## Monitoring of Sunflowers Grown in Soils Subjected to Different Lead Doses

## Monitoramento de Girassóis Cultivados em Solos Submetidos a Diferentes Doses de Chumbo

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## Abstract

Heavy metals are elements present in the soil that, among other harmful effects, affect human health and the environment. Phytoremediation has been seen as a promising method for recovering areas contaminated by heavy metals and may thus represent the recovery of such areas through depollution. The sunflower is an oilseed that has a high commercial value, with a high-quality edible oil, allowing for the utilization of by-products from the extraction of its oil and production of biofuel and turning out to be an excellent bioaccumulator of these metals. The objective this paper assessed the remedial potential of this plant in contaminated soils at different concentrations of lead. The experiment was set up using nine hanging flower beds subjected to three test treatments T1: 0 mg of Pb kg<sup>-1</sup> of soil (references beds); T2: 14.4 mg of Pb kg<sup>-1</sup> of soil and T3: 7.2 mg of Pb kg<sup>-1</sup> of soil (with three doses of the metal in the soil), with three repetitions. Morphometric measurements such as plant height, leaf length and width, stem and capitulum diameter were assessed. At the end of it vegetal cycle, the material was harvested and dehydrated to carry out an atomic absorption spectrometry analysis. The treatments showed differences indicating a major influence of lead element on the crop. The roots of the plants show a high capacity for lead uptake, characterizing them as phytoremediation plants for this metal and, consequently, showing a strong mitigating capacity, while the stems presented multiple capitula, which characterized the plant as a bioindicator as well.

Keywords: Environment. Heavy Metals. Phytoremediation.

## Resumo

Os metais pesados são elementos presentes no solo que, entre outros efeitos nocivos, afetam a saúde humana e o meio ambiente. A fitorremediação tem sido vista como um método promissor para recuperação de áreas contaminadas por metais pesados, podendo assim representar a recuperação dessas áreas através da despoluição. O girassol é uma oleaginosa de alto valor comercial, com óleo comestível de alta qualidade, permitindo o aproveitamento de subprodutos da extração do seu óleo, produção de biocombustível e revelando-se um excelente bioacumulador desses metais. O objetivo deste trabalho foi avaliar o potencial corretivo desta planta em solos contaminados com diferentes concentrações de chumbo. O experimento foi montado utilizando nove canteiros suspensos submetidos a três tratamentos T1: 0 mg de Pb kg¹ de solo (testemunha); T2: 14.4 mg de Pb kg¹ de solo e T3: 7.2 mg de Pb kg¹ de solo (três doses do metal no solo), com três repetições. Foram avaliadas medidas morfométricas como altura da planta, comprimento e largura das folhas, diâmetro do caule e capítulo. Ao final do ciclo vegetal, o material foi colhido e desidratado para realização de análise por espectrometria de absorção atômica. Os tratamentos apresentaram diferenças indicando grande influência do elemento chumbo na cultura. As raízes das plantas apresentam alta capacidade de absorção de chumbo, caracterizando-as como plantas fitorremediadoras desse metal e, consequentemente, apresentando forte capacidade mitigadora, enquanto os caules apresentam múltiplos capítulos, o que caracteriza a planta também como bioindicadora.

Palavras-chave: Meio Ambiente. Metal Pesado. Fitorremediação.

# Introduction

Internationally, the issue of soil pollution has been a concern among experts, authorities and society. The incorrect disposal of both industrial and domestic waste bears a great impact on the atmosphere, drinking water and soil, especially regarding heavy metals.

Lead (Pb) is a ubiquitous heavy metal in the earth's crust whose physical properties have made it useful to humanity for centuries. It is still produced by means of various industrial processes, although to a lesser extent than in the previous decades (Rosen *et al.*, 2017). During most of the 20th century, however, Pb was widely used in gasoline, paints, pesticides,

batteries and pipe fittings (ATSDR, 2007).

The use of phytoremediation plants to clean areas contaminated by these metals has been targeted by research, especially due to their low cost, easy development and application, to reduce the damage caused by heavy metals in soils (Nascimento, 2013).

The phytoremediation process occurs directly or indirectly in the plant through stimuli on the rhizosphere microbiome, allowing for the decontamination of the environment through extraction or degradation performed by a number of techniques classified as: phytoextraction, phytodegradation, phytovolatilization and phytostimulation, in addition to phytostabilization (Accioly; Siqueira, 2000), and this strategy is based on advances in the knowledge of soil chemistry, ecology, plant physiology, soil microbiology, ecotoxicology and agriculture (Koptsik, 2014).

