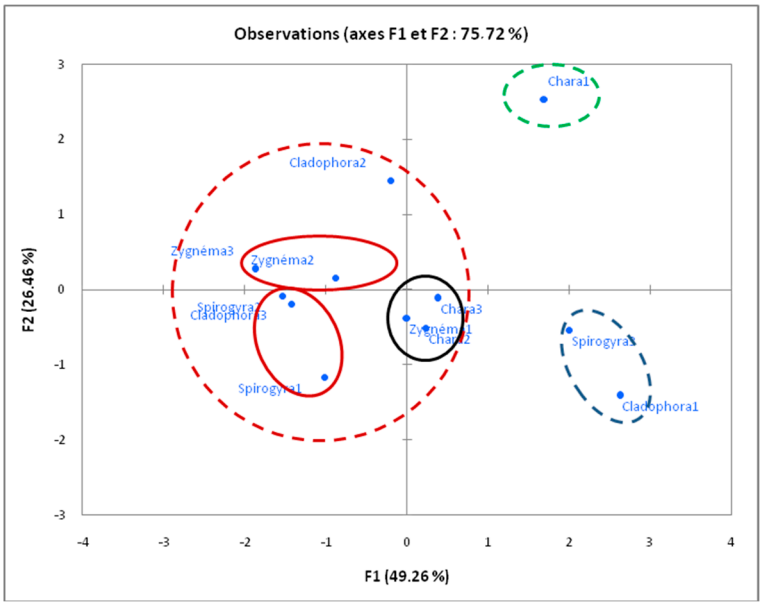



**Figure 8.** F1xF2 factorial design of individuals (samples).

The findings of the PCA are overlaid with those of the AHC (see the dashed groups on the factorial plan of the PCA and the dendrogram of the AHC shown in Figure 9).

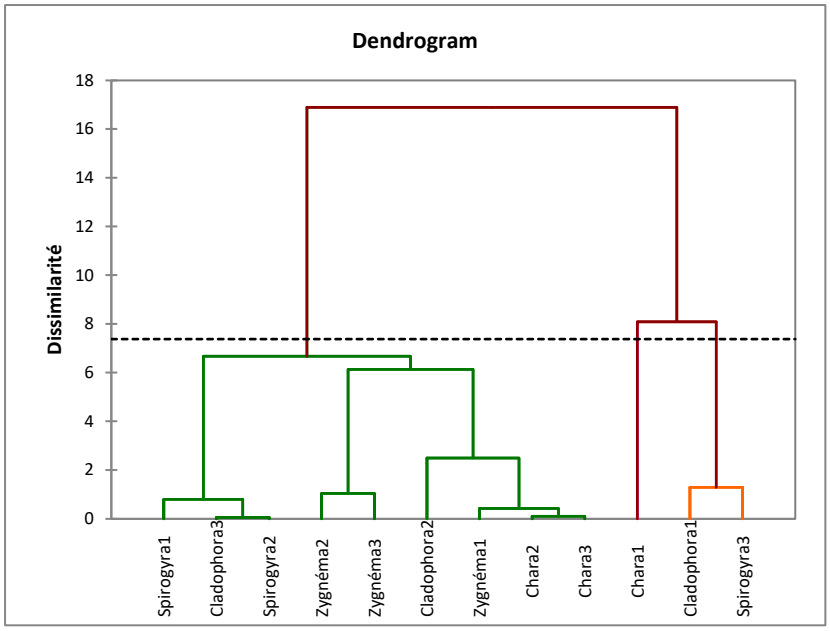



**Figure 9.** Structure dendrogram of individuals (samples) according to the CAH.

*3.2. Principal Component Analysis (PCA)*

In Tables 2 and 3, the findings of the Principal Component Analysis (PCA) are shown. This analysis enabled the identification of four primary components. The total of the variances is % (Table 3), which is still more than 70 %. These factors account for the variance expressed and explain the information sought. The representation using these factors gives a satisfactory account of the structure of the scatterplots. The affinity that exists between all the variables taken is given by the correlation matrix (Table 2). Table 3 shows significant correlations between algae and trace metals. The results of the PCA are used to select the different factors needed to interpret the different data. The analysis of the graphs of the space of variables, in the factorial plane F1–F2 (Figure 9), shows that this plane alone represents 75.72% of the information sought, that is, of the variance expressed. The community circle (Figure 8) shows that the most important factor F1 is determined by heavy metals. This grouping is dominated by heavy metals that come from urban and industrial pollution. Factor 1 highlights metal pollution of anthropogenic origin related to industrial, agricultural, and domestic activities [33–35].

The F2 factor is determined by the different species of algae, which shows the important role of these bioindicators to accumulate these heavy metals. The presence of trace metals (Pb, Fe, Cu, and Zn) with levels exceeding the recommended standards reveals metal pollution of anthropogenic origin.

## 4. Conclusions

The current study showed that the levels of heavy metals in algae studied in the three chosen sites were higher than the acceptable limit. The accumulations of heavy metals were very high, which reflects the pollution of those sites due to the increased discharge of waste materials and pollutant.

- The high bioaccumulation abilities of *Cladophora, Spirogyra, Chara, and Zygnema* algae for selected metals have been confirmed.
- The results indicate that some species may serve as better biomonitors than others for certain elements. Based on the agreement with the available literature, we suggest *Cladophora* for Fe and Zn, Chara for Pb, and *Spirogyra* for Cu.

- Interestingly, algae could serve as a good bio-indicator for pollution. The dispersal of heavy metals and their increased levels indicate the need for the government to monitor water of those sites more effectively and create public awareness of heavy metal accumulation in food.

The purpose of this study is to assess the level of heavy metal pollution in certain algae from the city of Djelfa. The research conducted on the wastewater of the three locations within the city of Djelfa have enabled the identification of the key findings:

- In the three sites of the city of Djelfa, the levels of Fe and Pb in the four species of algae exceed the IAEA standard (497 mg/kg and 0.574 mg/kg, respectively). The levels of Cu also exceed the standard (23, 2 mg/kg) in the four species for site 1, in *Chara* and *Zygnema* for site 2, and in *Chara* and *Spirogyra* for site 3, whereas the levels of Zn remain low and are significantly lower than the standard.
- Our findings on the amounts of Cu and Zn show the significance of these two metals to the biological processes. We have prioritized the accumulation rates of the four examined metals throughout this investigation. For the three sites, the metal accumulation gradient is as follows: Fe > Cu > Pb > Zn.

Therefore, algae can be a very good aquatic bio-filter plant for the bio-assessment of polluted urban stream ecosystems and polluted waters.

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