Environmental impact of pollutants and the potential contribution to phytoremediation

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Sandra Ávila Gaspar[1], Denise Nagamatsu[2], Fábio de Almeida Bolognani[3], Morgana Teixeira Lima Castelo Branco[4], Alexandre dos Santos Pyrrho[5], Simone da Silva[6], Paulo Sérgio Torres Brioso[7] and Marcia Cristina Braga Nunes Varricchio[8]

ABSTRACT

This chapter revisited an experiment that evaluated elicitors for the germination of Vigna unguiculata (Cowpi Beans): the plant extract of E. tirucalli, the ultra-diluted and dynamized solution of Sulfur 5CH and the two in combination. At that time (2017), due to the significant results observed for the engorgement of seeds exuding an ammoniacal odor, the participation of the sodium/potassium pump, detoxifying enzymes from the thiol group and the suggestive participation of the nitrogenous route of oxidative stress was discussed, by monitoring the factors abiotic and detection of cyanuric acid in similar hydroponic solutions where Cowpea was cultivated, thus questioning the potential phytoremediation effect of test solutions as herbicides. Later, with the literature showing the association of the thiol group with mercury as another mechanism for phytoremediation, removing it from contaminated soil and water, it was also shown that very few plant species have this ability (two). So, based on the figure and scheme drawn up by environmental engineer Alexandre Pereira (2022), this potential was thought to be possible for the succulent species studied, E. tirucalli. From then on, the experiment was re-discussed thinking about the association with another likely mechanism for the results previously verified. Now also thinking about the potential participation of dairy ducts as structures capable of extracting and even bioaccumulating pollutants, in this case, mercury, since biotransformation was empirically observed in this species through phytovolatilization, which caused

[1] Agricultural Sciences from the Federal Rural University of Rio de Janeiro (UFRJ).
Apiary 4x4 Station
E-mail: sandra.avila.gaspar@gmail.com
[2] Lato senso Specialization in Homeopathy Pharmacy by the Hahnemannian Institute of Brazil (IHB).
Pharmacy from the Federal University of Rio de Janeiro/ SAPB-LIPAT Project.
E-mail: nagamatsud@yahoo.com.br
[3] Notorio Sapere in Homeopathy from the Open International University of Sri Lanka.
Institution: Homeopathy Service at the 7th Ward of the Santa Casa da Misericórdia General Hospital in Rio de Janeiro/Brazil.
E-mail: fabiobolognani@gmail.com
[4] Dr. in Sciences (Biophysics)/Carlos Chagas Filho Institute of Biophysics/Federal University of Rio de Janeiro (UFRJ).
Immunology Laboratory, Institute of Biomedical Sciences and Clementino Fraga Filho University Hospital (HUCFF), Federal University of Rio de Janeiro (UFRJ).
E-mail: morganalima@ufrj.br
[5] Dr. in Sciences (Biophysics) from the Federal University of Rio de Janeiro (UFRJ).
Institution: Laboratory of Immunoparasitology and Toxicological Analysis of the Faculty of Pharmacy (UFRJ).
E-mail: pyrrho@pharma.ufrj.br
[6] Post-Doctorate in Biodiversity and Biotechnology from Federal University of Amazonas (UFAM).
Amazon Biobusiness Center (CBA) & Institute of Technology and Education Galileo da Amazônia (ITEGAM).
E-mail: Simonida Silva@gmail.com
[7] Dr. in Sciences – Genetics/Agronomic Engineering at Federal Rural University of Rio de Janeiro (UFRRJ).
Official Phytosanitary Diagnostic Laboratory – Phytopathology at Federal Rural University of Rio de Janeiro (UFRRJ).
E-mail: brioso@bighost.com.br
[8] Post-Doctorate in Intellectual Property of Brazilian Traditional Ethnic Knowledge (INPI).
Dr. in Sciences (Vegetal Biotechnology) from the Federal University of Rio de Janeiro (UFRJ).
Immunology Laboratory, Institute of Biomedical Sciences and Clementino Fraga Filho University Hospital (HUCFF), Federal University of Rio de Janeiro (UFRJ).
E-mail: varichio2@gmail.com

intoxication through inhalation of researchers, using an odorless substance released (NAGAMATSU et al., 2019). To conclude, therefore, based on practical experience with this plant species, this appears to be a viable mechanism that will be investigated in future water cultivation trials of Vigna unguiculata, this time with micropropagated Euphorbia tirucalli to be carried out in partnership with agricultural engineering, agronomic (phytosanitary) and environmental. The aim is through research in plant biotechnology to study the effects on phytoremediation, the consequent interdisciplinary promotion of environmental health.

Keywords: Vigna unguiculata, SUD 5CH Sulphur, Euphorbia tirucalli, Phytoremediation, Heavy metals.

INTRODUCTION

According to the World Health Organization (1993 In RIBEIRO, 2004):

> "Environmental health is all those aspects of human health, including quality of life, that are determined by physical, chemical, biological, social and psychological factors in the environment. It also refers to the theory and practice of valuing, correcting, controlling and avoiding those environmental factors that could potentially harm the health of current and future generations" (WHO, 1993).

The large number of environmental factors that can affect human health is an indication of the complexity of existing interactions and the range of actions necessary to improve environmental factors that determine health. However, environmental improvement programs have actions that are quite different from those of medical care, even though they cannot be separated from them (RIBEIRO, 2004).

As already reflected by professor Helena Ribeiro (2004), concerns about environmental aspects, both in relation to Public Health and in relation to other characteristics of different social groups (social, cultural and economic organization), have existed since the beginning of human history and constituted an important analytical basis for social thought in the past. Today, these questions are once again attracting the attention of different sciences. Clarence Glacken (1967) summarizes these concerns in three major questions, which have been highlighted at different moments in human history:

1. What is the meaning of human creation and what is the conception of the earth? In other words, the earth was created for human beings?
2. What is the influence of the physical environment - the environment - on the characteristics of human beings and societies?
3. How have human beings been transforming the earth?