Lately, a lot of interest has been directed to the use of the sunflower (*Helianthus annuus*) for the phytoremediation of organic pollutants and heavy metals due to the capacity of the plant to absorb metals from the environment. The site where these heavy metals accumulate differ from one plant to another (Ojuederie; Babalola, 2017).

Angelova *et al.* (2012) showed that the distribution of heavy metals across sunflower organs is selective for each metal, ranging from 59% of Pb accumulated in the leaves to as low as 1% accumulated in the seeds. Similar findings were seen for Zn and Cd, with accumulation rates of 47% and 79% in sunflower leaves, respectively (Ojuederie; Babalola, 2017).

Koptsik (2014) points out that the hyperaccumulation of metals seems to be an evolutionary adaptation of plants to life in unfavorable habitats with high contents of heavy metals, in soils of ultrabasic rocks, and that the advantages that plants show due to metal hyperaccumulation remain a mystery. Bhargava *et al.* (2012) considers that these advantages are probably related to tolerance to these metals, resistance to drought, ability to compete with less tolerant plants, tolerance to unintentional uptake of heavy metals and protection against pathogens and herbivorous animals.

Plants considered hyperaccumulators can develop in highly contaminated environments. Thus, they can accumulate high levels of potentially toxic elements in their tissues, for example, contents above 1,000 mg kg<sup>-1</sup> of Pb (Accioly; Siqueira, 2000; Pereira *et al.*, 2010; Zeitouni *et al.*, 2007).

According to Anjum *et al.* (2012) lead levels in the environment range from 4 to 20 mg g<sup>-1</sup> of dust. Non-contaminated waters contain this element at concentrations ranging from 0.001 to 0.06 mg L<sup>-1</sup>. In soil, they reach 5 to 30 mg per kg of soil. To be deemed ideal, a phytoextractor plant must present, as its key characteristics, tolerance to the high concentration of the pollutant present in soil, grow quickly, have low water requirement, obtain efficiency in the removal of the pollutant, be non-invasive, pose some resistance to pests and diseases, have low input requirements and be easily harvested (Herzig *et al.*, 2014).

Sunflower plants can be used for the phytoremediation of soils contaminated with heavy metals, being characterized as suitable plants for the removal of high amounts of lead (Pb), copper (Cu) (Yeh *et al.*, 2015) and zinc (Zn), in addition to tolerating high metal concentrations in soil (Herzig *et al.*, 2014; Zalewska; Nogalska, 2014). Once grown in soils contaminated with heavy metals, they possess the ability to accumulate expressive levels of the element. It is important to consider, however, the fact that the sunflower is a very versatile crop and can be used to feed both humans and animals, in addition to its ornamental use. Its grains are used for the production of oil and margarine and nearly 400 kg of oil per ton of grains is extracted from it; in addition, 350 kg of cake is obtained for animal feeding

purposes (Amabile et al., 2002).

As a protein source, the sunflower is also classified as the fourth option for animal feed and human use (Briega *et al.*, 2018). It accounts for approximately 13% of all plant oil produced in the world and stands out for a quick increase in terms of human consumption, which results not only from the great quality of its oil, but also from its excellent physico-chemical and nutritional characteristics (Amabile *et al.*, 2002).

Currently, Brazil holds the 27th position on the ranking of top sunflower producers, with a planted area of 30,900 hectares and production of 50,400 tons of grain, the major producers being the states of Mato Grosso, Goiás and Rio Grande do Sul (CONAB, 2021). In other words, the fact that it is an important oilseed to produce edible oils worldwide, among other derivatives for human consumption, in addition to its ability to bioaccumulate heavy metals, further reinforces the need and the importance to know the extent of this ability of the plant, and, even more so, to grow it in soils known to be free of these elements.

The presence of heavy metals in soil may lead to morphological, physiological, biochemical and structural changes in plants. An example that can be mentioned is the fact that, in the presence of an excess lead (Pb) concentration, there are interferences with cell division and inhibition of the root system extension (Sharma; Dubey, 2005), in addition to a considerable decrease in plant growth due to a reduction in functional activity and changes at different structural levels occurring in the photosynthetic apparatus in view of high metal levels (Kosobrukhov *et al.*, 2004).

Therefore, the purpose of this paper was to assess morphometric and morphological changes, in addition to Pb uptake, accumulation, translocation and tolerance in sunflowers grown at three different concentrations of the metal and confirm that the sunflower behaves as a phytoextractor in the presence of different lead doses.

