POLITECNICO DI TORINO
Facoltà di Ingegneria
CdL in Ingegneria per l'Ambiente e il Territorio

FEASIBILITY STUDY ON PHYTOREMEDIATION OF HEAVY METALS FROM COPPER MINING WASTES

RANCAGUA MINES, CHILE

Supervisor:
Prof. Riggio Vincenzo Andrea

Candidate:
Martini Marco

Academic Year 2019/2020

A chi, nei momenti difficili, c'è sempre stato
a chi mi ha dato la forza, quando non ce l'avevo
ed a chi ha camminato sempre al mio fianco.
Ecco, siamo arrivati lontano,
insieme.

Abstract

High concentrations of heavy metals in the soil are an important environmental problem and contamination comes mainly from industrialization, often carried out without proper regulations. The mining sector is one of the major contributors to the release of dangerous substances into the environment. In Chile, this sector accounts for a major proportion of gross domestic product but has significant environmental problems linked to the risk to human health. The accumulation of waste materials, generally fine and rich in HM, takes place in large open-air deposits, called "tailing dams" and located in the vicinity of mines and processing sites. The recovery and treatment of such deposits is necessary given their increasing expansion and the resulting environmental impact.

Phytoremediation is rapidly gaining attention as a biotechnology to clean up substrates that are also contaminated with heavy metals, but the possibility of applying this technique must be assessed on a case-by-case basis. In this study, the feasibility and potential of phytoextraction intervention in tailing dams in Rancagua Copper Mine was investigated. The possible perspectives of the search have been evidenced, identifying the criticalities and fixing the bases for a future experimental planting.

First, the mining sector of copper in Chile was presented, analysing the extraction and transformation process, including the waste arising from it. The focus was on the analysis of tailing dams, deposits of fine material coming from floatation and thickening operations, subsequently reporting the environmental and structural risks related to their implementation and operation. Afterwards, the phytoremediation technique was presented, showing the various strategies used and the possible types of treatment. In the case of the extraction of HMs, the impact of some factors was assessed and the plants suitable for such remediation treatments were classified, highlighting the characteristics necessary for the growth and accumulation of contaminants. In addition, the assessment criteria were presented and a comparison was made with traditional remediation techniques.

For the development of research, the site under study was also characterized, both from the meteorological point of view (studying variables such as temperature, precipitation, humidity, evaporation, solar radiation and wind) and from the chemical point of view. By linking the chemical analyses to the guidelines for the hazardness of the specific elements, a risk-analysis was carried out, identifying the highest degree of contamination for Arsenic, Lead, Cadmium, Chromium and Zinc. Based on the above-mentioned evidence, the species that have shown phytoextraction potential for certain contaminants have been reported. In this study it has been shown that no hyperaccumulative species have been investigated in the Chilean territory. However, few endogenous and exogenous species could be used to clean up the contaminated site. *Pteris Vittata L.*, *Brassica Juncea L.* and *Noccea Caerulescens* showed adaptability and growth in hostile environments and resistance to multicontamination. Such plants could be the right candidates for future experimental planting.

Acknowledgement

This thesis work was carried out in collaboration with professor Vincenzo Andrea Riggio and Politecnico di Torino for the project REMIND (Renewable Energies for Water Treatment and Reuse in Mining Industries), patronized by the European Commission. The health emergency that we have experienced, and are still experiencing, has not allowed us to carry out this work as we planned, denying the possibility of carrying out activities on site and spending the period of experimentation in Chile. Despite all the difficulties I want to thank my supervisor, Professor Vincenzo Andrea Riggio, for supporting me throughout this period and for helping me to give the best in this project. This thesis would have not been the same without him. I would also thank Professor Jacopo Seccatore, from Universidad Adolfo Ibañez of Santiago de Chile, for helping me to organise the period and my stay in Chile. COVID-19 made everything vain but I'm sure I would have felt warmly welcomed from the very beginning.

I would like deeply to thank those who have allowed me to come here, those who have always been at my side and have believed in me. This is not just my goal. I would thank those I have met during these years, those who have even spent a few moments with me, because all have contributed to make me the person I am in this moment. I have had beautiful and exciting moments but also difficult and hard times, and each time it was, for me, an incentive to do better and a challenge to give more in my life. When I looked at my side I was lucky to always find someone, because both the joys and the pains, if shared, have a better taste.

I would thank friends of Grosseto, those who despite the commitments and distances were present; I do not need to write names because each of them is aware of how important it has been and still is for me. I want to thank also those who, despite the studies have gone through different paths, have always been able to be there, as the friends of the University of Pisa, in particular my group Trivella and Davide. A special thanks to my university family of the Politecnico di Torino. From the first day I found some special people, each of you helped me to give the best in this period and even the hardest periods, like the exam sessions, have spent in a better way. A special thanks to my friends from Porto, you have made the experience of Erasmus an indelible memory, have been six intense months, full of emotions, travels and moments that I will never forget.

Contents

List of Figures

List of Tables

Acronyms

AMD	Acid Rock Drainage
BCF	Bio-Concentration Factor
CEO	Chief Executive Officer
ETP	Evotranspiration
GDP	Gross Domestic Product
HMs	Heavy Metals
MMA	Ministry of the Environment
PET	Potential Evapo-Transpiration
PLS	Pregnant Leach Solution
SEIA	Environmental Impact Assessment System
SERNAGEOMIN	National Geology and Mining Service
SX	Solvent Extraction
TF	Translocation Factor
UTM	Universal Transverse Mercator

Chapter 1

Introduction

1.1 Background

Copper is one of the essential metals required for the development of the industrial economy. This metal has been known by mankind for over 10,000 years and has also characterised one of the periods of human history: the Copper Age. The metal gets its name from the Latin *Cyprium aes*, meaning *Cyprus metal*, since the island of Cyprus was where copper was mined during the Ancient Roman world. This evolved into the word *Cuprum*, from which the chemical symbol Cu is derived.

Copper has a typical copper-red colour, as showed in Figure 1.1, and his importance is due to its malleability and excellent electrical and heat conductivity. Other characteristics such as corrosion resistance, durability and antimicrobial properties make this material widely used in various industrial sectors (Garside, 2020). Also, its potential to be recycled as often as desired allows the range of uses to be extended and, in a small part, mining activity to be limited.

Figure 1.1. Copper product, pure cathode (99.99%).

Upon reflection, one would conclude that the innovative progress of the world would not be possible without copper. The growing need for energy, the high demand for electronic equipment and the need for interconnections and telecommunications are just some of the factors for which the demand for copper is constantly increasing (Aurubis, 2020).

The increase in demand for copper from industrial economies such as China and India is one of the main drivers of investment in the copper mining market. Currently, China is the largest consumer of industrial metals and accounts for a major share of global copper consumption. Other developing economies and technological progress will certainly feed future demand. The mining activity carried out in a few countries in the world should satisfy various industrial markets. The main end-user segments of this market are manufacturers of electronic equipment, generally the largest consumers, followed by the construction, infrastructure and transport industries. Production therefore had to follow the demand trend, involving an increase of about 5 million tons only in the last 14 years, as shown by the graph in Figure 1.2.

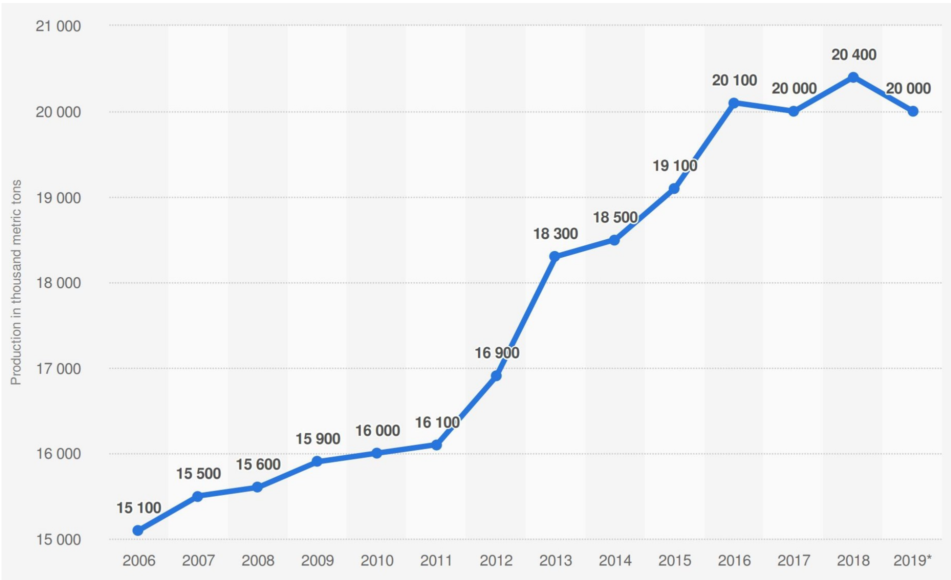

Figure 1.2. Total copper mine production worldwide from 2006 to 2019 (in 1,000 metric tons), from U.S. Geological Survey (2020a).

Globally, the copper mining industry is one of the leading markets in the metal mining market. Most of the available copper is distributed over large areas, generally mixed with mineralised materials and rocks. Currently, as a study of the U.S. Geological Survey (2020b) showed, South America is the main market for the copper mining industry, having the largest reserves of copper in the world. Chile, having reserves of 210 million metric tons, is the main producer of copper in the world and some of the largest copper mines in the world are located in this country.

Although the mining sector has generated and still generates important economic benefits for several countries, including those in South America, it has consequently also produced various social and environmental impacts. Environmental responsibilities are linked to the risks generated by mining and quarrying, often carried out in contexts without the right legislation. The main concerns stem from contamination of groundwater and surface water, loss of biodiversity, soil pollution and the resulting erosion. The mining industry, however, does not only cause damage to the environment but this situation is also affected on the health of the population around the mining sites. The spillage of chemicals, as evidenced by various epidemiological studies, often has harmful effects on the health of the inhabitants in the proximity of the site, mainly workers and families of them. The various attempts to move the villages of the workers to distances greater than 10 kilometers did not give the expected results.

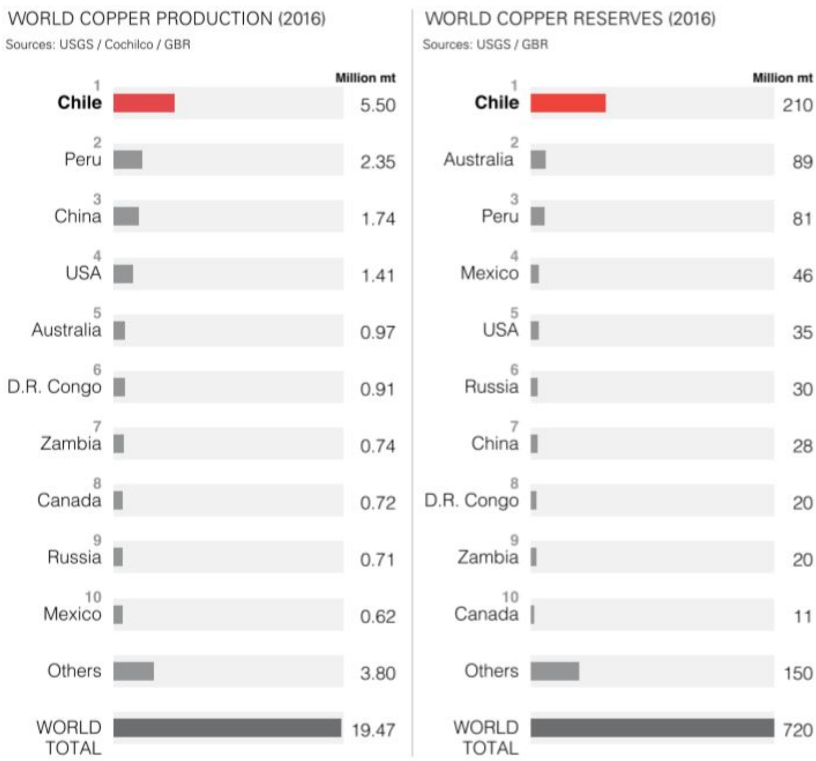

Figure 1.3. World copper production and reserves, in 2016, from FISA and GBR (2018).