Despite this large-scale and serious problem about pollutants in nature and non renovable resources, novel models of investigations may be able to answer complexity present in nature, aiming reduction of impacts of pollutants upon nature but also to living beings. So, some types of studies were developed in this theme all over the years focusing in water – removal of pharmacological waste and hipersalinity (KATHAR et al., 2023; GASPAR et al., 2023) - and in soil – by attended local cultives aiming to decrease toxicity of plants species (VARRICCHIO et al., 2006), besides persistence of endocrine disruptors (HANSEL-MARTINS et al., 2023; 2024; GASPAR et al., 2024), once it is known that some plant species can clean soil and water from heavy metal pollution from industries in a process of Phytoremediation, when is able to degrade, remove or stabilize toxic substances from contaminated soil or water, as previously discussed by KATHAR et al. (2023).

There are several hyperaccumulator plant species, that is, plants that can store metals that they do not need for their development (or that metabolize them for less harmful species) favoring the removal of these elements in nature. Because, if these polluting heavy metals are not removed or prevented from migrating, they will accumulate in edible vegetables or contaminate the water table, causing problems on contact such as allergic dermatitis, inhalation of volatile substances, resulting in perforations of the nasal septum, intoxication, headache, nausea and fainting and chronic poisoning can trigger cancer in animals, humans and non-humans. Using this technique, soil and water contaminated by petroleum derivatives can also be recovered (DE SOUZA, 2021), by herbicides (GASPAR et al., 2023; 2024, a), besides removal of waste of medicines (KATHAR et al., 2023).

The rate of reduction of pollutants will depend on factors such as climate, nature of pollutants and characteristics of plants (AGUIAR et al., 2012). Regarding pollution from heavy metals, their high concentration in the environment does not always indicate a high accumulation by plant species, as the interpretation of results can be complicated, the removal system through the roots used by plant species has not yet been developed. is fully clarified (MULGREW & WILLIAMS, 2000; AGUIAR et al., 2012). Mulgrew & Williams (2000) in their study demonstrated that plant species have been used as biological indicators and bioaccumulators of various pollutants, many of which are removed from the air, soil and water, thus reducing pollutant levels in the environment.

Some plants are suitable for phytoremediation (DUSHENKOV et al., 1995; AGUIAR et al., 2012). Plants that are grown hydroponically tend to develop more robust root systems, providing a greater surface area for the absorption of pollutants (MANT, 2001; AGUIAR et al., 2012). Brazil has a vast number of plant species with great potential for phytoremediation and hydroponics; however, these species are still little explored and their studies are almost scarce regarding their potential as phytoremediators (AGUIAR et al., 2012).

Among environmental pollutants, there is mercury. The expression “mercury cycle” refers to the transformations that the metal mercury can undergo in the environment, such as the formation of methyl mercury, harmful to living organisms and the bioaccumulative form of the metal in them. It is a metal that is liquid at room temperature, used both in industry, through various products, and also in mining, for example.

The chemical element mercury is symbolized by Hg, a reduction of the Greek word Hydrargyrus, in Latin, it represents liquid silver, given that mercury was known in this way in ancient times. However, the name “mercury” is a legacy of the Egyptians who, knowing seven stars (Sun, Moon, Venus, Mars, Saturn, Jupiter and Mercury), promoted the association with the seven metals known until then: gold, silver, copper, iron, lead, tin and mercury, respectively. The associations were not by chance: Mercury, as it apparently moves faster than the other planets, would adequately represent the volatile metal, in addition to appearing in liquid form at room temperature (DAMAS, 2014).

In the formation of mercury metal, the distribution of the various species that enter the aquatic system is guided by physical, chemical and biological processes, which occur in the air/water and water/sediment relationships. The conversion between these different forms is the basis of the complex distribution pattern of mercury in local and global cycles and its biological enrichment. Regardless of the processes that regulate these cycles, the mercury that enters different environmental compartments can be inorganic and/or organic (BISINOTI & JARDIM, 2004).

Thus, the methylation of mercury in the aquatic environment is possible due, among others, to sulfate-reducing bacteria. Methylation can be separated into two pathways, biotic and abiotic. As for the first, it is mediated by microorganisms or fungi and is related to methylcobalamin, known as vitamin B12, which is a coenzyme capable of transferring the methyl group to the Hg2+ ion. Several bacteria commonly present in the aerobic environment are sensitive to mercury methylation, such as Aerobacter aerogenes, Bacillus megaterium and Candida albicans, for example (MICARONI; BUENO & JARDIM, 2000).

In inorganic form it can be found under three different oxidation states: elementary Hg (Hg0), which is found mainly in gas form, the mercurous ion (Hg2 2+), a form that is not very stable in natural systems, and the mercuric ion (Hg2+). In organic form, the mercuric ion is covalently linked to an organic radical, with methyl mercury (CH3Hg+) and dimethylmercury ((CH3)2Hg) being the most common, or with natural organic ligands. In addition, there is a generic biogeochemical cycle in which methyl mercury and Hg2+, dimethylmercury and Hg compounds are interconverted in atmospheric, aquatic and terrestrial systems (MICARONI; BUENO & JARDIM, 2000).

A small portion of the Hg0 that reaches the atmosphere is converted into water-soluble species (probably Hg2+), which can be re-emitted into the atmosphere as Hg0, through deposition on soil or exchange at the air/water interface. The atmospheric cycle involves retention of Hg0 in the atmosphere for long periods; consequently, this compound can be transported over great distances. The sediment at the bottom of the oceans is considered to be the place where mercury is found abundantly deposited in insoluble form (HgS) (MICARONI; BUENO & JARDIM, 2000).

For this reason, it is believed that fires release a significant portion of mercury through volatilization or sulfate, capable of stimulating methylation processes. Thus, mercury methylation can occur in both soils and sediments, aerobically and anaerobically (DAMAS, 2014). However, it can also be associated with sulfur, which forms a cinnabar ore (HgS), which can then be heated and condensed to obtain metallic mercury. It is also found in volcanic eruptions, natural evaporation and mercury mines, used in various things, as previously mentioned (MICARONI; BUENO & JARDIM, 2000; BISINOTI & JARDIM, 2004).

However, due to the bioaccumulation of methylmercury, methylation prevails over demethylation in aquatic environments, as the reaction can be reversible. Once formed, methylmercury enters the food chain through rapid diffusion and strong binding with the proteins of living beings in the aquatic environment, promoting its maximum concentration in fish tissues at the top of the aquatic food chain due to biomagnification (BISINOTI & JARDIM, 2004).