### 2 Material and Methods

The study was conducted on Homeopassos Farm, in the city of Passos, Minas Gerais, Brazil (Latitude 20° 43' 08" S, Longitude 46° 36' 35" W, altitude 745 m). According to the Köppen climate classification, this location is under the influence of the Pampas temperate, mesothermal Cwa climate.

The annual mean temperature is  $20.9 \,^{\circ}\text{C}$  with a mean rainfall of  $1423 \, \text{mm}$ .

The full life cycle of a sunflower from planting to senescence is 95 days, from October 16, 2017 (planting) to January 20, 2018 (harvest). The average temperature of the cultivation days was 29 °C, the temperature of the vase was 32 °C, and the lux meter (light intensity meter) showed an illuminance of 42,000 lux.

Nine beds were built containing three plants each and subdivided as follows: three beds containing soils subjected to a concentration of three reference beds (Treatment 1 - beds A), three beds at a concentration of 7.2 mg of Pbkg<sup>-1</sup> of soil (Treatment 2- beds B); and 14.4 mg of Pb kg<sup>-1</sup> of soil (Treatment

3 - beds C), all distributed across an open-air area. To prepare the element, dilution was performed in 5 mL of nitric acid (HNO<sub>3</sub>) to obtain the decomposition of the metal (Pb). The contamination of soil with this metal was based on the resolutions of CONAMA (2013).

Seeds of the IAC (Instituto Agronomico de Campinas) - Uruguay cultivar was planted and grown at a distance of 20 cm between plants.

Nine beds were set up and each experimental unit consisted of a plastic vase with a capacity of 21 L.

To prepare the soil, the following mixture was used: 4 kg of gravel, used for drainage, and 19 kg of soil collected in a layer of 0 - 20 cm, simulating the characteristics of the region's soil, where the Cerrado soil prevails. The water from irrigation in the beds had to be drained, avoiding contamination of the local soil, and, for that purpose, hoses were placed at the bottom of the beds and connected to PET bottles, closing the circuit.

Beds with already contaminated soil were subjected to a seven-day rest time, allowing the solution containing the heavy metal to settle in the soil. Once the rest period was over, seeds were planted.

For planting, a soil analysis was carried out and, as recommended, lime and gypsum were applied in such a way to be incorporated into the soil. According to soil fertility calculations, based on the analysis table for the soil collected, it was determined that 11.13 g of lime and 4.41g of agricultural gypsum should be added into the nine experimental beds.

After the plantation was fertilized, topdressing fertilization was performed - 35 days after germination - according to the methodology studied, the fertilizer added was intended for supplying phosphorus in an amount of 2.69 g, in an 8-cm deep seeding furrow. Subsequently, four sunflower seeds were placed in wet soil at 5 cm. After the pit was closed, it was fertilized with urea on the surface, in an amount of 0.26 g to supply N. The plants were continuously monitored from germination to the final stage of their development - physiological maturation.

For the crops, during the monitoring of the beds for environmental conditions, a Manaus infrared thermometer, model TD-950, was used for temperature measurements to obtain the temperature of the soil surfaces in the beds, and a TFA digital thermometer, model NR 30.1026, was used to record the maximum and minimum temperatures of the environment.

As for the luminosity index, a Menipa lux meter, model MLM-1010, was used.

Morphometric measurements related to the development of the aerial parts (stem and leaves) were taken using a digital caliper, Western brand, model DC-60, and a tape measure.

Temperature, luminosity and morphometric data were used to collect data on the climatic factor for possible interferences with the development of the plants.

When the plants reached maturity, they were harvested and separated by parts (root, stem, leaves and capitula) and by levels of lead concentration and then taken to a drying oven, where they were dehydrated at a temperature of 70°C and subsequently

ground in a knife mill.

The ground material was intended for the performance of atomic absorption spectrometry (AAS) analyses in the Soil and Leaf Analysis Laboratory at Minas Gerais State University (UEMG). The analysis consists of a technique that uses this phenomenon to determine the number of elements (metals, semimetals and some non-metals) in samples such as biological, environmental, geological, technological and food materials by means of thermal energy for atomization processes (Krug *et al.*, 2006).

The experimental design used was completely randomized in a split-plot scheme over time. Thus, three doses, 12 days of measurements and three repetitions were used. It should be noted that the plots were the doses, and the days were the subplots. Subsequently, an analysis of variance (ANOVA) was performed, which was the procedure employed to compare the three treatments and the three repetitions each.

The data were subjected to the Scott-Knott test for comparison of means which is a technique that uses the likelihood-ratio test to group n treatments into k clusters. The statistical analyses were performed using the GENES software package (Cruz, 2013).