The deposits of tailings, both operational and those abandoned for years, generate a series of risks of contamination of soil and water with metals and metalloids. This process is due to atmospheric agents, such as wind and water, which encourage the dispersion and dragging of the material to the surrounding areas, resulting in problems for human health, natural ecosystems and agriculture. The presence of such deposits in arid and desert places, without aquifer and uninhabited, may involve less risk, but mining activities in the vicinity of populated cities need greater attention and immediate action to mitigate such impacts. As previously pointed out, the absence of legislation regulating the mining phases, both operational and post-closure, has attracted high foreign investment capital but has had a negative impact on the environment. Since the beginning of the century environmental awareness has grown and become more important thanks to the foundation of bodies for the protection and control of mining activities. The relevant regulations have introduced important laws for tailings deposits, establishing terms and conditions for the physical and chemical stabilisation of contaminated soils before abandoning them.

Several technologies or remediation strategies have been developed to address the problems associated with increasing metals dispersion on soil. However, the remediation of metal enriched substrates is very difficult because these elements cannot be degraded chemically. Traditional treatments, from an economic point of view, are impracticable in developing countries and leave the substrate in a condition where its physical, chemical and biological properties have been altered, which limits the possibilities of its subsequent use. In this context, the development of alternative solutions strengthening the management of tailings deposits is inevitable, in order to improve the control of their performance and therefore their relationship with the environment, promoting a sector that applies global sustainable practices.

1.2 Chilean framework

1.2.1 Copper sector in Chile

Mining is one of the main industrial activities in Chile and is directly linked to the economic development of the country. The sector, having a high prevalence in the country, has a direct influence on the economy, employment and per capita income of the inhabitants. Historically Chile is a mining nation, having developed since the sixteenth century an industry capable of exploiting its deposits of silver, copper and gold. During the 19th and 20th centuries, through the extraction of saltpetre, a material with chemical properties suitable for use as a fertiliser for agricultural land, the sector experienced exponential growth and generated important structural changes in the Chilean economy. From this period international trade and foreign investment, attracted by the high amount of natural deposits, led to important factors of economic growth by requiring the government to take on an important role in managing the large tax revenues generated.

Subsequently, from the 19th century, the central role of saltpetre was superseded by the extraction of Copper. Previously, the extraction of red ore concerned only a small portion of mining activity, involving only the exploitation of small deposits. This rather sudden change was due to the increase in the world demand for ore for uses in nascent industries such as electricity, for the expansion of the construction sector and for technological innovation. This period of change had also made the exploitation of deposits with copper content between 1% and 2% economically sustainable. The real economic boom of the extraction of copper occurred between the years 1990 and 2000, when the sector succeeded in attracting foreign investments and exploiting the wealth of the subsoil, quantified as about a third of the world reserves of copper, according to data of FISA and GBR (2018). The period between 2003 and 2014 also led to a growth in the sector thanks to the boom in prices and the great demand for metal from China. The high cost of copper led to an unprecedented development of the sector, reaching its peak around 2011, allowing companies a high increase in production. The country's economy suffered strongly from this positive cycle and the country's GDP risen from \$77.8 billion to over \$258 billion.

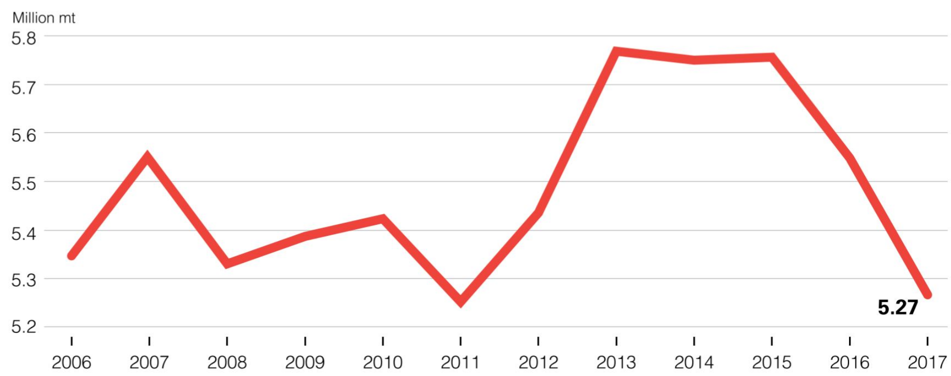

Figure 1.4. Evolution of copper production in Chile between 2006 and 2017, from FISA and GBR (2018).

Some setbacks, such as the difficult period between 2013 and 2016, were due not only to the fall in prices but also to the progressive introduction of regulatory policies concerning social and environmental impact. Nelson Pizarro, CEO of CODELCO, the largest mining company, defined the latter period as the "perfect storm" as it allowed a cost reduction in favour of the increase in productivity and the development of cutting-edge technologies in terms of innovation, efficiency and safety, eliminating the bottlenecks of the process and encouraging a recovery of the sector. The crises given companies the challenge of maintaining production capacity by adapting, modifying and improving production processes and related energy needs. Investments in the mining sector and the country's GDP are strongly linked to price fluctuations, as Chile has no control over the price nor the grade of the mineral in the rock but always acts and influences the productivity of the sector and the costs attached to it. The production experienced various fluctuations from 2006 to 2017, as can be seen from the graph in Figure 1.4, due to the factors previously explained, and in 2017 it was around 5.27 million meters.

To date, mining accounts for over 11% of GDP, compared to 8% in 1997, and has a 50% share in the country's exports. The continuous growth of exploration allows new deposits to be found while, as pointed out by a report published in 2018 (FISA and GBR (2018)), the number of mining concessions (group owned) in 2016 was 86 076 for a total of around 15 milion hectares. With regard to copper, SERNAGEOMIN reported in 2015 that the sector also had a majority share in the country's exports, reaching as much as 89% of the total mineral exports. The sector has an impact on the following economic and social factors:

- *GDP*: copper extraction alone has accounted for on average 10% of Chile's gross domestic product (GDP) over the past two decades, while, by comparison, the mining industry as a whole accounts for 10.9% of the GDP;
- *Employment*: The industry generated more than 380,000 jobs in 2016 considering both direct, indirect and adept workers for contractors. In addition, per capita income is higher than the national average in regions with the highest mining activity;
- *Local communities*: the poverty rate is one of the largest indicators of development in the country: it fell from 40.5% in 1990 to 8.5% in 2015;
- *Tax revenue for the country*: since 1990 the sector generates on average 7.8% of tax revenue, although it is not constant but fluctuates during the period
- *Related industries*: industries such as the energy sector are directly related to mining, enough to think that in the northern regions electricity utilization has a share of 38% while water consumption represents 3% of national amount.

However, although the increase in mining activity has been crucial to Chile's progress, it has had negative effects on the environment mainly due to the lack of proper regulation. Not only this, but also the decline in mineral grades in the rock, have led to an increase in processing waste and consequently a high degradation of the soil. This is enough to think that Chile is the third country in the world with the largest amount of tailings, with a total of 742 in 2019, of which 104 correspond to active deposits, 463 to inactive, 173 abandoned and 2 under construction, as report by Equipo Programa Tranque, Fundación Chile. The current deposits have an approved capacity of about 21 billion tons but currently the deposited tailings would amount to about 9.6 billion. Future estimates are really tragic, according to reports from the Ministry of Mines, in 2026 over 915 million tons of tailings will be produced per year, resulting in an increase of 74% in the generation of tailings compared to 2014 (525 million tonnes per year). The recent laws on the subject, set out in the next chapter, together with the national plan for tailings deposits, have enabled the creation of greater environmental awareness and the drafting of guidelines and tools to address these problems and create a more environmentally sound and secure industry.

1.2.2 Legislation framework

Although mining on the Chilean national territory has a long history, the regulations governing the sector have focused mainly on the organizational and regulatory aspects for mining concessions, mining and mining code regulations. These aspects, which have dealt with by the Ministry of Mines since 1960, have left no room for environmental and regulatory considerations. Only since 1994 with the "General Law on Environmental Bases" (Law no. 19 300) have environmental terms and impact assessment processes started to be established, subsequently updated by the Law No. 20 417 of 2010. Previously, only since 1992 could projects be voluntarily submitted to an environmental assessment process implemented by the Ministry of Mines while the previous regulatory vacuum allowed the degradation of the environment and the abandonment of places of copper production and dump.

Table 1.1. Regulatory framework for mining activity, modified from Ministerio de Minería (2019); Toledo (2016).

	Laws	Framework regulation	Year
Mining activity	Act No. 18 097	Constitutional law mining concessions	1982
	Act No. 18 248	Mining code	1983
	Decree No. 1	Mining code regulations	1987
Sustainability mining issue	Act No. 19 300	Environmental Foundation Act	1994
	Decree No. 40	SEIA Regulation	2002
	Decree No. 132	Mining Safety Regulation	2004
	Decree No. 248	Tailing dam regulation	2007
	Act No. 20 417	Update of Act No. 19 300	2010
	Act No. 20 551	Law on the closure of mining works	2011
	Decree No. 41	Regulations closing works and installations	2012
	Act No. 20 819	Update of Act No. 20 551	2015

In order to further control sources of pollution from tailings depots, SERNAGEOMIN Tailings Depot Office has, since 2015, compiled and updated all information on tailings depots in the country, creating the National Security Register. Within these reports are reported the characteristic parameters of each repository, such as tonnage, location, owner, method of construction, type of plant, geographical location (UTM), status (active, inactive, abandoned, under construction) and authorised and current volumes. As a decentralised body with legal personality and its own assets, the purpose of SERNAGEOMIN is to assist the State, through the Ministry of Mines in terms of control and training in mining safety, technical assistance and publications based on geology and mining activities, so as to contribute to the sustainable development of the country (Ministerio de Minería, 2019).

The laws and regulations governing mining activity in environmental terms are shown below. In addition, the activities carried out by the Ministry of Mines and the public body responsible for the review and approval of technical aspects (SERNAGEOMIN) are reported, including technical guides and relevant documents.

National laws regarding mining sustainability issue

- *1994: Act No. 19 300 "Environmental Foundation Act"*
  Created on 1 March 1994, it aims to introduce an environmental impact assessment system (SEIA), a management tool for the presentation of both investment projects and projects related to productive activities. The purpose of this system is to determine the effects that these activities will have on the environment, so as to prevent or avoid ecological damage.

- *2002: SEIA Regulation - Supreme Decree No. 40*
  Repair or recovery of polluted soils, which together cover an area of 10 000 square metres or more.

- *2004: Mining Safety Regulation - Supreme Decree No. 132*
  The aim is to protect the life and physical integrity of people working in the mining industry by requiring, for the first time, mining companies to apply a closure plan at the end of the production phase, including the closure of sterile deposits.

- *2007: Tailing dam regulation - Supreme Decree No. 248*
  It introduces new rules and regulations for the approval procedure for mine tailings projects regarding design, construction, management and closure.

- *2010: Act No. 20 417 (Update of Act No. 19 300)*
  This law underlines the fact that the Chilean Ministry of Environment (MMA) has a duty to "propose policies and formulate rules, plans and programs on contaminated waste and soil, as well as risk assessment of chemicals, genetically modified organisms and other substances that may affect the environment".