The process of biomagnification is understood as the increase in the concentration of a substance or element in living organisms, as it travels through the food chain and begins to accumulate at the highest trophic level; bioaccumulation, biomagnification, bioconcentration, trophic magnification. Trophic magnification is a phenomenon that occurs when there is progressively greater accumulation of a toxic substance from one trophic level to another along the food chain due to a reduction in biomass. Therefore, consumers have a higher concentration of toxic products than producers (BISINOTI & JARDIM, 2004; KEHRIG et al., 2011).

Furthermore, eutrophication produces changes in water quality, including the reduction of dissolved oxygen and aquatic biodiversity and, consequently, the loss of biodiversity quality, as it can lead to the death of fish. In this way, it was possible to notice the large amount of mercury in predatory fish, as they are at the top of the food chain in the waters, mainly in tributary areas of the Amazon River. However, it is possible to analyze its bioaccumulative aspects from the planktonic community to predatory fish (KEHRIG et al., 2011).

However, poisoning can occur in several ways, which include inhalation, digestive and cutaneous. In the lungs, its retention varies from 74% to 76% at an environmental concentration of 100 MG/m3. When the metal reaches the lungs, due to hematosis, via the serum it is distributed to the rest of the body, and can accumulate in the kidneys, central nervous system, liver, bone marrow, placenta, among other organs. The contamination reference point is based on exposure through methyl mercury, an organic form of this metal. Absorption through food is around 15%, while in the form of methyl mercury it is around 90%. Both forms of mercury, inorganic and organic, quickly bind to glutathione, present in most cells in the body. Mercury in its inorganic form is not captured by the body, being eliminated through feces, while organic mercury is eliminated through saliva, kidneys and skin (through sweating). Methyl mercury is eliminated through feces and can also be eliminated by epithelial cells through exfoliation (ROCHA, 2009).

In ecology, it is known that pioneer plants are those plants that originate from inhospitable, uninhabitable places, such as grasses in a desert. They are characterized by the fact that they can cope with the harsh conditions of bare land and, in addition, they have a very large annual seed production. They are described as species that survive due to their high reproductive capacity. They die and decompose, replacing the soil to increase the humus content. This leads to an increase in the soil's ability to hold water and mineral nutrients. As a result, pioneer plants create a gentle microclimate. This creates the basis for a migration of plants that are efficient in competing for light, nutrients and water. Although in the literature, Euphorbia tirucalli is not considered a pioneer species, the millennial existence and survival of the genus since Pangea, in addition to its cosmopolitan distribution, encourages us to persist in studying this intriguing species. E. tirucalli Avelós, is classified as follows: “Aveloz – euphorbiaceae. ORN – CV – MED (Ornamental, Cerca Viva, Medicinal by EMBRAPA (RIBEIRO, 2010, p. 19).

Even though it is considered a toxic species, it is regularly used by traditional, ethnic communities and indigenous peoples around the world (VARRICCHIO et al., 2008; CAMPOS et al., 2016) and even in popular use (MOLIN et al., 2015). Laboratory investigations have already investigated its angiogenic (BESSA et al., 2015), genotoxic and cytotoxic action on human leukocytes (MACHADO et al., 2016) in addition to its effect on gene expression in laryngeal carcinoma (FRANCO-SALA et al., 2016) , in vitro investigation of mechanisms in malignant neoplasia (SILVA et al., 2019).

An interesting review on phytochemistry and pharmacological activities was carried out by MALI & PANCHAL (2017) and new substances with antimicrobial activity produced by the roots of E. tirucalli from the Brazilian northeastern Caatinga biome were verified by LIMA (2019), described below. The chemical and biological investigation of three plant species collected in the caatinga: Euphorbia tirucalli Linn., Bredemeyera floribunda Willd and Bredemeyera brevifolia Benth. The LC-MS analysis, in negative mode, applied to the extracts of the three species made it possible to evaluate the phytochemical profile of the species and thus verify flavonoids as the predominant class of metabolites (LIMA, 2019).

The chemical study of hexane extracts from the aerial part and ethanolic extracts from the roots of *E. tirucalli*, applying chromatographic techniques, allowed the isolation of five constituents: two already isolated in the species, β-amyrin and 4-O-methyl-gallic acid, in addition to three new compounds for the species, ampelopsin, myricetin and 3,3'-dimethoxyelagic acid-4-O-αraminopyranoside. The evaluation of the antioxidant activity of *E. tirucalli* extracts against DPPH and ABTS tests made it possible to verify that the ethanolic extract of the roots showed better efficiency against the two radicals, while among the isolated compounds, myricetin showed better inhibition of the radicals with IC50 of 22.62 μg/mL for DPPH and 53.22 μg/mL for ABTS (LIMA, 2019).

The evaluation of antimicrobial activities revealed that only the ethanolic extracts presented antibacterial and antifungal potential, while all isolated compounds showed an inhibitory effect against strains of *Staphylococcus. aureus, Escherichia coli, S. brasiliensis* and *Candida Albicans*, with a greater effect on myricetin and ampelopsin (LIMA, 2019).

In parallel, the description of chronic, unexpected nasal intoxication of a volatile, odorless substance during daily contact with *E. tirucalli* for experiments provided clues to think about possible pathogenic effects and, consequently, potential biological applications (NAGAMATSU et al.

, 2019). Furthermore, it made us question its potential for phytotransformation through phytovolatilization, thus beginning our study in this area of phytoremediation research (KATHAR et al., 2023). Could there be an association of mechanisms if it were to play a phytoremediation role? Could this mechanism determine not only the type of phytoremediation, but which waste it can filter?

AIM

Revisit an experiment already carried out (GASPAR et al., 2024) to deepen the discussion regarding yet a new and possible mechanism of action in phytoremediation.

METHODOLOGY

The assay aimed to evaluate the biological effect of the total ethanolic extract 30% *E. tirucalli* and HUD 5DH Sulfur on the germination of cowpea (*V. unguiculata).* As methodology, assisted cultives of *V. unguiculata* in 200 ml of mineral water were carried out. Three seeds per pot, in three pots, in triplicate per row, with one more pot (N = 30). It was administered separately: 1 drop of total extract of *E. tirucalli*, HUD5DH, and respective controls prepared by the Hahnemannian method of multiple flasks. The action/time curve was verified on the first, third and seventh day of cultive: pH, free chlorine, total alkalinity, color, turbidity and presence of cyanuric acid were measured by the colorimetric method. Temperature measured by an Infrared thermometer B-Max. Morphology and germination were observed by ANOVA statistical programme (In GASPAR et al., 2017).

