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Bioaccumulation of lead by *"xanthoria parietina"* and *"hylocomium splendens"*, and its effect on some physiological parameters

N. Kouadria[1*], F. Belhoucine[1], N. Bouredja[1], M. Ait Kaci[1], Y. Abismail[1], A. Alioua Berrebbah[1]

[1] *Département le vivant et l'environnement, Faculté SNV, Université des sciences et de la technologie Mohamed Boudiaf, El M'naouar, BP 1505, Bir El Djir 31000, Oran, Algérie*

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nawel.kouadria@univ-usto.dz;
Phone: +213699135342;

Abstract

Air pollution is one of the most serious problems in the world. Other than natural sources, air pollution can also result from human activity; the level of atmospheric pollution has reached the dimensions that threaten human health due to the rapid urbanization and the increase of number of vehicles in the market, which are considered as the main emission sources of important quantities of toxic substances in the atmosphere that can remain intact in nature for long period affecting human, animals but also plants. The livings beings are the reflection of the environment in which they evolve and their observation in various levels of biological organizations can offer indications about the quality and the characteristics of that environment, therefore, the study of the bioaccumulation of xenobiotics in exposed organisms represents an important way of assessing pollution's level. Due to the absence of root system, and due to their bio-accumulating power, lichens and bryophyte are, since decades, used as bio-indicator of air pollution. In this paper we tested an in vitro contamination of two cryptogrammic species, known for their bioaccumulation power, by different concentrations of lead; a lichenic species "*Xanthoria parietina*" and muscicole species "*hylocomium splendens",* collected from the region of Bir El Djir "Oran ", in order to characterize their bio accumulating power of lead, as well as the impact of the different concentrations of that xenobiotic on their physiological parameters. This study allowed as to identify the perturbation of the different physiological parameters in lichen "*Xanthoria parietina*" and bryophyte "*hylocomium splendens"*. These changes were correlated with the important contents of lead accumulated by both thalli, from solutions in which the two species were exposed.

1. Introduction

It is known that the existence, life and health of people depends on the conditions and the quality of the atmosphere [1]. The atmospheric pollution and the improvement of air quality still represent huge and pressing issues worldwide [3]. Nowadays, the level of atmospheric pollution has reached the dimensions that threaten human health, with the rapid urbanization and the increase of energy conception [2], but also due to the fast increase of number of vehicles, which are responsible of the release of considerable amounts of xenobiotic, notably many heavy metals [4] [5]

The presence of heavy metals in the atmosphere involves the whole environment and human life and it leads to a multitude of adverse consequences to humans, ecosystems and climate [3]. These heavy metals have a separate precaution in pollutants especially in terms of human health, because they can remain intact in nature for long period [2].

Lead (Pb), one of the most hazardous heavy metals, derived from both natural and anthropogenic sources, can persist in the environment, its occurrences as long-range trans-boundary air pollutants and wet and dry deposits of atmospheric emissions represent a serious risk factor for human health by inhalation or entrance and bioaccumulation in food chains, with a consequent increase of acute and chronic diseases [3]. Currently, air pollution is involved in a large number of respiratory and cardiovascular diseases. More recently, the role of particulate matter from pollution has been studied in autoimmune diseases; urban areas have been shown to be associated with a higher incidence of respiratory diseases. [6]

The livings beings are the reflection of the environment in which they evolve and their observation at various levels of biological organizations can offer indications about the quality and the characteristics of that environment [7]. The use of lichens as indicators of air quality dates back to 1866 when Nylander evaluated air quality based on changes observed in the lichen community composition at Jardin du Luxembourg in Paris. Since then, the process of assessing changes in lichen community composition in order to evaluate "air quality" became one of the most used tools in most European countries. [8]

However, this technique of using biological material to monitor the environment, only became well established across the world following the ground-breaking work by Rühling and Tyler in the late 1960s, introducing the use of naturally growing bryophyte as monitors of heavy metal deposition from the atmosphere [8]. Therefore, the measurement of heavy metals concentrations in plants is important both for the monitoring of air quality and for the determination of their ability to remove these pollutants from air to increase its quality [9].