- *2011: Act No. 20 551 "Law on the closure of mining works"*
  This is the first law that regulates the closure phase of mining facilities, considering this phase as part of the useful life cycle of any mining project. The objectives of the planning remains those of safeguarding the environment and of mitigationing the negative effects via the obligation to carry out a risk analysis on the behaviour of long-term structures, assuring the chemical and physical stability of the dam. Companies must safeguard the life, health and safety of people and the environment by meeting certain standards.

- *2012: Regulations of the law on the closing of works and installations - Supreme Decree No. 41*
  This Decree implements the Act No. 20 551 and fix the regulations.

- *2015: Act No. 20 819 (amendment of Act No. 20 551)*
  This law introduces other legal changes in order to improve the way of calculating the useful life of mining projects, determining the constitution of a financial guarantee for projects and adding changes to the evaluation procedure.

Public sector initiatives - Ministry of Mines and SERNAGEOMIN

- *2002: Framework agreement on clean production for large mines*
  Management of industrial waste and good practices in the solid mining sector; Management of liquid industrial waste and good mining practices; Methodological guide on acid drainage in the mining industry; Methodological guide for closing mining operations; Efficient energy use in the extractive industry and good practices, Efficient water use in the extractive industry and good practices.

- *2003: Guide to Good Environmental Practice for the Small Mining Industry*
  Closure of mining sites, construction and management of dams, management of fuels and lubricants, other.

- *2006: Clean Production Agreement for Small Mines*
  Risk assessment manual for abandoned or paralysed mining operations (FMA/P)

- *2007: Land registry abandoned and paralyzed mining operations: Preliminary risk analysis*

- *2010: Update of abandoned and paralyzed mining sites of the Land Registry: preliminary risk analysis*

- *2015: Technical guides Risk assessment by closure*
  Chemical stability by closure; Manual of good practice for chemical stability, Inventory of chemical stability technologies.

The laws listed and briefly introduced are part of a path undertaken in 1994 to change the pre-existing mining practices (exploitation, extraction and mainly deposit) in order to achieve a greater sustainability of the most important Chilean industrial sector, Mining. However, this does not solve the existing environmental problems relating to the abandonment of mining facilities prior to these rules coming into effect, but limits its scope only to the resolution of environmental issues of new mining projects. It is difficult to clean up derelict and uncontrolled deposits, since these rules do not have a retroactive effect but act by mitigating the future harmful effects of mining, preventing the risks due to possible contamination of the environment. For this reason it will be necessary to introduce a law on mining environmental responsibilities generated in past years (Toledo, 2016).

1.3 Copper processing

In this chapter, the copper production process is analyzed. Starting from the extraction of the material to the formation of the cathodes, this process leads to the production of a pure material which is 99.99% copper. The extraction process and transformation process will be explained next.

1.3.1 Extraction process

The mining operation are necessary to extract the mineral from ore deposits. Copper can be extracted from its ore by:

- *Open pit:* this solution is used in 90% of ore extraction and allows to mine massive deposits. In this method the ores near the surface can be quarried after removal of the surface layers (overlying waste).

- *Underground methods:* it is used when the open pit solution is not suitable and is economically viable at a great depth. It consists in sinking a vertical shaft into the Earth to an appropriate depth and driving horizontal tunnels into the ore.

In order to compare the two different methods, it is important take into account that open pit mining has the advantage of having lower development/maintenance costs and some safety advantages compared with underground methods but the environmental cost can be really high. The choice of extraction method depends on different parameters, mainly physical, technological and economical.

Metals are often found as compounds in ores. An ore is a rock or mineral that has enough metal in it to make it worth extracting. The main ores of copper are sulfides and oxides. From their chemical formulas you can calculate the percentage of copper in each mineral. However, this is not the same as the percentage of copper in the ore, as that also contains unwanted silicates and other minerals, called gangue, which will have to be separated (Copper Alliance, 2020).

(a) *Open Pit mine.* **(b)** *Underground mine.*

Figure 1.5. Some examples of extraction mines.

1.3.2 Transformation process

The manufacturing process of copper, besides being very complex, involves many steps that allow the transformation of the ore from the raw material, obtained in the mining operation, to a purified material. The raw material contains generally between 0.5 and 2.0% copper whereas final material, represent by copper cathodes, must reach a high purity, around 99.99% copper. In order to obtain high quality copper, necessary for the production of electrical equipment, two different methods can be used: pyrometallurgical extraction or hydrometallurgical extraction (Ayres et al., 2002).

Pyrometallurgical is an extraction metallurgical method that consist in a specific high-temperature treatment that allows copper to be extracted from its ores and concentrates (U.S. Congress, 1988). This process is used with copper sulfides and it has generally four different steps (roasting, smelting, converting and fire refining) carried out obviously after the crushing and concentration stages.

Hydrometallurgical extraction is a method based on chemical reactions made in an aqueous medium that allow copper recovery. This methods is used with low grade copper oxide and some sulfide mine wastes and has been practiced for copper recovery for more than 300 years (Szymanowski, 1996). Nowadays, the main smelting method is pyrometallurgy, with around 85% of output in the world. In this chapter both processes are mentioned.

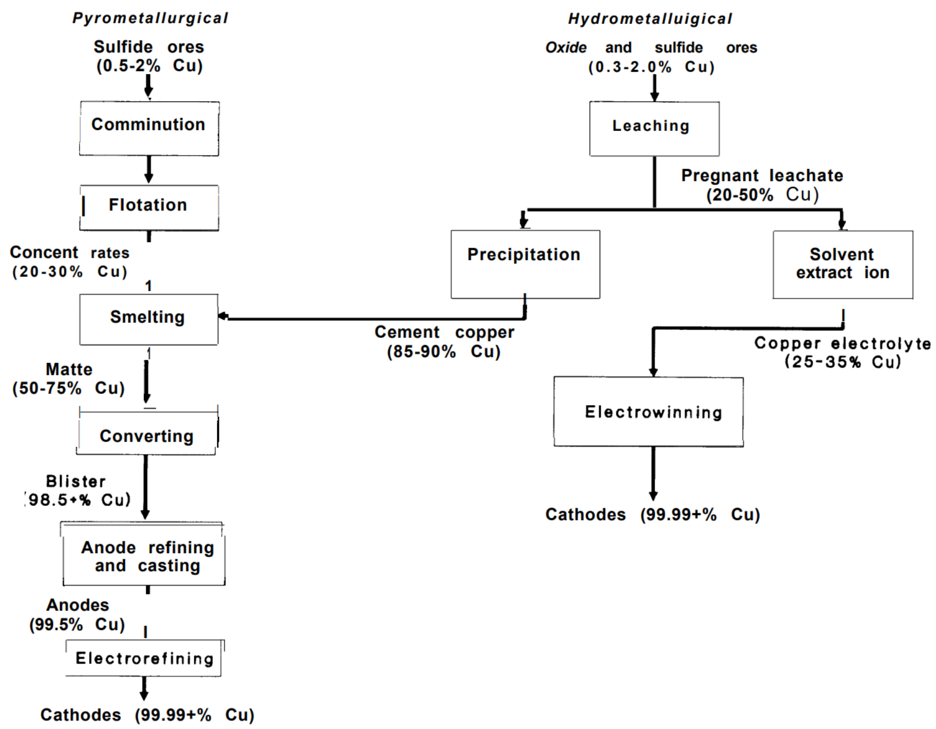

Figure 1.6. Copper transformation processes: Pyrometallurgical and Hydrometallurgical steps, from U.S. Congress (1988).

Pyrometallurgical method

1. **Stage 1: Crushing and grinding**
   The comminution of the ore chunks—essentially from boulders to grains of sand represents the first stage after the extraction operation.

   Crushing is done in three different steps (primary, secondary and tertiary crushing) that gradually reduce the particles to 25 mm diameter and prepare the material for finer reductions in the grinding phase. The Grinding operation is carried out by jaw machines, gyratory, and cone crushers, which use compression force in order to fracture rocks. The first machine is generally used for the first stage (Figure 1.7) while the other machines are used for the second and tertiary crushing stages.

   Grinding also takes place during the same stage. This process operation can be operated wet or dry but generally the logical choice, if the following stage is flotation, is to use the wet type. In this type of grinding work dust control is not required and power consumption is lower. The Water is mixed with the material that comes from crushing in order to create a slurry of around 40 percent solids (U.S. Congress, 1988). Grinding is operated by tumbling the ore with steel rods or balls, or particles of the ore itself.

Figure 1.7. Primary comminution in El Teniente mine, from Gleeson (2020).

2. **Stage 2: Froth flotation**
   This stage (Figure 1.8) is the prevalent concentration method in the copper industry that selectively separates valuable minerals, the hydrophobic part, from waste gangue, the hydrophilic one (Leonida, 2020). To start, the slurry coming from previous grinding operations is mixed with chemicals (surfactant or collector chemical) that make the desired mineral water repellent (hydrophobic).

   After air intake into the tank and agitation, the copper minerals attach to the air bubbles and flow to the surface. The material that overflows the tank is concentrated and has around 20-30% of Copper by mass. The unwanted waste rock (gangue) falls to the bottom and is removed.

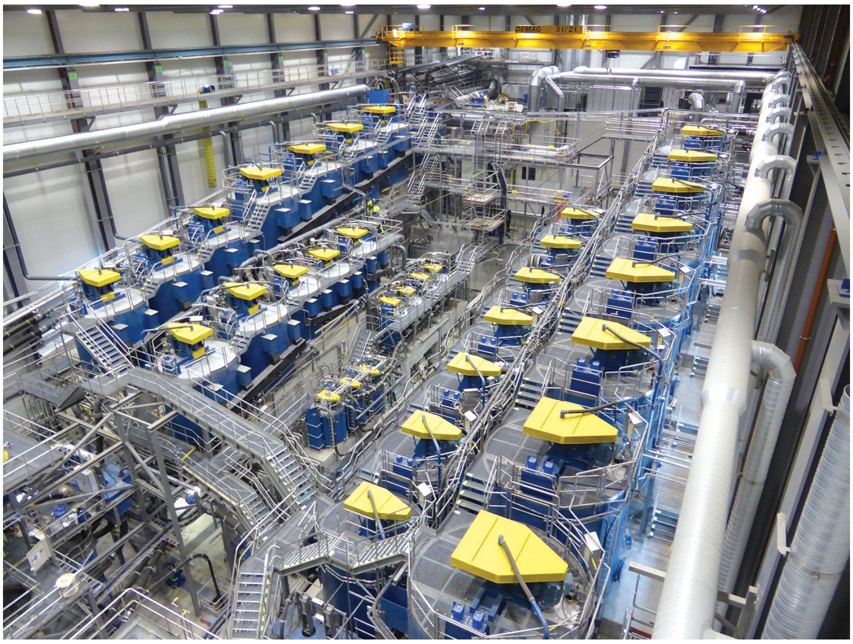

Figure 1.8. Series of tanks used in copper Froth Flotation plant, from Leonida (2020).

3. **Stage 3: Roasting**
   In order to prepare the material for the following smelting it is necessary to remove sulfur and some volatile contaminants (e.g. Arsenic).

   The roasting operation is performed by a chemical reaction that is undertaken at a lower temperature compared of the melting points of the sulphides and oxides involved (in general for copper the temperature is below 900-1000°C). The product from the roaster is called *calcine* that is a solid mixture of oxides, sulfides and sulfates (Copper Alliance, 2020).

4. **Stage 4: Smelting and Anode casting**
   The calcines that come from the previous operation are processed a high temperature, at around 1200° C, and react. The products of this reaction are a copper-rich liquid, called matte, and a mixture of impurities called slag that floats on the surface ready to be removed. During this reaction sulphur dioxide (SO_2) is also produced and needs to be captured to avoid any dispersion and pollution. The material is poured into a mould and it cools and solidifies.

   The blister copper has 99% copper which may seem enough but it contains high levels of sulfur, oxygen, and other impurities. During this stage the blister copper is re-heated and subjected to fire refining and anodes are created by pouring the liquid into a mould.