RESULTS

Results showed that it had an inhibition on the germination of *V. unguiculata* ($p<0,05$), curiously associated with engorgement of the seeds, an evidence of metabolic route deviation. Hormetic ponderal phytoextracts with also HUD 5DH Sulfur test solutions, induced cyanuric acid at water where seeds were suggestive of protein breakdown, possibly due to erosion of the bean skin. Cyanuric acid is commonly used as pesticide, mosquito inseticide and mainly as a repellent of invasors plant species and their microbiological pathogens, through allelopathy mediated by radicule secretions emissions by roots (BEZERRA, 2015; HANSEL – MARTINS et al., 2023).

Cyanuric acid is also able to compete with some chemicals. It is a precursor to polyesters, polyurethanes, bleaches, disinfectants and herbicides. This last function, in turn, can be used in plant biotechnology in the bioremediation of soils contaminated with herbicides, used to combat invasive weeds. So, this experiment will be repeated separately to further study the mechanisms. Under this vision, the use of herbal extracts and diluted solutions (infinitesimals) seeks to become an alternative for the cultivation of small farmers, reducing their exposure to insecticides and pesticides currently used in industry, such as organochlorines, which are considered endocrine disruptors (HANSEL – MARTINS et al., 2023) that can cause serious health problems, including mental disorders (HANSEL – MARTINS et al., 2024).

Attended cultive of *Vigna unguiculata* elicited with *E.tirucalli* extract and HUD 5CH Sulphur as elicitors: Biotechnological potential on phytoremediation?

DISCUSSION

ATTENDED CULTIVE OF *VIGNA UNGUICULATA* ELICITED WITH *E.TIRUCALLI* EXTRACT AND HUD 5CH SULPHUR AS ELICITORS

Was it found one more biotechnological potential on phytoremediation? A biotechnological potential for bioremediation of soils contaminated with herbicides, a response through nitrogen oxidative stress, was evidenced. Assays on this route will be carried out in phytopathology laboratory and cyanuric acid will be the pathogenetic marker of biological activity for HUD solution tested (GASPAR et al., 2024), as already evidenced at this present assay. Furthermore, herbal extracts obtained from roots (VARRICCHIO, 2008; LIMA, 2019) and high ultradiluted sucussioned solutions may be an alternative to the cultive of small farmers, reducing exposure to pesticides and soil contamination. So, there are phytoremediation potential but, perhaps, also a bioremediation potential (GASPAR et al., 2017; 2024a).

Furthermore, it might be another possible explanation and aplication of these prepares from now on this preliminar observation. This assay was evaluated through abiotic factors whose, naturally, suggested mechanism of action through sulfur enzymatic pathway, object of several of our studies along years (APOLINARIO et al., 2000). There is a relationship of mercury with the covalent bonds to thiol groups, cellular enzymes in lysosomes and mitochondria is one of the factors that contribute to knowing the level of toxicity, due to the fact that the metal interrupts metabolism and cellular functions (ROCHA, 2009).

However, there is no exact target for interaction preference, because thiol groups exist in proteins that have extracellular and intracellular membranes, and can be related in both function and structure. As a consequence of toxicity, several mechanisms can be enumerated, such as: inactivation of enzymes, structural proteins, transport processes and even changes in the permeability of the cell membrane. Furthermore, it is worth mentioning that, despite its low affinity, it can bind to the carboxyl, amide, amine and phosphoryl groups of enzymes. There are some studies that relate toxicity to increased permeability of the blood-brain barrier, inhibition of proliferation and formation of microtubules, interruption of protein synthesis, among others (ROCHA, 2009).

One of the effects of mercury on the body is related to oxidative stress, due to the loss of levels of glutathione, superoxide dismutase, catalase and glutathione peroxidase, thus providing less protection to cells. Furthermore, mercury can induce lipid peroxidation, mitochondrial disorders and changes in the heme group. Furthermore, mercuric chloride can cause depolarization of the inner mitochondrial membrane, increasing the formation of peroxidase. Another factor related to increased oxidative stress is related to changes in mitochondrial calcium homeostasis, influenced by mercury chloride in kidney cells. Consequently, biochemical changes occur, such as excretion of porphyrins in the urine (ROCHA, 2009).

One of the main causes of the affinity of methyl mercury in myelin is its lipid solubility, which inhibits the excitability of neurons. Furthermore, HgCl2 has an inhibitory activity when it binds to the sulfhydryl groups of calcium transport proteins, consequently, muscle contraction does not occur, in addition to the inhibition of neurons. One of the main targets of methyl mercury in the body are microtubules, which are essential in cell division and migration. Therefore, when they are affected, they block the development of the nervous system. Additionally, methylmercury inhibits GABA receptors (Gamma-AminoButyric Acid receptors), which are located on Purkinje cells and neurons (ROCHA, 2009).

So, returning to the point of the article, the dramatic engorgement of the seeds under the effect of SUD 5CH suggested that it was able to act on the sodium/potassium pump, but it also may have be able to act as a phytoextractor. Indeed, there is the possibility that phytoextractive action was increased and even noticed due to the effect of ultradilution and succussion by Samuel Hahnemann's multiple flask method, HUD mineral, as already discussed by GASPAR et al. (2024a).

Novel assays with raw extracts obtained from Cowpea bean of differents assisted cultive and micropropagation will go on being evaluated through abiotic factors as this present assay discribed and about their mechanisms of action through sulfur enzymatic pathway, when under effect of HUD of *Mercurius solubilis* and it's controls, not more as a homeopathic effect, but an isopathic effect. The relationship of mercury with the covalent bonds to thiol groups, cellular enzymes in lysosomes and mitochondria is one of the factors that contribute to knowing the level of toxicity, due to the fact that the metal interrupts metabolism and cellular functions (ROCHA, 2009) and it will be used as a pathogenetic biological marker to understand the mechanism of action (HANSEL-MARTINS et al., 2024; GASPAR et al., 2024, a).

In turn, *Vigna unguiculata* (Caupi Bean) is the bean with the highest concentration of iron and proteins still used by quilombolas and northeastern people in Brazil (GASPAR et al., 2017; MUSMANNO et al., 2019), some of them exposed to environmental pollutants as herbicides and agrotoxics (HANSEL-MARTINS et al., 2023; 2024).