It is in this context, and as a part of our work that we found it useful to study the ability of "*xanthoria parietina*" and "*hylocomium splendens*" to bio accumulate lead, in which they were exposed in the laboratory, under controlled conditions, and to identify the impact of this xenobiotic on some physiological parameters in these species.

2. Material and Methods

2.1. Presentation of the sampling region

Bir el djir is one of the biggest cities in Oran; it occupies an area of 101.7 Km^2. It is located in the north of Oran, 8km from the city center. According to the direction of environment of Oran, Bir el djir is a city that remained essentially agricultural until the end of the years 80; it now hosts a population of 118,000 inhabitants and becomes a major pole of the Oranean agglomeration. It houses several corporate offices with modernist architecture ; the new University Hospital1st November and University education Institutes, where USTO is the biggest. According to the National Meteorological Organization 2014 The Oran region has a classical mediterranean climate marked by a summer drought, mild winters, and a bright and clear sky. During the summer months, precipitation becomes rare or even non-existent.

2.2. Sampling

The sampling site is chosen due to the presence of an important biomass of lichen and mosses, among which, "*xanthoria parietina*" and "*hylocomium splendens*" were the most seen.

Figure 01. *Xanthoria parietina*

Figure 02. *Hylocomium splendens*

The sampling of the two species, shown in figure 01 and 02 above, were done at the sciences and technology university of Oran. For the lichen species, we detached thalli from their phorophyte using a knife, as for the muscicole specie, we carefully detached thalli from the soil. The samples taken were placed in a well-labelled bag used for short-term preservation [10], on which all the information of samples are mentioned, and transported to the laboratory.

2.3. In vitro contamination of "xanthoria parietina" and "hylocomium splendens"

Firstly, we have prepared 3 solutions with different concentrations of lead, 50, 100 and 350 µg/l, while the concentration of 50µg/L is the admissible dose in water, following the technique of (Durfor, 1964) [11]. The contamination of thalli was ensured by soaking 5g of thalli from each species in solutions of different concentrations of lead.

2.4. Analytical study

In order to study the impact of the different concentrations of lead on the studied species we opted for the dosage of the following parameters.

2.4.1. Lead measurement

After the complete mineralization, the measurement of lead content were done following SAA (atomic absorption spectroscopy) technique. The results are directly read on the spectrophotometer (PERKIN-AIMER model 400.

In the other hand, the lead content was measured in the different solutions where thalli of lichens and mossed have stayed.

2.4.2. pH measurement

Using a pH meter, we followed the temporal evolution (each week) of the pH of all samples in solutions treated at different concentrations.

2.4.3. Chlorophyll a, b and a+b measurement

We measured the content chlorophyll a and b following the method proposed by (Rao and le Blanc, 1965) [12].

2.4.4. Chlorophyll a, b and a+b measurement

The calculation of the proline content is determined following the formula proposed by (Mon neveux and Nemmar, 1986) [13].

3. Results and discussion

3.1. Results of lead measurement

The results of the lead measurement in the three solutions in which lichen's "*xanthoria parietina*" thalli were soak after 15, 30 and 45 days given in figure 03, allowed us to notice a remarkable over time decrease of concentrations of lead in the three solutions.