#### 3 Results and Discussion

Regarding all the plant structures analyzed (stem diameter, plant height, leaf width and length), it was possible to observe significant differences between them in the three treatments (1, 2 and 3) established for their respective beds (A, B and C).

The height of the plants in the contaminated soils (B and C), a delay in their growth could be observed in comparison with the control bed. Abreu *et al.* (2016), studying the behavior of the sunflower in lead-contaminated soils, also found significant differences in the growth of leaves, stems and roots, and a reduction in the dry mass of these structures was observed, when compared with plants grown in reference soils. For them, lead significantly reduced the calcium (Ca) and magnesium (Mg) contents in the leaves, stems and roots, which may explain, at least partially, the reduction in growth induced by Pb in the *H. annuus* plants. Boffe *et al.* (2017), in experiments on *H. annuus L.*, also noted that the accumulation of this metal reduced biomass production.

These results are also in agreement with the studies conducted by Andrade *et al.* (2009), who evaluated phytoextraction by growing sunflower (*H. annuus L.*), black oat (*Avena strigosa Schrebere*) and bahia grass (*Paspalum notatum Flugge*) and noted significant differences in plant development.

During the experiment, some symptoms of phytotoxicity were identified in the leaves of plants in soils containing lead, characterized by the presence of chlorotic leaves with already necrotic spots. Results like this support those obtained by Andrade *et al.* (2009), who, by examining the phytoextractor potential of the sunflower in a soil contaminated by the same metal, found that the plants showed symptoms of chlorosis and foliar necrosis.

In a study conducted by Abreu *et al.* (2013) evaluating the chlorophyll content in sunflowers grown in a lead-contaminated nutrient solution, it was possible to observe that the more lead was applied to the solution, the more intense the foliar chlorosis in the plants. This fact may be connected to the intervention of lead with the uptake of the micronutrient iron (Fe) and the macronutrient magnesium (Mg) by the plant, elements that are essential for the biosynthesis of chlorophyll molecules. This could account for the reduction in the length and width of plant leaves.

Alkhatib *et al.* (2011) studied the effects of lead toxicity on leaf gas exchange, chlorophyll content, chlorophyll fluorescence, chloroplast ultrastructure and stomatal opening in tobacco plants (*Nicotiana tabacum L.*) and found that the growth of roots and shoots, the net photosynthetic rate and stomatal conductance were significantly reduced in plants treated with 100, 300 and 500 M of Pb  $(NO_3)_2$ . They also observed that mesophyll cells showed altered chloroplasts with ruptured thylakoid membranes.

Several authors (Burton *et al.*, 1984; Ghoshroy *et al.*, 1998; Sharma; Dubey, 2005) have shown that exposure to Pb results in a strong reduction in plant growth and significant changes to many metabolic pathways. The visible symptoms of toxicity are characterized by leaves and a stunted growth of shoots and roots. The leaves show chlorosis and necrosis, while the roots turn black or dark brown, supporting this study, in which sunflower leaves also showed these spots.

Regarding the reproductive structures reflected in the floral capitula of sunflowers, it could be observed that there was an anomalous development in plants grown in soils with a higher lead concentration.

As for the reproductive structure analyzed (capitulum diameter), the capitula of plants grown in beds with higher lead concentrations stopped developing around early January, while the capitula in the other treatments continued to expand until the time of harvest, that is, at the end of the cycle, which occurred in late January 2018.

It should be noted that plants grown in the more contaminated beds showed, on average, four stems with a capitulum at the apex per plant; therefore, there was the formation of a larger number of capitula in this crop, albeit with reduced diameters compared with the others. One of the factors that may explain these stems on the same plant is the large amount of lead (Pb) to which this bed was subjected, since *H. annuus* is known to be a plant that accumulates heavy metals as mentioned by Garbisu and Alkorta (2001).

Experiments carried out by Garbisu and Alikorta (2001) demonstrated that lead (Pb) is an element that is retained at higher concentrations in the roots in detriment of the aerial part, showing that tolerant species accumulate elevated concentrations of contaminants in the root system, limiting metal translocation to the aerial portion in view of the physiological barriers imposed by the plant.

To assess the phytoextractor potential of plants, usually a comparison method is used for the concentrations of the pollutant accumulated in the roots and the aerial portion (Marques, 2009).