Although its ethnobothanic use (MOTA et al., 2022; MOTOO et al., 2022; BINCKLEY & ZAHRA, 2023) there is a point of use of *E. tirucalli* to Malary (VARRICCHIO et al., 2008; VEIGA & SCUDELLER, 2015), studies regarding its antitumor activity for solid tumors remain in the spotlight (MUNRO et al., 2015; PALHARINI et al., 2017), with increasing emphasis on research into its antioxidant and antimicrobial activity (LE et al., 2021).

As Alexandre Pereira ponders, Phytoremediation (phyto = plant; remedium = cure, restoration) is the process in which plants are used for environmental decontamination, through removal, reduction, degradation and containment of the contaminant. In this process, various plants can be used, including aquatic plants and algae. Phytoremediation strategies have the potential to treat a broad spectrum of contaminants. Brazil has great potential to use phytoremediation in the treatment of contaminated areas due to its high biodiversity and climate, which are favorable to biological processes to remove contaminants (PEREIRA, 2022).

The technique is often used in the remediation of heavy metals, radionuclides, as well as recalcitrant organic pollutants. For satisfactory performance in metal remediation, it is desirable that plants have tolerance to the contaminant, good metal absorption or adsorption capacity, high growth rate, deep root system, easy harvest. Knowledge of potentially phytoremediating plant species is an initial and fundamental step for decontamination studies and projects (PEREIRA, 2022).

Therefore, considering the great plant diversity in Brazil, this review aimed to survey Brazilian flora species that potentially phytoremediate heavy metals. To this end, the capacity and tolerance, as well as the predominant phytoremediation process in the decontamination of heavy metals by different species and botanical families native to Brazil, were investigated in the literature. 108 species of native plants were found used in metal phytoremediation studies. The identified species belong to 42 different botanical families. With 19 species, the Fabaceae family is the most represented in studies, showing potential to phytoremediate 8 heavy metals. Asteraceae also stood out with 9 species remediating 4 types of metals, and Poaceae with 5 species studied remediating 6 metals. (PEREIRA, 2022).

In relation to metals, it is clear that most phytoremediation studies used copper and zinc as decontamination targets. Although there is much to be explored regarding the use of species from Brazilian flora, the results are satisfactory, contribute to the advancement of knowledge, and already offer a significant number of possible plant species for metal remediation. The plant species used for the phytoremediation of heavy metals are called metallophytes, which have good tolerance to high levels of metals. Another definition is that they are plants that have evolved, adapted and even thrived in soils rich in heavy metals (BOTHE et al., 2011, SHEORAN et al., 2011 In PEREIRA, 2022).

Metallophytes are divided into three categories: metal excluders, metal indicators and metal hyperaccumulators. Excluders accumulate heavy metals from the substrate in their roots, but restrict their transport and entry into their parts. Indicators accumulate heavy metals in other aerial parts.

Hyperaccumulators are plants, which can concentrate metals in their above-ground tissues to levels much higher than those present in the soil (MALIK and BISWAS, 2012 In PEREIRA, 2022).

In Brazil, several research teams are already focusing on this process, on a laboratory scale. For example, a type of fern is studied for arsenic phytoextraction and cowpea is used for the recovery of soils containing herbicides. The whiner for the recovery of waters containing petroleum derivatives; castor, sunflower, Amazon pepper and tobacco for the treatment of soils containing cadmium, lead, copper, zinc and nickel. One of them is *Thalaspi caerulescens*, from the Brassicaceae family, and it is one of the main focuses of research on phytoremediation (AGROAMBIENTE, 2023).

In the US, GM Indian mustard has been used to treat arsenic-containing soils in California. In Canada, lead, copper and zinc were removed from the soil thanks to 3 species: willow, Indian mustard and fescue (pasture grass). The University of Georgia is developing 60 transgenic cotton plants to clean soil contaminated with mercury on land in the city of Danbury, in the US state of Connecticut, where they will remove mercury deposited by an old hat factory (AGROAMBIENTE, 2023).

Most plant species have the capacity to absorb, translocate, immobilize and/or degrade contaminants (CUNNINGHAM & OW, 1996), thus being able to repair contaminated environments. Plants act in such a way that they are able to modify the physical and chemical properties of contaminants in the soil by releasing exudates through their roots. These strategies that allow phytoremediation can occur through a single or a combination of the following basic mechanisms: removal, accumulation and withdrawal of the contaminant with the removal of the plant; contaminant removal and degradation; removal and volatilization to the atmosphere; or facilitating soil treatment (accumulation, immobilization and stimulation of rhizosphere microbiota). Therefore, the types of phytoremediation carried out by plants adapted to the presence of heavy metals can be classified as phytoextraction, rhizofiltration, phytostabilization and phytovolatilization (In PEREIRA, 2022).

Regarding the advantages, phytoremediation stands out for: having good public acceptance, low cost, and can be used in large areas; because it is a natural process that requires little mechanical work and because it makes the place more pleasant. Taking into account these characteristics and the expected low cost, phytoremediation can be used on a larger scale than would be possible in the case of other methods (PIRES et al. 2003).

The time required is generally long due to the plant's growth and development cycle. There is the possibility of contamination of the food chain, the phytodegraded toxic compound can become an even more toxic compound (LAMEGO & VIDAL 2007, PROCÓPIO et al., 2009 In PEREIRA, 2022). Whether the treatment process takes longer or longer will depend on some factors or conditions such as the species and number of plants; the factors (physical, chemical and biological) of the soil; the weather; the rhizosphere; the type and concentration of the contaminant. The size and depth of the contaminated area also influence. So, types of phytoremediation were shown by the environmental engineer Alexandre Pereira (2022), named as Table 1 and Figure 5 in his work.

Table 1: Summary of phytoremediation techniques.

Tabela 1 - Resumo das técnicas de fitorremediação.

Resumo das diferentes técnicas de fitorremediação.	
Técnica	**descrição**
Fitoextração	Acúmulo de poluentes na biomassa colhível, ou seja, pode retirada.
Fitoestabilização	Limita a mobilidade e biodisponibilidade dos contaminantess no solo através das raízes.
Fitoestimulação	Estimula a planta para a biodegradação microbiana dos contaminantes, mediante exsudatos radiculares e/ou fornecimento de tecidos vegetais.
Fitovolatilização	Conversão de poluentes para a forma volátil e posterior liberação para a atmosfera.
Rizofiltração	Absorção e adsorção de contaminantes na rizosfera por microorganismos rizosféricos.