Figure 1.9. Liquid material poured into rotating moulds, from Encycl. B. (2020).

5. **Stage 5: Electrolytic refining**

   This process is performed at the end of copper transformation and takes place using electrolysis. The goal is to remove impurities from the copper anodes and also recover or reuse them in other process.

   During this operation, the anodes (Figure 1.10) that come from fire refining are immersed in an aqueous solution containing copper sulphate and sulphuric acid. The anodes behave as positive terminals and, during electrolysis reaction, the copper corrodes, moving to the cathodes. At the end of the process the cathodes reach high purity with 99.99% copper and the impurities are recovered from where they are deposited, at the bottom of the tank.

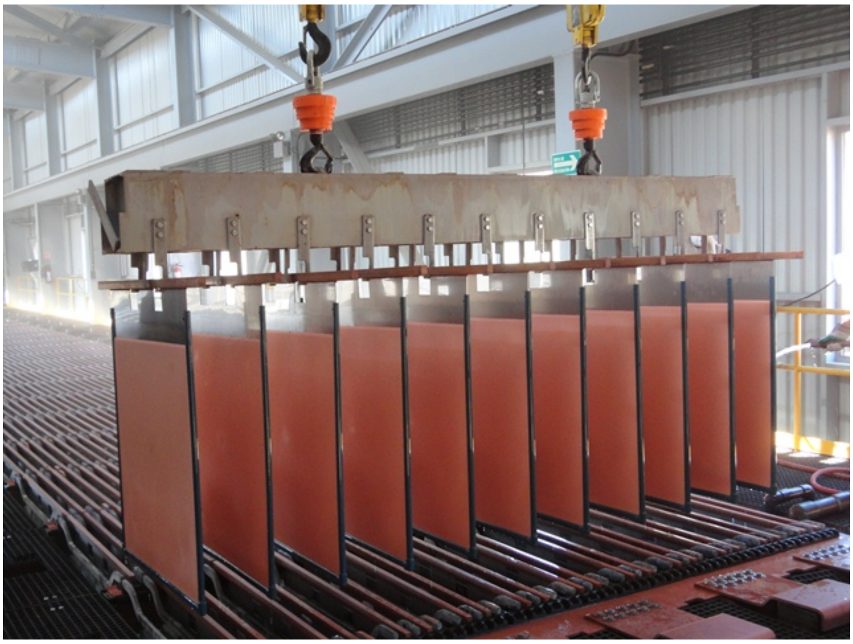

Figure 1.10. Copper electrorefining tankhouse, from Copper Alliance (2020).

Hydrometallurgical method

1. **Stage 1: Leaching**
   Leaching involves the use of aqueous solutions to extract the metal or its compound from the solid material that contains it. There are two main leaching methods: heap leaching and in-situ leaching. In both methods, the material that come from the comminution process is first agglomerated with sulfuric acid and seawater. A special mobile belt transfers the material to a big stock pile where the leaching process takes place. The liquid that "leaches" through the rocks, known as a pregnant leach solution (PLS), is collected because it contains the dissolved copper. The PLS is then captured in production wells, collected and later processed (U.S. Environmental Protection Agency, 1994).

   The leaching solution should be regulated in terms of pH, redox potential and temperature to optimize the aqueous phase dissolution of the desired metal component.

Figure 1.11. Heap leaching system, from Agua royal spring (2020).

2. **Stage 2: Solvent extract ion**
   This methodology of copper extraction enables the recovery of the mineral concentrate from the leach solution (PLS), otherwise it would not be possible to apply electrorefining methods because the mixture is too dilute. At the beginning of this stage, the PLS is mixed with an active organic solvent. This special additive is able to selectively extract the ore, in this case copper, while the impurities remain in the aqueous solution.

   The copper electrolyte, after solvent extract ion stage, has around 25-25% copper and is ready to be sent to Electrowinning. The solvent extraction method is dependent on the solubility of the reagents and the equilibrium constants that control the reactions.

3. **Stage 3: Electrowinning**
   This method is used to recover copper from the electrolytic solution produced in the previous SX step. Electrowinning uses inert (insoluble) anodes made of lead or stainless steel, called sheets. To achieve the development of this

process it is necessary to pass the electrolyte inside the circulation tanks in which the electrolytic solution of supply is constantly pumped. Copper is plated on stainless steel cathodes or thin copper starting sheets. These copper plates are then removed and have a purity of 99.99%.

Figure 1.12. Storage plant of copper cathodes with minimum 99.99% of copper content, ready for worldwide shipping. Photo retrieved from www.reinhausen.com.

1.4 Mine wastes

The study and analysis of the copper transformation mining process is an important step in understanding the derivation of mine waste and then to find specific solutions. In the past, little attention was given to the analysis of the waste resulting from the process and to the environmental problems it creates.

Mine wastes are problematic because they contain hazardous substances, like heavy metals, metalloids, radioactive elements, acids and process chemicals, that can be (or are) released into the environment around the mine. They require special treatment, secure disposal, and monitoring. The increasing attention regarding environmental and dispersion of pollution has focus on their reduction and many studies was developed in this field.

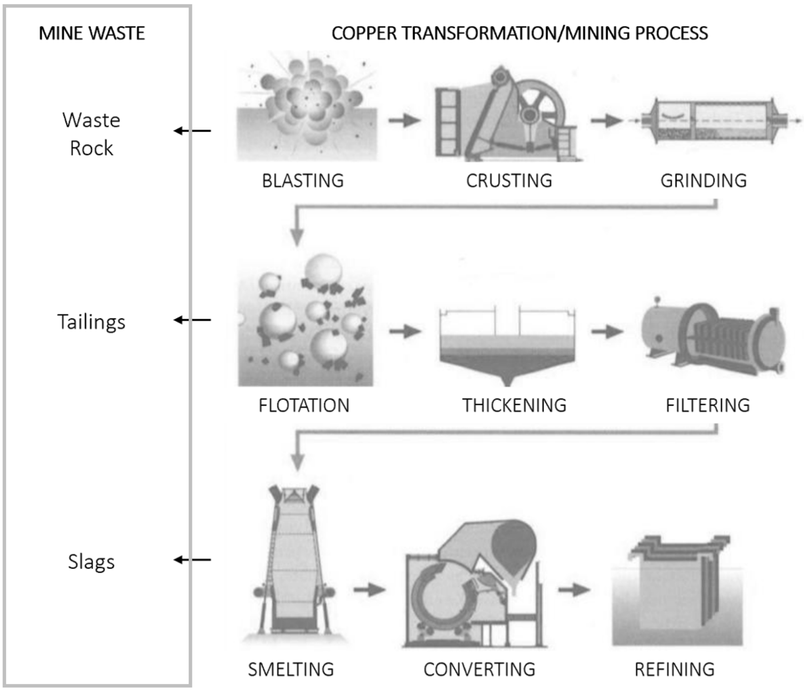

Figure 1.13. Wastes from the copper mining and transormation process with identification of where they are released, from Spitz and Trudinger (2009).

The wastes are not only produced during mining operation but, as we can see from Figure 1.13, tailings come from flotation, thickening and filtering operations and slags come from smelting furnace.

Waste Rock Waste rock, also called sterile material, consists of unmineralised rocks and rocks containing mineralization that are below the "cut-off" grade and will not be further processed because it is not economically viable with the existing technologies. The amount of waste generated by mining operations depends to a large extent on the shape of the ore field, the mining plan and the total mineral production. Rock landfills contain a wide variety of different rocks and minerals that is site-specific, depending on the nature of the ore deposit and the host rock. The main potential environmental problem is related to the presence of sulfide minerals, especially pyritic ores, that can potentially create acid rock drainage (AMD) (Wirtez, 1999). However, water, like rainfall and snowfall, and the presence of oxygen can increase or decrease the risk and the analysis of potential impact must include these two factors. Rock dumps are generally not sealed at their base and the risk of acid water entering the surface drainage system or underground aquifers is very high. The high porosity of rock dumps is also a characteristic that can greatly increase the risk of acid drainage production.

Tailing waste Generally, tailings are materials that remain after the flotation operation and they account for around 50% of total wastes. This is composed of both solids and liquids, and contains very fine host rock and other non valuable material, like nonmetallic minerals. Solids, typically in the field of fine sand and silt, are discharged (with a solid content of 20% to 40%) with process water exhausted in tailings dams. The chemical composition of tailings is highly site-specific, depending on the ore and gangue minerals and type of process used (U.S. Environmental Protection Agency, 1994). The solids in the tailings consist mostly of common rock minerals but they also contain a significant fraction of pyrites and 10% to 15% of heavy metals, including copper (Ayres et al., 2002). Liquid could be removed from the tailings slurry in thickeners but the functioning of this operation depend by site, availability of water or special needed of water reuse.

Slag Slag does not fall under the scope of the study. Slags represents the most important pyrometallurgical waste and it is composed of iron oxides and silicates in a glassy structure. This type of structure enables slag to be considered as very stable waste which does not require special disposal. However, sometimes this waste can contain impurities, like arsenic, and requires a more careful disposal procedures (Wirtez, 1999). Generally, this material can be found stored either near the mine, if smelting was conducted nearby, or often stacked in heaps near the smelter.

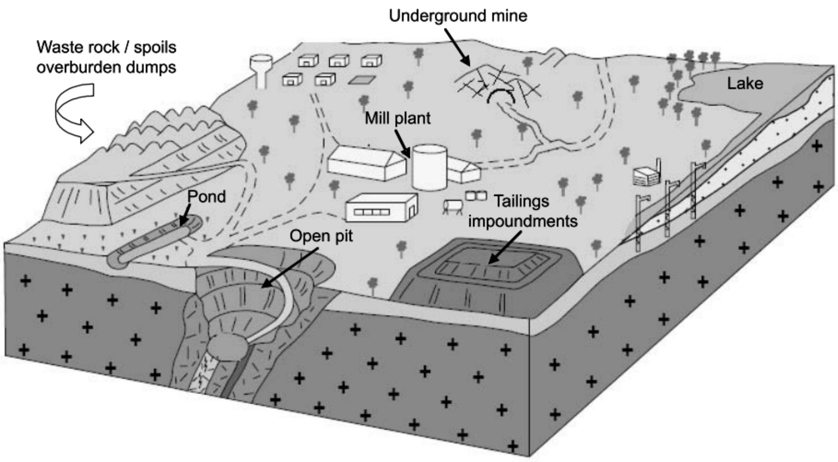

Figure 1.14. Schematic diagram illustrating the various components of a mining operation, from Yilmaz (2011).

1.5 Tailings dam

The tailings, as previously explained, are a mixture of finely ground rock and process fluids that remain after the extraction of the mineral. They are transported through piping systems and are stored in landfills: large tanks built specifically for the purpose of containment. The definition of "Deposit of Tailings" is provided the Supreme Decree No. 248 (dated 2007) and is:

> "any work securely structured to contain tailings from a plant for a wet concentration of mineral species. In addition, it contemplates his annexed works. Its main function is to serve as a deposit, generally, solid materials from the tailings transported from the plant, thus allowing the recovery, to a large extent, of the water that transports such solids"

To get an idea of the amount of material which results it is sufficient to observe how the average degree of copper ore has reduced considerably over the years, as demonstrated by the study done by COCHILCO (2017) and shown in Figure 1.15, reaching a value of 0.65%. The study also stresses that this reduction in Chile is more marked than the world average and is mainly due to the high over-exploitation of ore deposits with higher mineral concentration and the search for new deposits with lower degree of copper mineral. This implies that the tailings represent more than 99% of the extracted material. In addition possible water transport and other elements used in the processes must be considered. Such materials are obviously not economically further treatable and are composed of solid material suspended in a liquid solution.