In this paper, through atomic absorption spectrometry (AAS) analysis (Table 1) it was possible to observe a significant uptake of this element by the plant, with greater accumulation in the roots (178.8 mgPb kg<sup>-1</sup>) of more contaminated plants, in contrast to 0.609 mgPb kg<sup>-1</sup> in the roots of the plants in the control beds, that is, it was confirmed that the species H. annuus L. presented different concentrations in relation to the buildup of the metal in the root system and the aerial part.

Table 1 - Data obtained from atomic absorption spectrometry analyses

Sample -	D 1	Sample	D 1	Sample -	D 14
A*	Result	- B*	Result	C*	Result
mg kg-1					
Leaf	0.24	Leaf	1.91	Leaf	2.01
Stem	0.39	Stem	4.12	Stem	5.34
Root	0.61	Root	8.6	Root	178.8
Capitulum	0.29	Capitulum	0.11	Capitulum	0.6
Soil	0.36	Soil	16.25	Soil	32.69

\* A = Control; B =  $7.2 \text{ mgPb kg}^{-1}$ ; C =  $14.4 \text{ mgPb kg}^{-1}$ 

Source: research data.

Similar results were found in the studies by Zeitouni (2003), which showed a low lead concentration in the aerial portion of the sunflower used in his experiment.

Regarding the results obtained from the statistical procedure using analysis of variance (ANOVA), it is observed in table 2 that the effects of doses and days were significant at 1% probability for all characteristics under study.

Table 2 - Analysis of variance of sunflower with 12 assessment dates (mean squares)

FV G	CI		Mean		
	GL	Height	Stem	Width F.	Length F.
Doses	2	74.224818**	0.915825**	13.286796**	11.210756**
Error a	6	3.34	0.38792	0.29078	0.811981
Days	11	414.604873**	4.344278**	58.324101**	86.939355**
Interaction	22	2.836823**	0.044106*	0.595775 ns	0.48388 ns
Error b	66	1.12	0.022063	0.391806	0.321565
Total	107				
CVp (%)		10.76	8.1129	8.8554	10.9733
CVs (%)		6.25	6.1184	10.1407	6.9056

Significant at 1% = \*\* Significant at 5% = \* non-significant = ns

Source: research data.

As illustrated in Table 2, for the interaction effect, the characteristic height was significant at 1%, stem was significant at 5%, and leaf width and length were not significant. About the data in Table 3, at week 11 of the experiment, the stem had greater thickness than a week 1, showing that there was a natural growth of the plant.

Table 3 - Test of means (Skott-Knott) at 12 weeks of data collection

	Stem	Height	Width F.	Length F.
Weeks	(mm)	(mm)	(mm)	(mm)
1	0.94 g	2.22 g	1.48 g	1,19 h
2	1.80 f	10.94 f	4.19 f	5.65 g
3	1.92 f	11.54 f	4.26 f	6.42 f
4	2.02 e	14.06 e	4.25 f	6.29 f
5	2.03 e	14.71 e	4.65 f	7.01 e
6	2.38 d	16.73 d	5.18 e	7.73 d
7	2.76 с	17.61 d	6.56 d	8.87 c
8	2.93 b	20.04 с	7.81 c	10.65 b
9	2.95 b	21.97 b	7.92 c	10.61 b
10	3.01 b	24.76 a	9.09 b	11.36 a
11	3.21 a	24.57 a	9.79 a	11.52 a
12	3.17 a	24.47 a	8.84 b	11.17 a

Means followed by the same letter in the column do not differ from each other by the Scott-Knott test (P<0.05%)

Source: research data.

However, it was possible to verify a small decrease (albeit not statistically significant) in the value of the stem measure at week 12, which can be explained by the fact that the plant was already at its 90th day of development, meaning that it already reached senescence, since its cycle was 110 days.

The results for the lead (Pb) doses available in the planting soil can be seen in Table 4.

Table 4 - Test of dose means (Skott- Knott)

Doses	Stem	Heigh	Width F.	Length F.
1	2.61 a	18.58 a	6.87 a	8.80 a
2	2.32 b	16.53 b	5.77 b	8.12 b
3	2.34 b	15.81 b	5.87 b	7.70 b

Means followed by the same letter in the column do not differ from each other by the Scott-Knott test (P<0.05%)

Source: research data.

In this Table, it is evident that the treatment used for the reference bed (without the addition of lead) represented by dose 1 differed statistically from the beds represented by doses 2 and 3, (7.2 mgPb kg<sup>-1</sup> of soil and 14.4 mgPb kg<sup>-1</sup> of soil, respectively), and no statistically significant differences were observed between them.

As regards the data evaluated (weeks x doses), the Scott-Knott test of means divided the data into two small groups, both with 11 treatments. After the division, 36 combinations were obtained (Table 5).