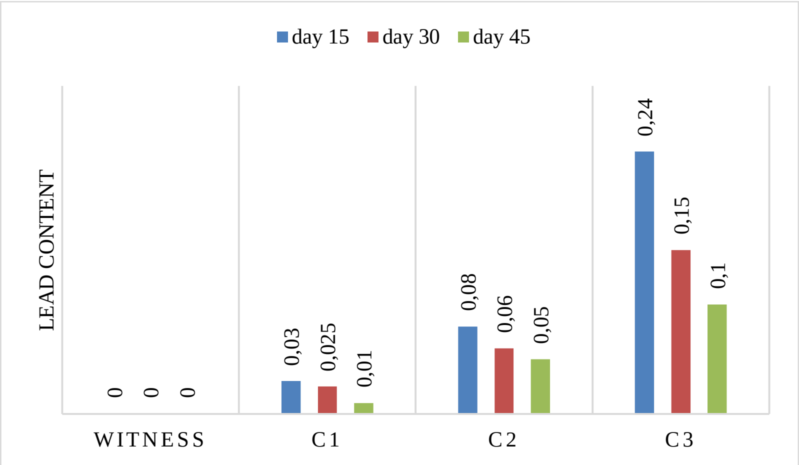

Figure 03: variations of lead content in solutions where "*xanthoria parietina*" stayed.

Lead concentrations shown in figure 04 reveals decrease of concentrations of lead in the three solutions in which bryophyte's "*Hylocomium splendens*" thalli were soak and that after 15, 30 and 45 days in, reflecting the bio-accumulation of lead by the bryophyte.
The measurement of lead content after 15, 30 and 45 days in thalli of both species allowed us to notice a remarkable over time increase of lead content in thalli which have stayed in the three solutions. Going up to 10 µg/l in lichen's thalli and 3.6 µg/l in bryophyte thalli as it is shown in the figure 05.

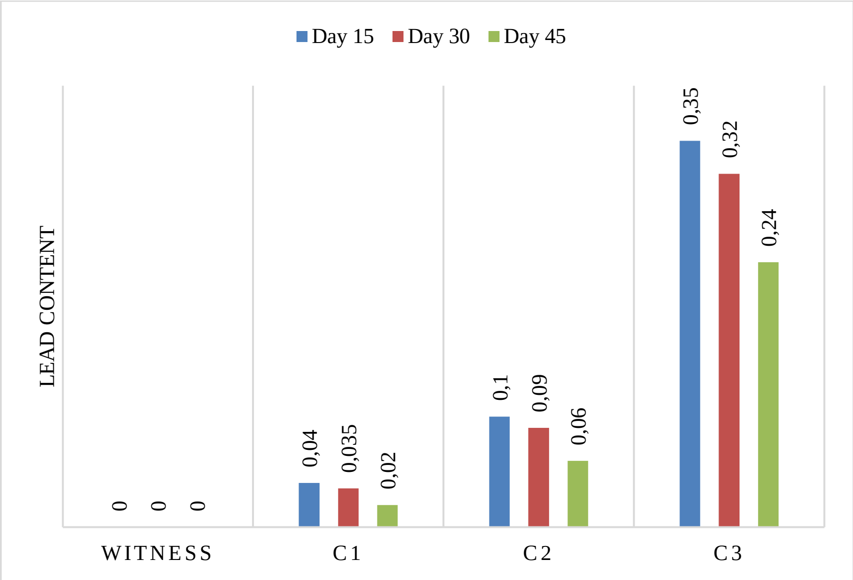

Figure 04: variations of lead content in solutions where "*Hylocomium splendens*" stayed.