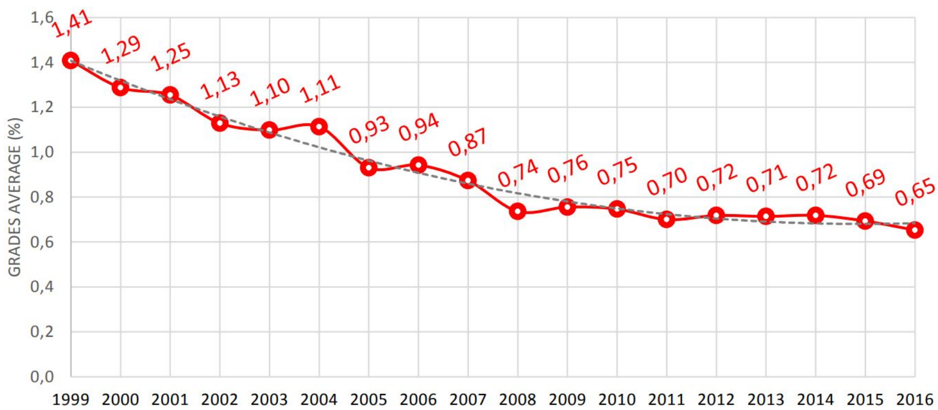

Figure 1.15. Average Copper Mining Grades in Chile, 1999-2016, retrieved from COCHILCO (2017).

The waste material in output can be deposited in different physical forms in relation to the characteristics of the production plant and possibly the need to recover the process water through special filtration treatments. Tailings may be presented as:

- *Thickened tailings:* this type of sediment is previously subjected to a sedimentation process with thickening, removing much of the water. These tailings, however, still contain a fraction of water so they must be deposited in appropriate dam that allows the recovery of the same and avoids dispersion, otherwise pre-filtration treatments need to be carried out, partial or total;

- *Filtered sterile:* in some cases filtration treatments can be carried out to reach a moisture level of less than 20%. Obviously, given the investments in such plants and the associated operating costs, the operation may not be economically viable in such places where the water supply is scarce and expensive;
- *Pastes:* this situation is intermediate between the two previously exposed. This material is similar to a high density pulp, with a water content of between 10 and 25% and a concentration of fine particles by weight (less than 20 microns) greater than 15%.

Tailings must be deposited in safe places in such a way as to ensure stability over time, avoiding water infiltration and consequent contamination of groundwater and soil. The various forms of deposition depend on factors such as the type of tailings, the distance from the processing plant, the storage capacity and the topography of the soil. The "Regulation for the approval of projects for the design, construction, management and closing of tailings deposits" of Supreme Decree No. 248 considers these different types of deposits:

- according to construction mode:

  Tailings Dams (upstream, downstream and center-line methods);

  Tailings Reservoirs.

- according to the type of tailings to be deposited:

  Deposit of thickened tailings;

  Deposit of filtered tailings;

  Deposit of sterile pasta.

Tailings Dams: This type of containment works is characterized by having the supporting wall composed of the thickest fraction of the tailings, the sands. They can be realized through three different construction techniques.

- The *"upstream" method* is characterized by being made of a small dam of compacted loan material to which is added, upstream, the fraction of coarser sand (tailings) while the finer part is placed further away from the wall. This separation is achieved by hydrocyclones and the method has advantages due to the low amount of material needed for the construction. Despite this, however, the wall presents problems of seismic stability, which is why its implementation is prohibited in Chile.
- The *"downstream" method* is always realized starting from a wall of compacted material, but in this case the cyclones positioned in the upper edge of the dam deposit the sand downstream in slightly inclined or horizontal layers. This technique allows much more resistant dams to be realized, although they are more costly due to a higher demand for sand.
- The *method of the central-line or "mixed" method* start from the same point as the others but develops vertically due to the deposit of the sand upstream and fine particles downstream. This method is intermediate between the first two and has a vertical inclination of the upstream wall, while downstream varies from case to case.

In Figure 1.16 are showed three different methods.

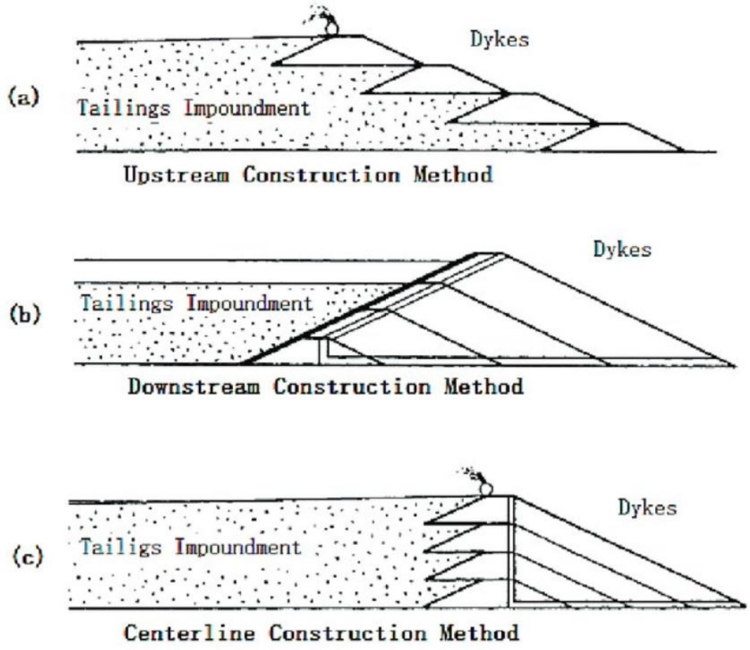

Figure 1.16. Three different construction methods of Tailings Dams, from Bhanbhro (2014).

Tailings reservoir: these types of deposits are made through the construction of the retaining wall entirely with loam material that is compacted and waterproofed along the internal slope. The tailings are deposited in the basin and a mechanism is put in place to recover the water from it. Unlike normal dams, realized considering a determined value of maximum containment, the reservoirs can often be realized gradually, widening the wall and therefore distributing the investments in time as the deposit increases. This type of structure is more solid and resistant to seismic events.

Deposit of thickened tailings: These types of deposits do not require the construction of a retaining wall and the deposit takes place in the form of a cone with slope in relation to the concentration of solids, studied through the viscosity curve. Despite this, however, a small wall is required to delimit the area downstream and collect the water that flows. The maximum slope of this type of deposits is 6% so the realization is possible in certain places where the topography allows it.

Deposit of filtered tailings: The passage of the material through mechanical filtration operations allows the deposit of tailings with a low presence of water, with humidity of less than 20%. Obviously, the tailings cannot be transported through pipes but must be carried out through conveyor belts or with earth moving equipment. These types of deposits, called "dry stacks", are stable and can be suitable for areas with high seismic activity. Although the contamination of subterranean water by infiltration is very small, an environmental problem can result either from the wind with the generation of dust or in very rainy climates for the operation of the site. In Figure 1.17 a "dry stack" filtered sterile deposit is shown.

Figure 1.17. Dry deposition of tailings with mechanical methods.

Deposit of tailings in paste: If tailings are dehydrated, they are presented in the form of paste, that is, a high density material with a low water content. The paste is transported through a system of pipes and volumetric pumps since the high density avoids the creation of sedimentation and means it can be transported efficiently. The deposition of the same occurs in layers that gradually dry out and form cracks that will be filled by the next layer. This allows stable structures to be created and the low water content reduces the risks associated with the infiltration of heavy metals underground. In Figure 1.18 it can be seen how a deposit of tailings in paste is formed.

Figure 1.18. Deposit of paste material.

1.5.1 Identification and environmental problem

Starting with the definition of risk as "the combination of the probability of occurrence of an event and the extent of its consequences", tailings deposits, in all forms and of all ages, may present different liabilities and environmental risks. They should be analysed and taken into account at each stage, design, operational and closure, in order to limit their impact and the associated consequences. The main risks are related to:

- Structural risks associated with physical stability with regard to breakdowns in the dam wall due to seismic or hydrogeological events;
- Environmental risks associated with soil contamination as a result of infiltration into the underlying layers or as a result of wind action, allowing the migration of contaminated (such as HMs) into other environmental matrices. These risks are also linked to the disfigurement of the landscape and the deprivation of such land for agricultural or other purposes.

As stated in Law No. 20 551, regulating mining operations and works, it is important to specify two concepts:

> **Physical stability:** Structural security situation, which improves resistance and reduces destabilizing forces that may affect the works or deposits of a mining site, for which measures are used to avoid bankruptcy phenomena, collapse or removal.
>
> **Chemical stability:** The control of the presence of the chemical characteristics of the materials contained in the works or deposits of a mining site, in the water, air and soil, with the aim of preventing or eliminating chemical reactions. These reactions can cause acidity and avoid contact of water with acid-generating residues found in large mining works and deposits, such as tailings deposits, landfills, waste deposits and leaching rubble.

Of course it is important to analyse in detail all the possible risks, both of a structural nature (physical stability) and of an environmental and chemical nature, of the structures involved.

Seismic risk As Chile is a country characterized by seismic events, it is necessary to consider the resulting structural importance and determine the potential risks. The design of the walls supporting the dams must be such as to guarantee, through the constructive modalities and the dimensions, a stability in time and a durability of the work in relation to the magnitude of the seismic event, related to the place where the deposit is located. Events such as soil liquefaction can lead to deposition failure by releasing tailings and compromising the safety of adjacent areas.

Hydrological risk Direct or indirect contamination of water has always been one of the major risks associated with tailings deposits. Rainfall, flooding and river overflow can lead to the dissolution and migration of heavy metals to the groundwater, and thus to even the most underground strata. This transfer of heavy metals causes acidification of water and an alteration of the subsoil leading also to instability of structures.

Risk of generation of dust The tailings, being fine waste material of the processing, are affected by the action of the wind and generate dust. The particles are carried by the wind and have adverse effects on the health of living beings by interacting with the respiratory system. Not only human beings but also plants can be affected by the harmful effect of dust, which has a negative impact on the photosynthesis process and on agricultural land.

Soil pollution Soil and water are the environmental matrices most affected by the harmful effects of contamination. The metals contained in the tailings can migrate, contaminating productive soils and compromising their use. Toxic elements can come into contact with plant species through the roots, causing even fatal risks to plants or being accumulated in leaves and causing harmful effects to animals and living beings who use them for food.

Water consumption The aspect of water consumption has always been the focus of discussion regarding mining activity. Tailings are often deposited in the basins along with process water and other chemical elements which subsequently evaporate or are retained in the form of moisture. This results in a significant loss of water especially if this situation occurs in areas where it is scarce. To limit this problem, water recovery methods can be used using filters or through special piping systems.

It is also important to mention that such deposits have a high visual impact on the environment by increasing waste from neighbouring communities and raising awareness of the risks arising from the storage of such materials.

All these risks of contamination are, of course, ultimately a health hazard. Particles released into water, soil and air can potentially come into contact with the human organism and manifest a toxic action given the strong affinity between their cationic forms (due to the sulfide present) and sulphide groups present in enzymes. The effects depend on the type of metal and the chemical form through which they enter the body. For example, copper, although it as an essential micronutrient, may cause damage to organisms due to excessive concentrations while lead mainly affects the nervous system and adversely affects activities such as the production of blood cells and affects organs such as the kidneys and the reproductive system. Other substances such as mercury and arsenic cause damage to the human organism in relation to the severity of the contamination (López et al., 2003).

1.6 Goals of the study

The goal of this study is to evaluate the feasibility of using plants for phytoextraction of tailings resulting from the extraction of copper at a Chilean mine. The increasing consumption of copper and other metals has resulted in an increase in mining and land use. It is therefore necessary to study new forms of mitigation of the negative effects produced by the mining industry, considering each extractive and transformation phase in order to obtain the final product. Particular attention must be paid to the large quantity of sterile waste produced by the activity, thus going to study new forms of treatment and reclamation of the contaminated material.