**Table 5 -** Test of means weeks x doses (Skott- Knott)

Weeks x Doses	Stem	Height	Width F.	Length F.
1	1.09 f	2.43 h	1.14 f	1.23 g
2	1.88 e	11.27 g	4.65 e	5.49 f
3	2.06 d	12.13 g	4.69 e	7.29 d
4	2.22 d	15.04 f	4.85 e	7.18 d
5	2.21 d	16.12 e	5.31 d	7.95 d
6	2.60 с	18.86 d	6.03 d	8.93 с
7	3.11 a	20.97 с	7.72 с	9.69 b
8	3.02 a	22.88 b	8.20 c	11.34 a
9	3.24 a	24.25 b	9.01 b	11.36 a
10	3.19 a	26.35 a	9.26 b	11.78 a
11	3.35 a	26.17 a	10.47 a	11.80 a
12	3.33 a	26.46 a	10.08 a	11.62 a
13	0.79 f	2.11 h	1.18 f	1.20 g
14	1.72 e	11.17 g	4.12 e	6.02 e
15	1.93 e	11.76 g	4.25 e	6.39 e
16	1.96 e	13.90 f	4.24 e	6.24 e
17	2.01 e	14.41 f	4.59 e	6.93 d
18	2.34 d	16.09 e	5.06 d	7.67 d
19	2.53 с	16.48 e	6.09 d	8.75 с
20	2.91 b	19.95 d	7.38 с	10.47 a
21	2.70 с	21.56 с	6.99 с	10.39 a
22	2.65 с	23.67 b	8.41 c	11.11 a
23	3.16 a	23.24 b	8.80 b	11.38 a
24	3.15 a	23.98 b	8.10 c	10.86 a
25	0.93 f	2.12 h	1.12 f	1.14 g
26	1.81 e	10.38 g	3.79 e	5.43 f
27	1.77 e	10.75 g	3.83 e	5.58 f
28	1.89 e	13.24 g	3.66 e	5.46 f
29	1.84 e	13.61 f	4.05 e	6.15 e
30	2.19 d	15.26 f	4.47 e	6.61 e
31	2.63 с	15.40 f	5.88 d	8.16 c
32	2.84 b	17.29 e	7.86 с	10.16 b
33	2.90 b	20.11 d	7.76 с	10.08 b
34	3.18 a	24.29 b	9.59 b	11.21 a
35	3.10 a	24.29 b	10.11 a	11.38 a
36	3.01 a	22.97 b	8.34 c	11.05 a

Means followed by the same letter in the column do not differ from each other by the Scott-Knott test (P<0.05%)

Source: research data.

When comparing the influence of lead dosage and plant development in terms of the combinatorial analysis established by the program, only combinations 8, 9, 10, 11 and 12 had statistically positive results for the parameters under study (stem diameter; stem height; leaf width and leaf length), with the results obtained for stem diameter and leaf length being slightly superior.

It was also possible to observe a positive relation between week and dosage among the structures in the combinations 23 and 24, except for leaf width. The same could be seen for the combinations 34, 35 and 36, whose parameter "leaf width" once again did not show a positive relation.

Review studies such as the one by Kumar *et al.* (2020) showed that numerous studies have presented lead as one of the most toxic heavy metals, which led those authors to emphasize

three characteristics of this metal: i) Pb bioavailability in soil, ii) Pb biomagnification, and iii) Pb remediation, addressed in detail through physical, chemical and biological lenses.

For the authors, however, specific integrated approaches to the site and the source should be adopted to formulate suitable remediation strategies. Biological remediation, such as phytoremediation and PGPR, can be an ecological and cost-effective strategy to alleviate Pb toxicity in moderately contaminated soils, as presented by Saleem et al. (2018) when demonstrating that lead contamination reduced plant growth, physiology and yield at all levels of lead stress, but that the application of LTPGPR to lead-contaminated soil improved plant growth, physiology, yield, antioxidant activities and proline content and lowered the malondialdehyde content (this is reduced by the application of different strains in lead contamination) in the sunflower compared with plants grown in soil without inoculation. Inoculation also promoted lead uptake in the root and shoots and reduced lead uptake in plant achenes, compared with plants subjected to lead contamination without inoculation.

Moreover, several studies have reported the presence of pesticide residues, heavy metals and mycotoxins in edible oils and fats (Kivevele; Huan, 2015; Bhat; Reddy, 2017). These products can be contaminated by environmental pollution and/or during the processing stages, before refining (Xia *et al.*, 2021), which may, in addition to harming the environment, lead to the loss of biodiversity and consequent environmental imbalance, as well as threaten human health, exposing consumers to a wide range of diseases.