Figura 5: Phytoremediation techniques.

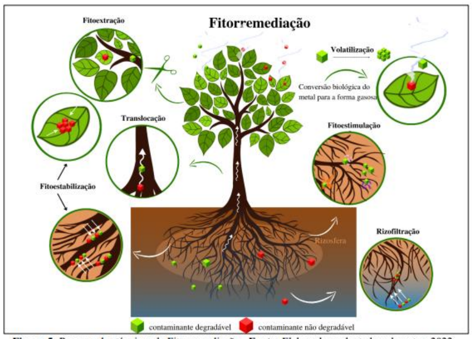

Figura 5- Resumo das técnicas da Fitorremediação. **Fonte:** Elaborado e adaptado pelo autor, 2022.

Source: Prepared and adapted by PEREIRA (2022).

As Alexandre Pereira (2022) reflects, when selecting plants that have phytoremediation capacity, it is desirable that they present some indicator characteristics (PIRES et al. 2003). It is unlikely that a species or variety brings together all these characteristics, however, one should look for those that bring together the greatest number of desirable attributes (MILLER, 1996). The main desirable characteristics are: suitability for the climate (temperature and humidity), tolerance to contaminants; resistance to pests and diseases; the ability to absorb, concentrate and/or metabolize the contaminant; retention of the contaminant in the roots (phytostabilization); rapid growth and biomass production; practicality in harvesting, removal if necessary; ease of acquisition or propagation; ease of control or eradication; stimulation of soil biota; depth of the root system; natural occurrence in contaminated areas (CUNNINGHAM et al., 1996; NEWMAN et al., 1998; ACCIOLY & SIQUEIRA, 2004. All in PEREIRA, 2022).

The importance of studies of native plants, whether at a local or regional level, is due to the fact that they are already adapted to the climate and the chemical properties of the soil. Native and exotic plant species are classified according to their origin. According to the IBF – Brazilian Forestry Institute, a native species is defined as a plant that is natural, originating in the region in which it lives, that is, that grows within its natural limits including its potential area of dispersal (PEREIRA, 2022).

With the exception of water hyacinth (aguapé) and cattail (taboa), no other plants were found that naturally hyperaccumulate Hg, but plants that can develop mechanisms to deal with the presence of this contaminant in the environment, as previously proposed by WAGNER-DÖBLER (2013).

Indeed the fourth circulation through laticiferous ducts of *E. tirucalli* and its "roots in hair" may be the way to study this strongly suggestive potential of action. So, also HUD might be a viable alternative, being discussed through other reasoning (VARRICCHIO, 2008).

What toxicology at primary effect and secondary effect obtained thorough HUD administration can show to us?

PRIMARY AND SECONDARY TOXICOLOGICAL EFFECTS (PATHOGENESIS)

SYMPTOMS OF MERCURY POISONING

In humans, contact with mercury can cause everything from mild symptoms, such as itching and redness of the skin and eyes, to serious interference with cellular metabolism, in case of prolonged exposure. Know the main symptoms of mercury poisoning: Fever, tremors, allergic reactions to the skin and eyes, drowsiness, delirium, muscle weakness, nausea, headache, slow reflexes, poor memory, malfunction of the kidneys, liver, lungs and nervous system.

- Pathogenesy HUD of mercurium black oxide (ILLAH, 2019; VARRICCHIO, 2024): MERCURIUS OXYDULATUS NIGER (*Mercurius solubilis Hahnemanni*)

"Dizziness more when sitting than when standing, lack of clarity and darkness before the eyes, especially towards dusk. Dizziness as if he were drunk; he gets up and walks from one side of the room to the other staggering, then anxious heat breaks over him, with nausea; Dizziness, cold hands with feverish trembling, then confusion of the head, forcing him to lie down. A type of dizziness; when lying down he feels as if he were swaying longitudinally. It takes away the sharpness of his intellect, makes him bewildered, he does not hear what is said to him, he cannot retain well what he reads, and he is apt to make mistakes when speaking. Unconsciousness and mutism; In the evening, a dull painful sensation in the head until he falls asleep; loud talk distressed him. Pressive headache as if it were tightly tied. As if the head was going to explode, at the same time the eyes were watering. Dilated pupils (after 1 h). Black insects or flies always seem to be flying before the view. Heat in the eyes and tearing. Painful in both eyes as from sand. Sore in eye, when it is moved;