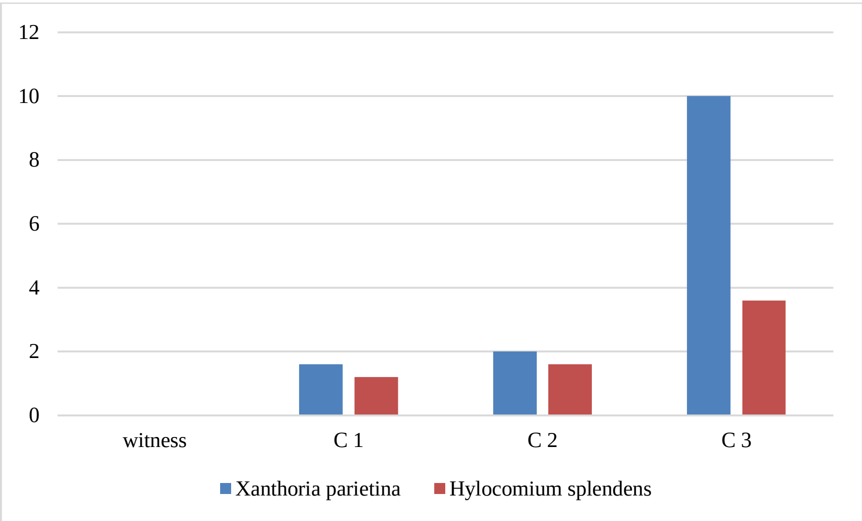

Figure 05: content of lead (µg/g) accumulated by thalli of "*xanthoria parietina*" and "*Hylocomium splendens*".

3.2. Results of pH measurement

For both species whatever the lead concentration, the pH of the solutions has increased, as it is given in figure 06 and 07 due to the absorption of some elements resulting from the dissolution of lead nitrates, which implies the dissociation of its constituent ions.

3.3. Results of chlorophyll a, b and a+b measurement

In the chlorophylien plant as we can see in the witness in both figure 08 and 09, the content of Chl a is superior than Chl b, but chlorophyll a, b, and a+b is conversely proportional to accumulated lead doses both in lichens and mosses, and decreases as a result of the reduction in photosynthetic intensity.

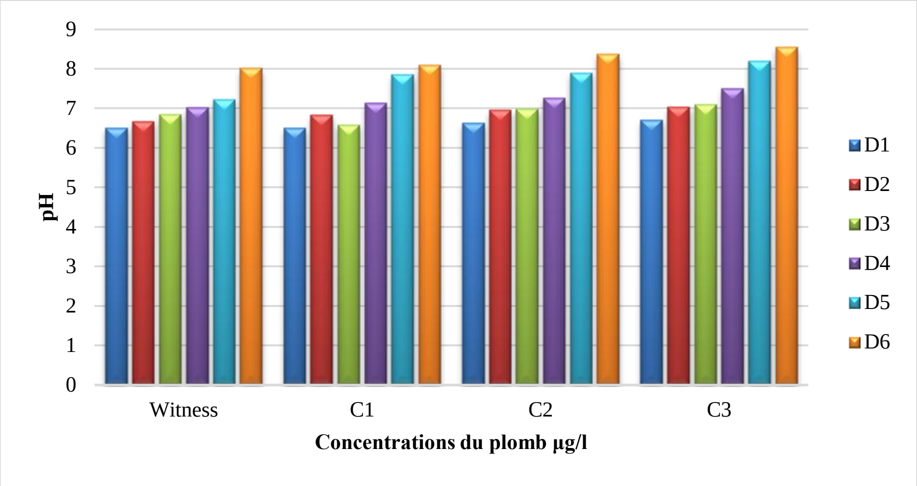

Figure 06: variation of pH of solutions where «*Xanthoria Parietina* » stayed.