## 4 Conclusion

Lead cause stress in the plant, which led to its weakening and increase vulnerability to pest attacks when exposed to higher doses.

The presence of heavy metals in soil, especially lead, can affect the entire sunflower production chain, impacting even human health through the consumption of seeds or edible oils.

The contaminant probably interfered with the plant's physiology, since the one subjected to the highest dosage showed a reduction in its growth and presented deformations in the leaves, in addition to presenting more than one stem per plant, when compared with the reference plant, which could elevate this sunflower to bioindicator status under these circumstances.

Lead caused stress in the plant, which ended up triggering its weakening, and its vulnerability to pest attack was enhanced when subjected to higher doses.

The presence of heavy metals in soil, especially lead, can affect the entire sunflower production chain, impacting even human health through the consumption of seeds or edible oils.

## References

ABREU, C.B. et al. Nutritional and biochemical changes induced in sunflower (Helianthus annuus L.). Semina Ciênc. Agr., v.37,

n.3, p.1229-1242, 2016.

ABREU, C. *et al.* Teores de pigmentos em girassol *Helianthus annuus* (Compositae) sob doses de chumbo em solução nutritiva. In: *CONGRESSO NACIONAL DE BOTÂNICA*, Belo Horizonte, 2013.

ACCIOLY, A.M.A.; SIQUEIRA, J.O. Contaminação química e biorremediação do solo. In: NOVAIS, RF *et al.* Tópicos em ciência do solo. Viçosa: Sociedade Brasileira de Ciência do Solo, 2000. p.299-352.

ALKHATIB, R. *et al.* Physiological and ultrastructural effects of lead on tobacco. *Biologia Platarum*. v.56, n.4, p.711-716, 2011.

AMABILE, R.F. *et al.* Girassol como alternativa para Sistema de produção do Cerrado. Circular Técnica, n.20, 2002.

ANDRADE, M.G. *et al.* Metais pesados em solos de área de mineração e metalurgia de chumbo. I - Fitoextração. Rev. Bras. Ciênc. Solo, v.33, n.6, p.1879-1888, 2009.

ANGELOVA, V.R. *et al.* - Potential of Sunflower (*Helianthus annuus* L.) for Phytoremediation of Soils Contaminated with Heavy Metals. BALWOIS - Ohrid, Republic of Macedonia. 2012.

ANJUM, N.A. *et al.* The Plant family Brassicaceae: contribuition towards phytoremediation. Sci. Business, p.342, 2012.

ATSDR-Agency for Toxic Substances & Disease Registry. 2007. Disponível: https://www.atsdr.cdc.gov/spl/previous/07list.html. Acesso: 10 abr. 2024.

BHARGAVA, A. *et al.* Approaches for enhanced phytoextraction of heavy metals. J. Environ. Manag., v.105, p.103-120, 2012.

BHAT, R.; REDDY, K.R. Challenges and issues concerning mycotoxins contamination in oil seeds and their edible oils: updates from last decade. Food Chem., v.215, p.425-437, 2017. doi: https://doi.org/10.1016/j.foodchem.2016.07.161

BOFFE, P. M. *et al.* Potencial fitoextrator da espécie vegetal *Helianthus annuus* L. em solo contaminado por chumbo. Rev. Esp., v.38, n.9, p.8, 2017.

BRIEGA, D. *et al.* Métodos de utilização do girassol. Ciênc. Vet., v.1, n.1, 2017.

BURTON, K.W. *et al.* The influence of heavy metals on the growth of Sitka-spruce in South Wales forests. II. Greenhouse experiments. Plant Soil, v.78, p.271-282, 1984.

CONAB. Companhia Nacional de Abastecimento. Acompanhamento da Safra Brasileira, v.8, n.7, 2021. doi: https://www.conab.gov.br/info-agro/safras/graos/boletim-da-safra-degraos

CONAMA. Conselho Nacional do Meio Ambiente. Resolução No 460. 2013. https://www.ibama.gov.br/component/legislacao/?view=legislacao&legislacao=13 1499.

CRUZ, C.D. GENES: a software package for analysis in experimental statistics and quantitative genetics. Acta Sci. Agron., v.35, n.3, p.241-276, 2013. doi: https://doi.org/10.4025/actasciagron.v35i3.21251

GARBISU, C.; ALKORTA, I. Phytoextraction: a cost-effective plant-based technology for the removal of metals from the environment. Bio.Technol., v.77, n.3, p.229-236, 2001.