The introduction of phytoremediation as a method of remediation of sites contaminated by heavy metals (such as tailing dams), being still in a phase of study and preliminary implementation, requires an in-depth study from an environmental point of view as it depends heavily on the location, weather conditions and chemical conditions of the soil. The identification of the candidate plant species for planting will take place through a process of study and analysis of the species, analyzing the advantages and disadvantages of the plants that will be used in the project.

The present study revolves around the main objective of laying the foundations for the realization of a future experimental and operational planting in the site under study. To achieve this, it will be necessary to study the mechanisms and processes involved in detoxification and purification of contaminated soil. Phytoremediation in general will be treated first but the focus will be on the reclamation of soils contaminated by heavy metals, as in the case of the mining industry.

To determine the planting project it will be necessary to consider and treat in detail all the factors that affect the success of the intervention. The objective of the feasibility study, in addition to the description of the site and the spatial location of the same, will be to obtain an accurate characterization of weather, carried out through the analysis of atmospheric variables related to phytoremediation plants. The variables analyzed will be those of temperature, precipitation, evapotranspiration, relative humidity, solar radiation and wind. Accurately identifying the climatic conditions of the area will provide a complete picture of the environment in which the plant species will grow. In addition to atmospheric conditions, the chemical aspect of the soil will also be treated, identifying the main contaminants and their concentrations. It will enable the extent of soil contamination to be understood and will provide insights into the plant species to be considered promising in this area.

The selection of plants, carried out through a detailed decision-making process, will deal with both the Chilean national context, through the identification of endogenous species and the deepening of studies on the subject, and international aspect, thus shifting attention to similar studies carried out in other parts of the world. The objective will be to identify and report the plants in relation to the contaminant that they can treat more but also identifying the possibility of acting on soils affected by multi-contamination.

Chapter 2

Phytoremediation

2.1 Introduction

Phytoremediation technique has been the subject of attention as an innovative and economical alternative to the most established treatment methods actually used in hazardous waste sites. This technology is considered as an environmentally friendly approach and allows to treat and remove contaminants in a cheap and safe way, as the field experiment showed.

Bioremediation has been studied for several years, as many studies justify, and it is from some years that beginning to find employment in the mining industry, with technologies called *phytomining*. The environmental problem arising from waste from the mining processes are well known and dealt with in the previous chapters. The generic term *phytoremediation* consists of the Greek prefix *phyto*, that means plant, attached to the Latin root *remedium* indicating something to correct or remove an evil. The engineering definition of this methodology, given by Hinchman et al. (1995), is:

> "The engineered use of green plant to remove, contain, or render harmless such environmental contaminants as heavy metals, trace elements, organic compounds, and radioactive compounds in soil or water. This definition includes all plant-influenced biological, chemical, and physical processes that aid in the uptake, sequestration, degradation, and metabolism ofcontaminants, either by plants or by the free-living organisms that constitute the plant rhizosphere."

As can be understood from the previous definition, the discpline that studies this technology is very wide and complex. The engineering aspect of using specific plants and their properties to degrade, extract, contain or immobilize contaminants from soil and water must take into account many factors that may influence the success of the process. The various factors, starting from the various contaminants that can be tolerated or absorbed up to the different mechanisms put in place by the plants, require a consistent preliminary study and analysis at different degrees of detail.

2.2 Phytoremediation strategies and treatment typology

Plants suitable for Phytoremediation treatments can use various detoxification techniques according to the characteristics of the plant and depending on the chemical nature and physical properties of the contaminant to be removed. The main phytoremediation strategies, listed and described below, are six but more than one can be used at the same time (Favas et al., 2014).

1. Phytoextraction (Phytoaccumulation, Phytoabsorption or Phytosequestration);
2. Phytodegradation (Phytotransformation);
3. Phytostabilization (Phytoimmobilization);
4. Phytovolatilization;
5. Phytofiltration;
6. Rhizodegradation (Phytostimulation).

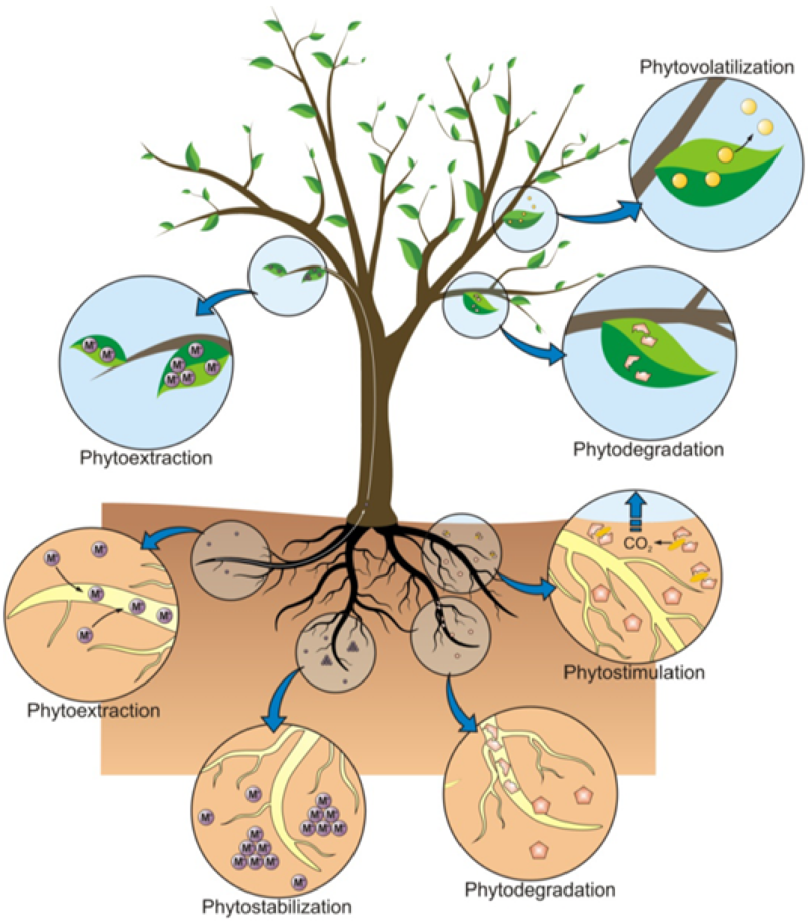

Figure 2.1. Schematic representation of phytoremediation strategies, from Favas et al. (2014).

Phytoextraction Phytoextraction methodology, also identifies with the names Phytoaccumulation, Phytoabsorption or Phytosequestration, involves the absorption of contaminants by the roots, followed by translocation and accumulation in the aerial parts, buds and leaves (Hazrat et al., 2013). This technique is used in situ for the treatment of contaminated soils and is mainly applied for the treatment of metals (Cd, Ni, Cu, Zn, Pb) in addition to elements such as Arsenic (As) and other organic compounds (Favas et al., 2014). The plants used are mainly of the hyperaccumulative type, that is, they are able to store high concentrations (0,01% to 1% of dry weight, depending on the metal), but the study of the suitable species must also take into account the objective of phytoextraction. The removal of contaminants occurs with the harvest of plants, so a high production of biomass is preferable even if the accumulation is lower. The process will continue with incineration and will end with landfill disposal.

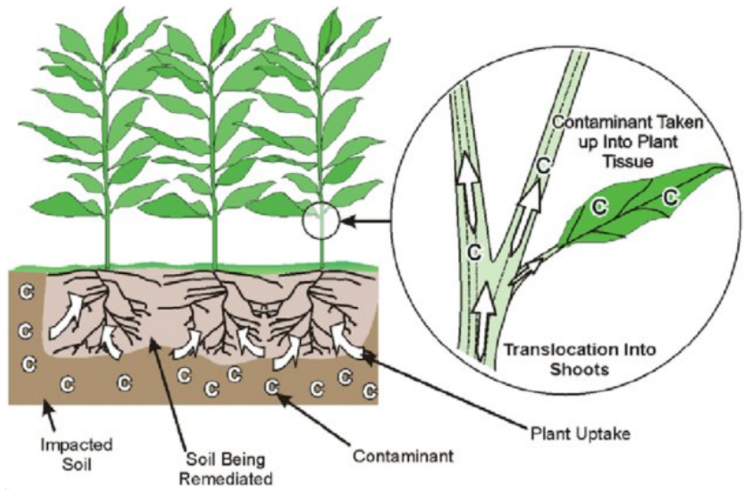

Figure 2.2. Phytoextraction mechanism, from ITRC (2001).

Phytostabilization The Phytostabilization reduces the mobility of contaminants and limits their spread in soil. The pollutants, usually organic or inorganic, are immobilized and incorporated into the lignin of the cell wall of the root system while the metals create insoluble forms that fall into the rhizosphere. Immobilization also allows to reduce the bioavailability of contaminants and inhibit them to possible migrations in the aquifer (Barceló and Poschenrieder, 2003). However, this technique is a management strategy, it is not able to remove the contaminant from the reclaimed soil until the plant is eradicated.

Phytodegradation Through Phytodegradation technique, contaminants, usually organic, are degraded or metabolized by plants within plant cells, with the help of specific enzymes. The involved enzymes in phytodegradation process are: (1) dehalogenase (transformation of chlorinated compounds); (2) peroxidase (transformation of phenolic compounds); (3) nitroreductase (transformation of explosives and other nitrate compounds); (4) nitrilase (transformation of cyanated aromatic compounds); (5) phosphatase (transformation of organophosphate pesticides) (Cristaldi et al., 2017). Unfortunately, the phytogradation is limited only to the removal of organic pollutants because heavy metals are not biodegradable (Favas et al., 2014). As some studies have shown, this methodology can be applied to the treatment, in addition to the various organic pollutants, also to synthetic herbicides and insecticides (Hazrat et al., 2013).

Phytovolatilization The Phytovolatilization is the process in which pollutants are first absorbed at the root level, then transported and released into the atmosphere through the aerial parts of the plant, in less toxic forms. This technique is applicable for decontamination of organic compounds such as tetrachloroethane, trichloromethane and tetrachloromethane and certain metals (typically Hg, Se and As) having a high volatility (Cristaldi et al., 2017). The pollutants removed, however, are only transferred from one compartment, that of the soil, to another, that of the atmosphere and therefore there is the possibility of a reintegration into the environment through precipitation (for example on lakes and oceans).

Phytofiltration The rhizofiltration allows, through absorption, concentration and precipitation in the root system, the removal of organic and inorganic pollutants from groundwater, surface and wastewater (Cristaldi et al., 2017). The plants used can be of aquatic or preferably terrestrial type, seen their more developed and fibrous root, and are maintained in a hydroponic filtration system applied in situ or ex situ. The main difficulties of this methodology are related to pH regulation, the need for a first greenhouse cultivation and the frequency of harvests and subsequent disposal (Barceló and Poschenrieder, 2003). This technique has shown excellent results for the removal of heavy metals (collected from roots and subsequently collected) and for radioactive elements.

Rhizodegradation The rizodegradation involves the biodegradation of organic contaminants at the level of the root system of the plant. This degradation occurs in an area of the soil called rhizosphere, in which degrading microorganisms are present, even in quantities greater than 100 times the surface) that use exudates and metabolites of plants as a source of carbon and energy. This process has some advantages, such as a low cost of installation and maintenance and an in situ treatment, but it is a slow process and with greater efficiency only in the first 20-25 cm of depth. Rhizodegradation improves soil characteristics and is particularly useful for the remediation of soils contaminated with a wide range of chemicals, including Pahs, pesticides, polychlorinated biphenyls (Pcbs), BTEX compounds (benzene, toluene, ethylene benzene, xylene) (Cristaldi et al., 2017).