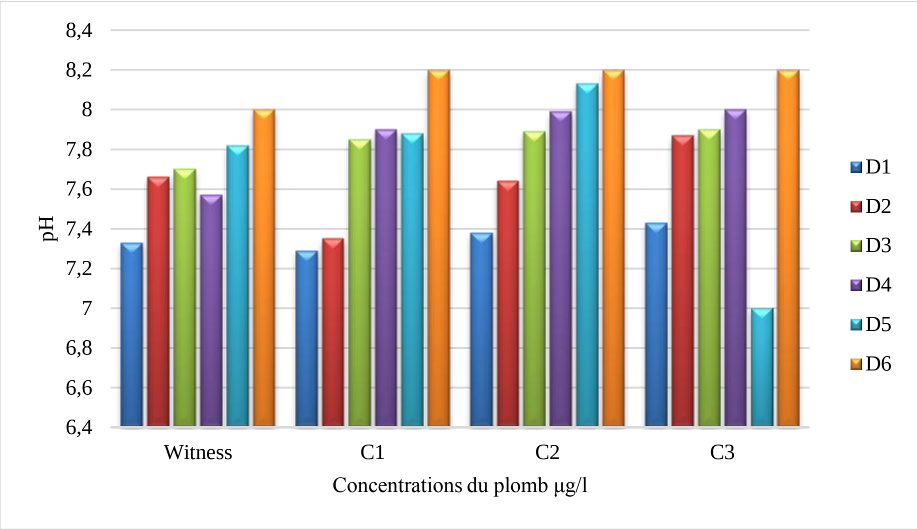

Figure 07: variation of pH of solutions where « *Hylocomium splendens* » stayed.

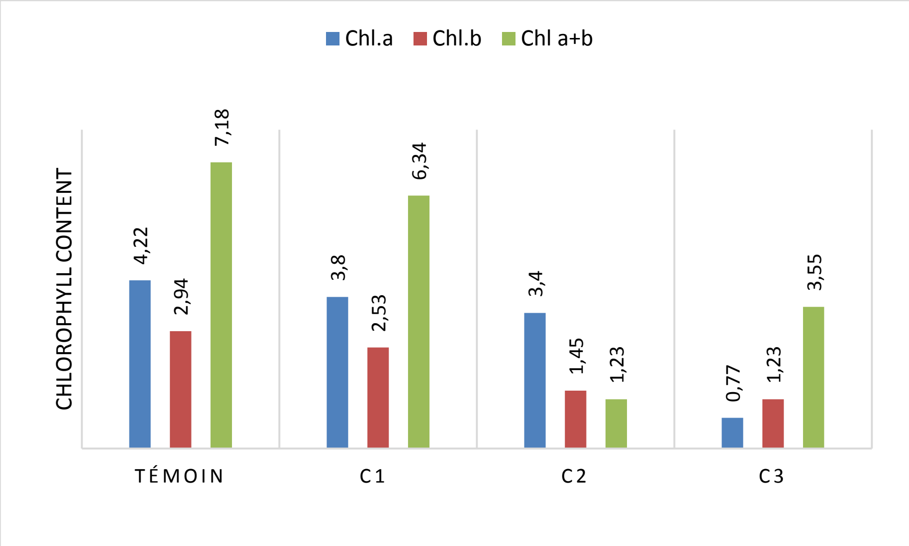

Figure 08: variations of chlorophyll a, b and a+b content "*xanthoria parietina*" thalli.

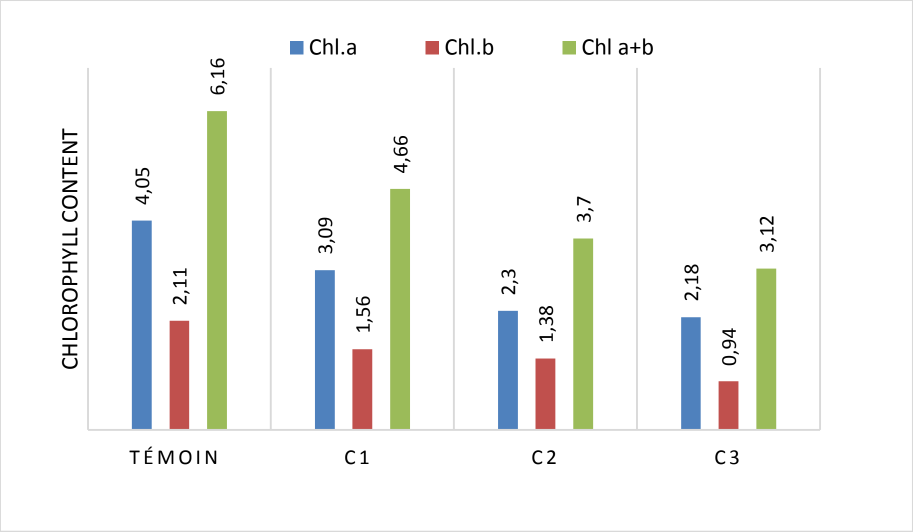

Figure 09: variations of chlorophyll a, b and a+b content "*Hylocomium splendens*" thalli.