GHOSHROY, S. *et al.* Inhibition of plant viral systemic infection by non-toxic concentrations of cadmium. Plant J., v.13, p.591-602, 1998.

HERZIG, R. *et al.* Feasibility of labile zn phytoextraction using enhanced tobacco and sunflower: results of five and one-year field-scale experiments in switzerland. Int. J. Phytoremediation, v.16, n.7, p.735-754, 2014.

KIVEVELE, T.; HUAN, Z.J. Influence of metal contaminants and antioxidant additives on storage stability of biodiesel produced from non-edible oils of Eastern Africa origin (*Croton megalocarpus* and *Moringa oleifera* oils). Fuel, v.158, p.530-537, 2015. doi: https://doi.org/10.1016/j.fuel.2015.05.047.

KOPSTSIK, G.N. Problems and prospects about the phytoremediation of heavy metal polluted solo: a review. Eura. Soil Sc., v.47, p.923-939, 2014.

KOSOBRUKHOV, A. *et al.* Plantago major plants responses to increase content of lead in soil: growth and photosynthesis. Plant Growth Regul., v.42, n.2, p.145-151, 2004.

KRUG, F. J. *et al.* Espectrometria de Absorção Atômica - Parte 1. Fundamentos e atomização com chama. 2006. Available at: <a href="https://edisciplinas.usp.br/pluginfile.php/4292094/mod\_resource/content/1/Apostila%20AAS-parte%201.pdf">https://edisciplinas.usp.br/pluginfile.php/4292094/mod\_resource/content/1/Apostila%20AAS-parte%201.pdf</a> >. Acesso em: 2 dez. 2023.

KUMAR, A. *et al.* Lead Toxicity: Health Hazards, Influence on Food Chain and Sustainable Remediation Approaches. Int. J. Environ. Res. Public Health, v.17, n.7, p.21-79, 2020.

MARQUES, L.F. Fitoextração de chumbo por girassol, vetiver, trigo mourisco, jureminha e mamona em áreas contaminadas. Areia: Universidade Federal da Paraíba, 2009.

NASCIMENTO, S.S. Tolerância de gramíneas forrageiras ao chumbo e sua disponibilidade no solo. Diamantina: Universidade Federal dos Vales do Jequitinhonha e Mucuri, 2013.

OJUEDERIE, O.B., BABALOLA, O.O. Microbial and Plant-Assisted Bioremediation of Heavy Metal Polluted Environments: a review. Int. J. Environ. Res. Public Health, v.4, n.12, p.1504, 2017.

PEREIRA, B.F.F. *et al.* Phytoremediation of lead by jack beans on a Rhodic Hapludox amended with EDTA. Sci. Agricol., v.67, n.3, p.308-318, 2010.

ROSEN, M.B. *et al.* A Discussion about Public Health and Lead in Drinking Water Supplies in the United States. Sci. Total Environ., v.590, p.843-852, 2017.

SALEEM, M. *et al.* Impact of lead tolerant plant growth promoting rhizobacteria on growth, physiology, antioxidant activities, yield and lead content in sunflower in lead contaminated soil. Chemosphere, v.195, p. 606-614, 2018.

SCOTT, A.J.; KNOTT, M. A cluster analysis method for grouping means in the analysis of variance. Biometrics, p.507-512, 1974.

SHARMA, P.; DUBEY, R.S. Lead toxicity in plants. Braz. J. Plant Physiol., v.17, p.35-52, 2005.

YEH, T.Y. *et al.* Chelator-enhanced phytoextraction of copper and zinc by sunflower, Chinese cabbage, cattails and reeds. Int. J. Environ. Sci. Technol., v.12, n.1, p.327-340, 2015.

XIA, Q. et al. Review on contaminants in edible oil and analytical technologies. Oil Crop Sci., v.6, n.1, p.23-27, 2021.

ZALEWSKA, M.; NOGALSKA, A. Phytoextraction potential of sunflower and white mustard plants in zinc-contaminated soil. Chil. J. Agricul. Res., v.74, n.4, p.485-489, 2014.

ZEITOUNI, C.F. Eficiência de espécies vegetais como fitoextratoras de cadmio, chumbo, cobre, níquel e zinco de um Latossolo vermelho amarelo distrófico. Camoinas: Instituto Agronômico, 2003.

ZEITOUNI, C.F. *et al.* Fitoextração de cádmio e zinco de um latossolo vermelho-amarelo contaminado com metais pesados. Bragantia, v.66, n.4, p.649-657, 2007.