The phytostabilisation and phytostabilization are related to containment processes whereas phytoextraction, phytodegradation and phytovolatilization are classified as removal processes.

Table 2.1. Phytoremediation overview and recap, from U.S. Environmental Protection Agency (2000).

Mechanism	Process Goal	Media	Contaminants	Plants	Status
Phytoextraction	Contaminant extraction and capture	Soil, sediment, sludges	Metals: Ag, Cd, Co, Cr, Cu, Hg, Mn, Mo, Ni, Pb, Zn; Radionuclides: Sr, Pu, Cs, U	Indian mustard, pennycress, alyssum, sunflowers, hybrid, poplars	Laboratory, pilot, and field applications
Phytodegradation	Contaminant destruction	Soil, sediment, sludges, groundwater	Organic compounds, Chlorinated solvents, phenols, herbicides, munitions	Algae, stonewort, hybrid poplar, black willow, bald cypress	Field demonstration
Phytostabilization	Contaminant containment	Soil, sediment, sludges	As, Cd, Cr, Cu, Hs, Pb, Zn	Indian mustard, hybrid poplars, grasses	Field application
Phytovolatilization	Contaminant extraction from media and release to air	Groundwater, soil, sediment, sludges	Chlorinated solvents, some inorganics (Se, Hg, and As)	Poplars, alfalfa black locust, Indian mustard	Laboratory and field application
Phytofiltration	Contaminant extraction and capture	Groundwater, surface water	Metals, radionuclides	Sunflowers, Indian mustard, water hyacinth	Laboratory and pilotscale
Rhizodegradation	Contaminant destruction	Soil, sediment, sludges, groundwater	Organic compounds (TPH, PAHs, pesticides chlorinated solvents, PCBs)	Red mulberry, grasses, hybrid poplar, cattail, rice	Field application

2.3 Heavy metals (HMs) and Bioavailability

Heavy Metals Heavy metals are conventionally defined as chemical elements with an atomic number >20 and metal properties. The most common are Cadmium (Cd), Chromium (Cr), Copper (Cu), Mercury (Hg), Lead (Pb) and Zinc (Zn). HM are natural components in the soil and some of them are the basis of the composition of many living organisms, and also of our own body. The HM naturally occurring in the soil are generally in low concentrations and result from pedogenetic and rock degradation processes .

The problems arising from these substances, however, begin to show themselves when concentrations in the environment and in our body reach levels higher than those that can be tolerated, bringing serious consequences also lethal. The definition *Heavy Metals* identifies those chemical elements which have the following common characteristics:

- density greater than 5.0 g/cm^3;
- behave as cations, that is, ions with a positive charge when they enter an electromagnetic field;
- low solubility of their hydrates;
- tendency to create complex chemical bonds;
- great affinity with sulfides, in which they tend to concentrate (chalcophile elements);
- may have different oxidation states depending on pH.

Generally, natural processes such as degradation from rock together with translocation or accumulation of such soil components generate concentrations of less than 0.1%. Higher percentages of heavy metals are often attributable to anthropogenic activities sources of contamination such as agriculture (fertilizers, pesticides), mining (during operation and after disposal), metallurgy, energy production, industrial emissions and many others (Adriano, 2001). Heavy metals have residence time in the soil for thousands of years, representing numerous dangers.

The HMs, on the plant point of view, can be divided into two categories: some play the role of micro-nutrients needed for plant growth, such as Zn, Cu, Mn, Ni and Co, while others have an unknown biological function, such as Cd, Pb and Hg. As far as the first category is concerned, these metals play an essential role in plant growth and, as an excess of them can have harmful effects, so too is a shortage of them. Regarding metals such as lead and cadmium, they have no biological function and their phytotoxic action is already evident in small concentration (Liphadzi et al., 2005; Pietrelli et al., 2004).

The toxicity, resulting from the presence of these metals in the soil, is due to their ability to interact in biological mechanisms at the cellular and molecular levels. Considering the overall effects, HMs generate an alteration of plant functions such as photosynthesis, respiration and the absorption of mineral nutrients. In addition, entry into the food chain endangers the health of living beings. The danger of these substances is mainly due to the fact that they cannot be biodegraded and the ability to bioaccumulate, causing various health diseases and disorders.

Bioavailability To determine the ecotoxicity of HMs for plant species it is not enough to know the total concentration of heavy metals present in the soil but rather it is important to evaluate the fraction that is bioavailable and therefore absorbable by organisms. The definition, given by Peijnenburg et al. (2007), identify Bioavalability as "the fraction of the total amount of a chemical present in a specific environmental compartment that, within a given time span, is either available or can be made available for uptake by (micro)organisms from either the direct surrounding of the organism or by ingestion of food".

In soil, heavy metals (HMs) are present in solid phases and in solution. Referring to the Figure 2.3, the mobility of metals decreases downwards, from high mobility in pore water to low mobility in the crystalline structure, but always dependent on chemical conditions of soil. Generally, in the solid phase, unless chemical changes, the HMs are immobilized, inert and harmless, while in the aqueous phase they are mobile and toxic (Ogundiran and Osibanjo, 2009). In addition, chemical and biochemical factors such as pH, oxidizing potential, soil texture, quality and quantity of organic matter, composition, temperature and groundwater regime influence the mobility of heavy metals (Plette et al., 1999).

To complicate the study, it is often not only necessary to determine such soil factors but the study should be concentrated in the area of the rhizosphere, that area of space that extends for a few millimeters, surrounding the roots of the plant, and is influenced by their activity. For these reasons it is important to perform a soil characterization through chemical methodologies, with selective and/or sequential extractions, or biological, through determination of the target organism and measurement of bioaccumulation.

Figure 2.3. Breakdown of heavy metals in soil, from John and Leventhal (1995).

TOTAL
- In pore water
  - Free aquo Ion
  - Inorganic complexes
  - Organic complexes and chelates
- Weakly adsorbed
- Associated with carbonate
- Associated with Fe, Mn oxides
- Complexed by organics
- Associated with sulphide
- In the mineral lattice

2.4 Plants used in phytoextraction

Phytoremediation includes different treatment techniques, as explained in Chapter 2.2, differentiated according to the type of contamination and the purpose to be achieved. As for heavy metal contamination, the most common technique is represent by Phytoextraction. Generally speaking, contaminated environments are often hostile to plant growth and the high rates of toxicity do not allow common species to resist. As Subhashini and Swamy (2013) report, the plant used in Phytoextraction should possess the following characteristics:

1. have a rapid growth rate and high biomass production;
2. hyperaccumulator of heavy metal and tolerate the toxic effects of heavy metal;
3. translocate metal from root to shoot;
4. resistant to pathogens and pests;
5. easy to cultivate and harvest;
6. have no attraction to herbivores to avoid its entry into food chain.

Some studies, as Rajesh et al. (2016), have highlighted that some species have developed detoxification and tolerance strategies, which can be classified as exclusion or resistance mechanisms. The first mechanisms, an exclusion one, prevent negative effects by acting within sensitive cells, preventing the accumulation of toxic concentrations. Instead, the mechanisms of resistance provide for the development of certain proteins able to counteract the negative effect of heavy metals.

The studies of suitable plants and their behaviour have as a starting point the classification and characterization of the species which populated polluted sites having high concentrations of toxic substances. Evidence from some studies, as (Baker and Proctor, 1990; Macnair, 1993), showed that generally species inhabiting such sites evolve a higher tolerance in relation to habitat, they do not already belong to species tolerant to metals for some hereditary characteristic.

Taking into account the mechanisms adopted and the complex biological variables in place, plants growing on metal-contaminated soils can be classified according to their ability to absorb and transfer metals to the aerial parts, in three different categories.

Excluders Metal "excluders" have as characteristic to limit, to keep low and constant the concentration of metals in the aerial parts. It occurs by limiting the entry of contaminants from the roots and often these plants have very reduced processes of transport from the roots to the leaves. The detoxification of metals occurs mainly in the roots but the problems appear when the high concentrations accumulate producing lethal effects for the vote of the plants. Most metallophytic plants are excluders (Wójcik et al., 2017).

Indicators Metal "indicators" are plants in which there is a linear relationship between the concentration of metals in the shoot and the concentration in the soil (Concentration coefficient shoot/soil is usually close to 1 (Wójcik et al., 2017). In these plant species the radical absorption and translocation to the aerial parts remains relatively constant for a wide range of HMs concentrations.

Accumulators Metal "accumulators" have excellent ability to concentrate elements in their tissues, regardless of concentrations in the soil and without showing signs of toxicity. This mechanism is made possible thanks to the storage of toxic substances in less sensitive parts of the plant (Wójcik et al., 2017). The ratios between concentration in soil and/or root and concentration in shoot are higher than 1. In the category of the accumulators we must also distinguish the hyperaccumulators, that is those plants that manage to accumulate in the aerial parts very high concentrations of metal, in relation to the normal range. They are able to tolerate concentrations higher than 2% of the dry weight of the plant and the threshold limit to differentiate between accumulators and hyperaccumulators plants has changed over the years and depends on the type of contaminant. The current values are shown in Table 2.3.

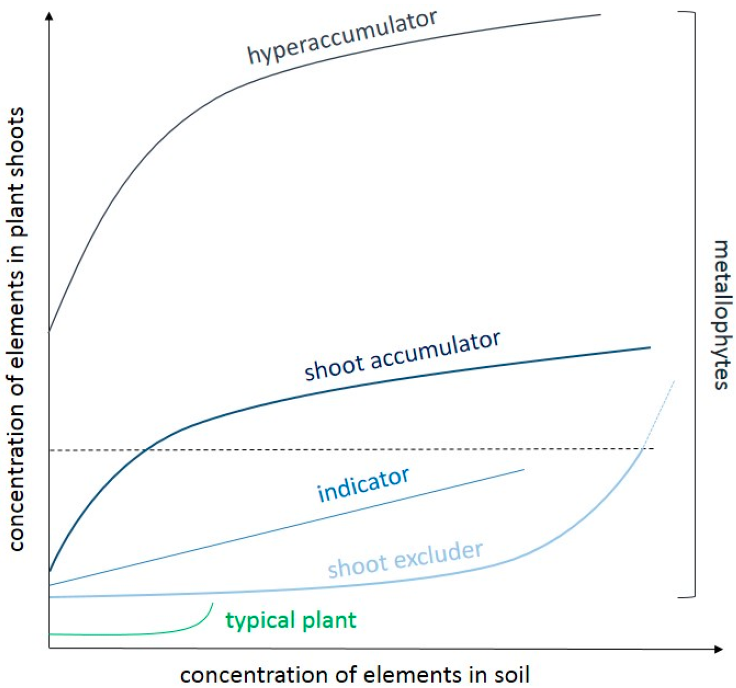

Figure 2.4. Conceptual response diagram demonstrating the potential relationship between the concentration of metals/metalloids in aerial parts of the plant and available content of metals/metalloids in the soil, from Muszyńska and Labudda (2019).

The graph in Figure 2.4 relates the concentration of elements in the soil to that in plant shoots, showing the behaviors of the various species. The normal plant has a short life cycle with increased bioavailable metals in the soil and can only tolerate low concentrations. Instead, the curve representing the "excluders" has a linear and constant trend of the concentration in the shoots as the one in the soil increases, showing, however, problems, till the death of the plants, due to high concentrations of HMs in the soil. The "indicators" species have a higher resistance to contamination, showing a linear curve. "Hyperaccumulators" have a logarithmic trend and have developed mechanisms that allow them to resist at higher concentrations of bioavailable metals much more than other species. The dotted line indicates the "hyperaccumulator" threshold for different metals and metalloids.

Table 2.3. Concentration range of heavy metals found in plant tissues, minimum limit for accumulator and maximum limit for hyperaccumulative species (mg kg^{-1}), from Baker and Brooks (1989).