It also hurts when touched. Itchy eyeballs. He can barely hear anything and yet everything resonates loudly in his ear. Bleeding from the nose (epistaxis) during sleep. Gums are painful when touched and when chewing, particularly hard food. In her sleep at night she grinds her teeth, and bites them so hard that it causes pain, which awakens her. Loss of speech and voice; she can't sleep and feels very exhausted; but she has an appetite for all kinds of food, and a thirst for beer; Swallowing is difficult and painful, as if it has burned the back of your throat. Pain and swelling of the salivary glands. Swelling of the ganglia of the neck and parotids, so that the jaws are closed, and cannot be moved at the cost of pain. Metallic taste in his mouth that almost makes him vomit. Salty expectoration. Sweet taste on the tip of the tongue. Putrid, very unpleasant taste in the throat. He has an aversion to sweet things. Extreme aversion to fresh meat. Aversion to coffee. Aversion to butter. No appetite for any hot food, only for cold things, bread and butter, etc. More thirsty than hungry, and constant slight shivering from the cold. After eating, violent sobbing. If he eats just a little too much, he becomes so bad-tempered that he can hardly bear it. Pain in the abdomen and a lot of noisy flatulence. A terrifying stab perpendicularly from the middle of the lower abdomen down to the anus. Inguinal bubo. Bloody stools, with painful acrid sensation in the anus. Brownish-red urine, very frequent with burning, burning pain. Pulling pain in the testicles and groin. Painful erections. Seminal emission without voluptuous dreams. Nocturnal seminal emission, mixed with blood. Suppressed menstruation. Harsh cough. Blood expectoration when walking outdoors. Periodic pain in the breasts, as if something in them was about to suppurate. Sharp needles in the spinal column, between the shoulder blades, in bed after midnight. Painful stiffness of neck, so that she cannot turn her head around, with heavy feeling in it. Visible fasciculation. Fatigue and restlessness in the legs, in the evening. At the same time heaviness, indolence and drowsiness. Jaundice with burning itching over the abdomen. Exhausted, not inclined to do anything and irritated. Fainting with an indescribable discomfort in the body and mind, which forces you to lie down. He doesn't like to talk, he can't read, his head is confused; he can't work, and falls asleep when he sits down. Exhaustion with sadness. Great exhaustion at dusk. First drowsiness, then insomnia. Sleep interrupted by frightened jerks, palpitations of the heart, and terrible images (e.g., as if he feared an epileptic fit). Very willing to sleep; sleeps very long and very soundly. Lots of delirious sleep talk. Sleeping is more unpleasant than pleasant for him. Anxious dreams with palpitation of the heart and yet he cannot wake up. Scary dreams at night, as if he fell from a height. Restless nights, dreams of highway robbers. Vivid dreams of the day's occupations; Dreams about water danger. Scary dreams of shooting. Violent thirst for cold drinks, particularly for fresh water. In the morning, immediately after rising, cold and shivering. Febrile attacks, particularly at night. Anxiety as if he had committed a crime. He doesn't rest, and needs to go from here to here, and can't stay in one place for long. He thinks he is losing his reason, that he is going to die; with fancies of the imagination, e.g. (e.g.), he sees water flowing where there is none (in the morning). With absence of thought he feels as if he has done something bad. No desire for serious work. He doesn't have the courage to live. He was indifferent to everything, even what he loved most. Throughout the day great seriousness with great indifference; he got irritated when others laughed at trifles, and at the same time he was extremely indifferent to everyone around him. Everything is unpleasant to him, even music. For no reason he is very dissatisfied with himself and his condition. Throughout the day, depression of spirit combined with anxiety; he always thought he was going to hear something unpleasant. All day long, grumpy; he was extremely laconic and serious. Throughout the day, nervous and moody; he believed that all his efforts would ultimately fail. Irritable, irascible, cheeky spirit. Very nervous and intolerant, easily irritated, very suspicious. Fights with anyone, stubborn, quarrelsome. Contentions. All day long, grumpy and suspicious; he almost insulted those around him, and considered them all as his greatest enemies. All day nervous, as if in contradiction and dissatisfied with himself, and he had no desire to talk and play. He is overwhelmed, acts like a clown, and does stupid, meaningless things; During his senseless acts he felt very much like crying, and when this paroxysm passed, he felt very exhausted. I almost involuntarily cry with relief."

The wealth of symptoms observed and methodologically collected from the secondary response was increased and opened a range of possibilities for the study of action mechanisms in search of effective contributions. Having noted the breadth of scope and similarity for its use for the current period, in accordance with the law of similarity to contemporary society. Will it be intoxicated?

MERCURY EXPOSURE ROUTES

Mercury is a liquid, heavy metal that, under normal conditions, is found in low concentrations in the environment. It is naturally released due to erosion processes and volcanic eruptions. Environmental contamination by mercury is, therefore, the result of human actions involving this element. The main anthropogenic sources of mercury are (AGROAMBIENTE, 2023):

- Burning coal, oil and wood: the process releases the mercury contained in these materials;
- Manufacture of products that use the substance as raw material, such as thermometers and fluorescent lamps;
- Inadequate disposal of mercury after its use in industrial processes, such as chlor-alkali production;
- Incorrect disposal of electronic products containing mercury;
- Gold mining, in which the substance is used to facilitate the particle separation process.

Brazil does not produce mercury, as it does not have reserves of cinnabar (a commercially exploited form of it). Therefore, the country imports it mainly from the USA and Spain. According to a study, the main sources of environmental contamination by the substance in Brazil are: Industrial effluents from the manufacture of caustic soda and gold mining in the Amazon region (AGROAMBIENTE, 2023).

The burning of large forest areas in the Amazon region is highlighted in the Ministry of the Environment's report as a significant source of mercury emissions in the country. Furthermore, there is the problem of soil contamination due to incorrect disposal of products containing the substance, covered by the National Solid Waste Policy (AGROAMBIENTE, 2023).

Most of the substance's atmospheric emissions occur in the form of metallic or elemental mercury (Hg^o). This form of the metal is quite stable, which allows it to be transported over long distances, in addition to remaining in the environment for a long period. As a consequence of contamination, inhaling high concentrations of metallic mercury vapor can cause lung damage. Chronic inhalation causes neurological disorders, memory problems, skin rashes and kidney failure. It is possible to identify poisoning by the elementary substance through a urine test. Elemental mercury binds to other elements, creating two other forms, organic and inorganic mercury compounds (AGROAMBIENTE, 2023):

Methylmercury $[CH_3Hg]^+$ (organic compound)

Methylmercury is just one of the representatives of organic mercury compounds. However, it is considered the most important due to its high toxicity to the human body. It is produced from the elementary substance and synthesized by bacteria present in aquatic environments as a result of the detoxification process. In this process, mercury (Hg) binds to a methyl group (a carbon linked to three hydrogens - CH_3).

Methylmercury is incorporated into the aquatic ecosystem and accumulates in the tissues of organisms. The higher the organism's position in the food chain, the higher the concentration of methylmercury in its body, naturally. Therefore, when consuming fish that occupy the top of the food chain, the individual is possibly eating food contaminated by methylmercury. They are salmon, trout, tuna and others. The routes of exposure are through ingestion of water and consumption of fish contaminated by methylmercury. Ingestion of methyl-Hg causes damage to the central nervous system. But also neural dysfunctions, and in severe cases, leads to paralysis and death.

Inorganic mercury

Inorganic mercury is represented by a set of mineral salts and compounds. These are formed by the bonding of the substance with elements such as sulfur and oxygen. Its main uses occur in the manufacture of batteries, paints and seeds; biocides in the paper industry, antiseptics, chemical reagents, protective paints for ship hulls, pigments and dyes. There is no industrial or commercial use for methylmercury (AGROAMBIENTE, 2023).

MERCURY CONTAMINATION - CONSEQUENCES TO HEALTH AND ECOSYSTEMS:

The main route of exposure is occupational. When workers come into contact with inorganic mercury through inhalation and dermal contact. Another route of exposure to consider is the ingestion of pharmaceutical products and the consumption of contaminated food. Contact with the dermis can cause rashes. Ingestion of a high concentration of the inorganic substance causes irritation and corrosion of the digestive system. Just like elementary mercury, inorganic mercury poisoning can be identified through urine testing (AGROAMBIENTE, 2023).