3.4. Results of proline measurement

The proline content of the sample (S) collected from its natural habitats, shown in figure 10 was very high in both species, tended to stabilize for the concentration C1, and increased respectively With C2 and C3 indicating acute stress during the sudden disturbance of plants. By analysing the results, the set of values clearly indicates a remarkable decrease of lead content in the three solutions, while lead content clearly increased in the two species' thalli, this can be justified by the capacity of lichens and mossed to accumulate lead via cations exchange mechanisms [14] [15].

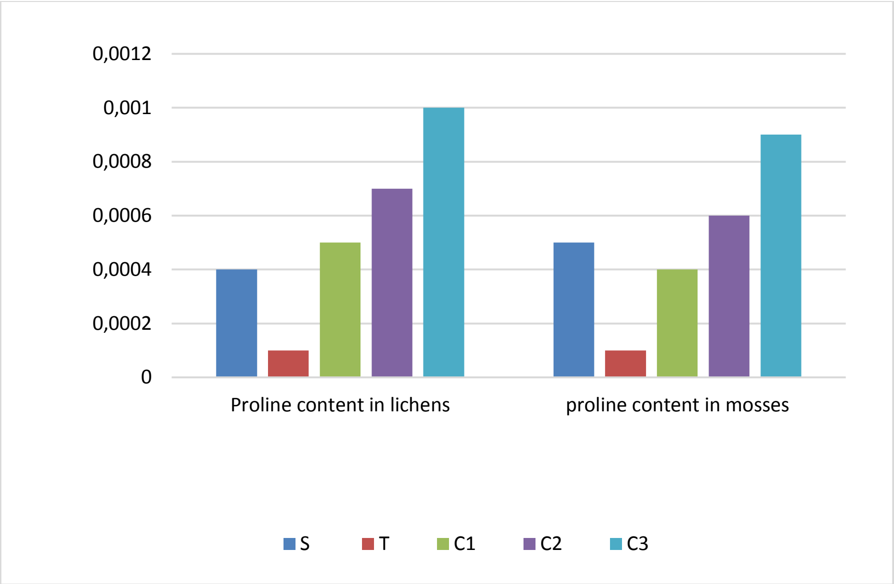

Figure 10: Variation in Proline content in *Xanthoria parietina* and *Hylocomium splendens.*

The physiology of these plants and their high exchange capacity allows a good accumulation of these particles [16], which, according to their physio-chemical characteristics, can be adsorbed, absorbed or eliminated by thalli [17]. Accumulation of lead by thalli is, according to previous studies, correlated with the appearance of oxidative stress induced by reactive species of oxygen ROS at the cellular level [14] [17], at the same time, it is responsible on physiological changes such as chlorophyll content, respiratory activity and photosynthetic activity, as well as proline content, which totally agree with our results. Our study have just reinforced results obtained by [18] [19] [20] by contaminating lichens and mosses by heavy metals in controlled conditions.

Due to their accumulator power, both species have proven to be very good bio accumulator of lead, which makes them excellent bio indicators for the estimation of air pollution [22] [21] [23] [24] [25] [14] [26] especially by metals (case of lead).

Conclusion

Atmospheric deposition of heavy metals can have long-term effects on species compositions of ecosystem and human health [27]. In the present study, the contamination of a lichenic species "*xanthoria parietina*" and muscicol species "*hylocomium splendens*" by different concentrations of lead under controlled conditions, allowed us to notice a significant accumulation of that xenobiotic by thalli of the both species, this lead was responsible of many physiological disturbances in the plants.

Due to their accumulating power, it is obvious to mention that the two studied species are proven good bio indicators of air quality.

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