Water provides a habitat for countless species, and many of these species absorb this mercury. Some of these species are consumed by humans, thus, there is exposure of humans through this route, as mercury is easily absorbed by our body. The biotransformation of inorganic mercury to methyl mercury by bacteria is the process responsible for the high levels of the metal in the environment, as already explained (LACERDA & MALM, 2008).

In general, the pollutants that cause the most damage to ecosystems can be divided into two large groups. The first includes substances present in effluents from large urban areas, especially associated with the improper disposal of solid waste (garbage) and inadequate or non-existent treatment of sanitary sewage (LACERDA & MALM, 2008).

The second group, made up of pollutants of industrial and mining origin, includes toxic substances such as metals, greenhouse gases and organic pollutants, especially those generated by the burning of oil. Unlike contaminants in the first group, whose effect is generally local or, at most, regional, these have the power to affect the environment on a global scale. For example, the emission of greenhouse gases (especially carbon dioxide and methane) and metals (such as mercury and lead) into the atmosphere largely originates from the generation of energy by burning fossil fuels (LACERDA & MALM, 2008).

Although the effects of these contaminants are much less visible, their impacts are much more difficult to remediate. Little is known about the response of natural ecosystems to chronic exposure to this group of contaminants. Several of them, especially metals, are non-degradable, progressively accumulating in natural ecosystems and affecting their functioning over decades or even centuries (LACERDA & MALM, 2008). For example, old mining areas, although deactivated since the penultimate century, such as the tailings of the gold rush of the American West, still affect the local biota today (In PEREIRA, 2022).

In everyday life, mercury is present in various forms and objects, made by industry or caused by it, such as burning fossil fuels, electrolytic production of chlorine-soda, production of acetaldehyde, garbage incinerators, paper pulp, paints, pesticides, fungicides, mercury vapor lamps, batteries, dental products, amalgamation of mercury in gold extraction, among others (MICARONI; BUENO & JARDIM, 2000; LACERDA & MALM, 2008).

Pollution caused by Mercury is a worldwide problem related to inadequate mining practices, causing intense environmental impacts and the health of workers exposed to this metal. Miners use mercury to collect concentrates in the form of an amalgam and recover the metallic gold by burning it, volatilizing the mercury, which is carried by the wind and then precipitates. The lack of technical-operational knowledge of this process and the lack of culture for recycling mercury, combined with the relatively low cost of the liquid metal, causes large quantities of mercury to be released into the soil, water and air (TUTUNJI; DE-PAULA & LAMAS-CORRÊA, 2008).

Mining can also morphologically damage the original form of the soil, eliminate vegetation and siltation of water courses, generating waste containing metallic mercury. Poor land use can also increase mercury methylation levels. Erosion can transport the metal to the water table, contaminating it. So, abiotic factors, such as pH, electrical conductivity, oxygen availability, temperature, as well as biotic factors such as biological activities and nutrient concentrations, among others, are important in the processes of organization of mercury and other heavy metals in soil and water (TUTUNJI; DE-PAULA & LAMAS-CORRÊA, 2008).

One of the widely used and dangerous methods in gold extraction is dredging. This process takes place using pumps measuring 5 to 12 inches in diameter, sucking gravel from a depth of up to 30m. This procedure can be carried out using lances, which are pipes with a system of cutting heads that allow penetration into the hard crusts at the bottom of rivers, or by divers (TUTUNJI; DE-PAULA & LAMAS-CORRÊA, 2008).

Submarine workers, as they are popularly called, spend more than 4 hours submerged holding gravel pulp suckers. Due to low visibility, fatal accidents caused by underwater slopes collapsing are frequent. Also frequent are the cuts of air supply to divers by opponents, who seek the points with the highest concentration of gold in the rivers (TUTUNJI; DE-PAULA & LAMAS-CORRÊA, 2008).

Another very relevant factor in this gold extraction process are the environmental consequences that the methods used, such as dredging, and their high levels of mercury contamination, environmentally affect the course of rivers, and consequently the bodies of people who work directly with This method indirectly affects people who live around rivers, and end up becoming contaminated with: water, soil, crops and fish contaminated and consumed by the population (AGROAMBIENTE, 2023).

Faced with the need to select plant species capable of carrying out phytoremediation of contaminated soils, several groups of plants are being researched and have been obtaining promising responses as phytoremediation agents for petroleum-derived contaminants. The groups include: grasses, legumes, vegetables, arboreal species and several other monocotyledons and eudicots (CUNNINGHAM et al., 1996; MERKL et al., 2004; HYNES et al., 2004; HUANG et al., 2005; ZULFAHMI et al., 2021. All In DE SOUZA, 2021).

Although this phytoremediation technique has limitations such as a long decontamination time, it is economically viable and is an excellent use of natural resources. It requires efforts to go beyond the laboratory scale and reach the fields (AGROAMBIENTE, 2023).

CONCLUSION

This chapter revisited an old experiment that tried to evaluate elicitors for the germination of *Vigna unguiculata* (Cowpea): the plant extract of *E. tirucalli*, the ultra-diluted and dynamized solution of *Sulfur 5CH and the two in combination.* At that time, due to the results, the participation of the sodium/potassium pump, detoxifying enzymes from the thiol group and the suggestive participation of the nitrogenous route of oxidative stress were discussed, with an increase in abiotic factors and cyanuric acid in similar hydroponic solutions where the Cowpea was cultivated, and the potential phytoremediation effect for herbicides in the soil was questioned.

With the literature showing the association of the thiol group with mercury, as a perspective of a mechanism for phytoremediation, allowing the removal of mercury from contaminated soil and water sources, in addition to showing that very few plant species have this ability. Based on the figure and diagram drawn up by environmental engineer Alexandre Pereira, this potential was considered for the succulent species studied, E. tirucalli, with the likely mechanism being re-discussed, considering the participation of the dairy ducts as capable of extracting and even bioaccumulating pollutants, in this case, mercury, since biotransformation was observed in this species from phytovolatilization that caused inhalation poisoning of researchers from an odorless substance.

To conclude, this seems to be a viable mechanism to be investigated in future trials for the cultivation of *Vigna unguiculata* with micropropagated *Euphorbia tirucalli* extratus in water, this time, to be carried out in partnership with agricultural, agronomic (phytosanitary) and environmental engineering, aiming through from research into phytoremediation, the interdisciplinary promotion of environmental health.

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