Metal	Norma range	Minimum accumulator level	Maximum hyperaccumulator level
Cadmium (Cd)	0.1-3	20	100
Cobalt (Co)	0.03-2	20	1 000
Chromium (Cr)	0.2-5	50	1 000
Copper (Cu)	5-25	100	1 000
Manganese (Mn)	20-400	2 000	10 000
Nikel (Ni)	1-10	100	1 000
Lead (Pb)	0.1-5	100	1 000
Zinc (Zn)	20-400	2 000	10 000

2.4.1 Plant selection process

Having explained the previous concepts, however, the technical approach to be used in the choice of the species is now under discussion.

Typically, the phytoremediation study involve the collaboration between engineers, hydro-geologists, and other professionals due to its complexity. The problem of remediation already presents, itself, some complications to which others are added, like interaction of the plants with the soil and the high dependence on the site conditions and location.

The main objective of the plant selection process, showed in Figure 2.5, is to choose a plant species, preferably autochthonous, with appropriate characteristics for growth and phytoremediation under certain site-specific conditions. To begin the process of selection of plants, of course, we have to take into account some important points from which to begin the research of the species that are suitable for our case.

1. *Plants present in similar sites:* samples of plants growing on the site, together with other site-specific knowledge, as well as extrapolations from phyto-purification research, may present important insights. However, these deductions must be followed by laboratory or field studies of the species concerned.

2. *Plants investigated in similar studies:* an important step to follow is certainly that of the analysis of the bibliography and scientific publications on phytoremediation. These documents can often be important, because in recent years much progress has been made in this area. In-field testing takes a long time and the available data can facilitate the search for the appropriate species;

3. *Native plants:* obviously, these types of plants have the characteristic of being able to grow under the conditions of the specific regional zone. The list of these plants can be available online.

In general, individual surveys produce a list of plants based on the criteria described. These lists, when compared, can often present situations in which some plants are common, means that are present in more than one list. They represent candidate plants, for which remediation is more likely to be effective, and therefore require a more in-depth study using additional information (U.S. Environmental Protection Agency, 2000).

The additional information allows to analyze and study the phytoabsorption capacity of the selected plants. The realization of phytoabsorption system requires a greater deepening in the study of the root system of the plants. This study requires the analysis of the type of roots and the depth they can reach. A root system with an high number of roots, such as the fibrous type, provides maximum contact with the soil thanks to the high surface of the roots.

The depth of the root must be investigated in relation to the layer of soil to be treated. It represents a greater degree of complexity, as laboratory studies can often lead to substantial errors. The growth rate can be under or over estimated if the study doesn't consider local conditions such as water depth and soil characteristic such as water content, structure, density, fertility or other conditions.

It is important that the depth of the root zone exceeds the depth of in situ contamination or of the excavated soil. In the shallower zone the extraction or absorption of contaminants is more marked as most of the mass develops in that zone and decreases along the depth. The depth depends on the type of plant and varies from values of 1.5/3 meters for Shrubs and Grasses, up to 9 meters of Legumes or even a mass of 30 for Trees (U.S. Environmental Protection Agency, 2000). These maximum depths are relative to normal environmental conditions while for contaminated areas, some cite studies, like Dobson and Moffat (1995), showed a reduction in root depth up to 90% less, thus making the possibility of bio-treatment for depths up to 2/3 meters.

Another parameter that can directly influence the remediation efficiency is the growth rate. This parameter should be defined differently for the different mechanisms of phytoremedition. Fast growth in terms of root depth, density, volume, surface area is preferred for rhizofiltration, rhizofiltration and phytoostalization. Instead, in relation to phytoextraction, a rapid growth rate of the surface plant mass is preferred. In general, a large radical mass and large biomass are preferred because it allows a greater mass of contaminants that can be accumulated and a greater assimilation of them. In addition, a rapid growth rate will reduce the time needed to reach a large biomass.

This situation, of course, concerns the ideal case. With regard to the metal hyperaccumulators used, they are able to extract a high quantity of some metals, but, however, the production of biomass is low and the growth rate is slow. For this reason it is important to make a comparison with plants capable of producing a greater amount of biomass even if they have a lower accumulation of contaminants because what counts in the end is the total mass extracted.

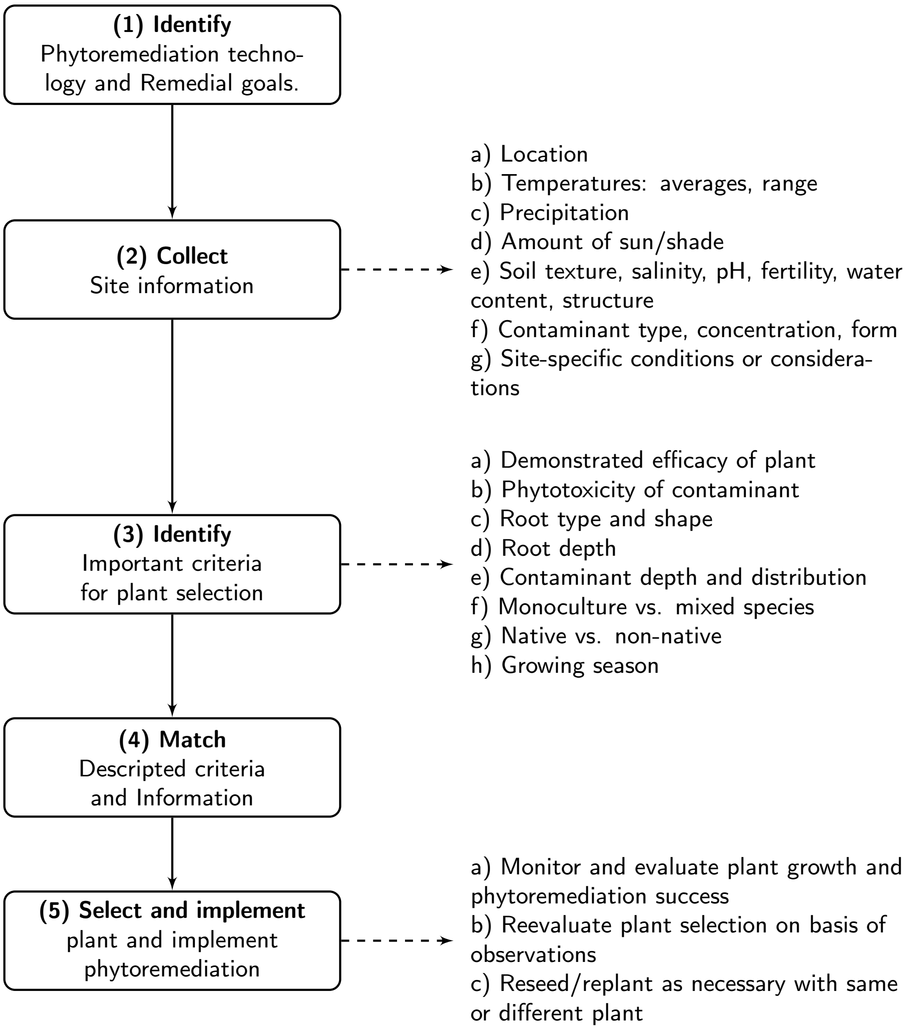

Figure 2.5. Flowchart of Plant Selection Process, from U.S. Environmental Protection Agency (2000).

2.5 Evaluation criteria of phytoextraction

The phytoextraction potential can be estimated by calculating the bioconcentration factor (BCF) (or biological absorption coefficient) and the translocation factor (TF).

Bioconcentration factor (BCF) The bioconcentration factor (BCF) from soil to leaves, defined as the ratio of the total element concentration in the harvested plant tissue (C_{plant}) to its concentration in the growing soil (C_{soil}) as follows:

$$BCF = \frac{C_{plant}}{C_{soil}} \tag{2.1}$$

where C are expressed in (mg/kg).

This factor is an indicator of how efficient a plant is in up-taking heavy metals from soil and concentrating them into its tissues **?**. Obviously, only with BCF it is not possible to establish if the studied plants may be considerate as hyperaccumulator species for certain metals, this is why we use also Translocation Factor (TF). Generally, BCF has been categorized into three groups, plants with BCF = 1 are called Indicator and have no influence, plants with BCF less than 1 can be an excluders and plants with BCF bigger than 1 can be considered Accumulators, with regards to high value that are considered Hyper-Accumulators (Radulescu et al., 2013).

Translocation factor (TF) The translocation factor (TF) also indicates the plant's efficiency in extracting heavy metals and it is defined as the ratio of the total concentration of elements in the aerial parts of the plant (C_{shoot}) to the concentration in the root (C_{root}) (Favas et al., 2014). The formula is:

$$TF = \frac{C_{shoot}}{C_{root}} \tag{2.2}$$

2.6 Comparison with other remediation methodologies

The phytoremediation methodology for the recovery of contaminated land is added to the many remediation technologies already in place. The fundamental principles on which all remediation operations are based are transformation, removal and immobilization. The first principle can be applied to organic contaminants and allows gradual transformation into simpler intermediate compounds by achieving complete mineralization. The removal is mainly used for heavy metals due to their persistence in soil and their non-degradation over time; for this reason it is necessary to separate the removal from the contaminated matrix. Immobilisation does not involve extract or degassing of the contaminant but is only applied in cases where other methodologies cannot be effective; this principle avoids only a further extension of pollution.

The classification of the remediation processes used to clean up the sites contaminated by HMs can be made on the basis of the place of intervention, differentiating works in-situ or ex-situ, on-site or off-site or according to the type of treatment, choosing between Biological, Physical and Chemical (showed in Figure 2.6). These techniques can also be used in combination to increase the removal efficiency and the choice is made on the basis of economic factors, site-specific characteristics and remediation objectives.

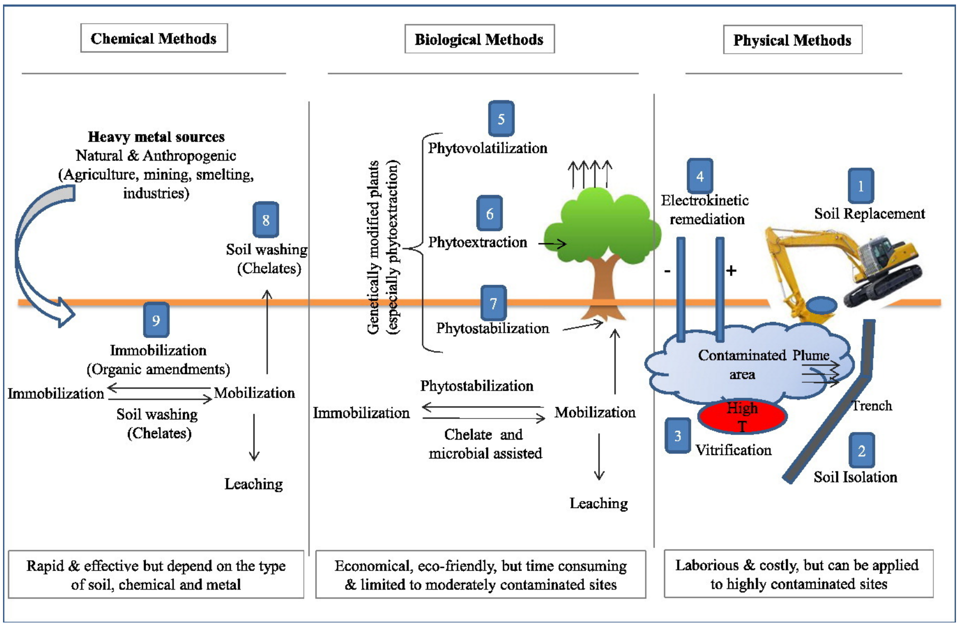

Figure 2.6. Different remediation technologies grouped by method, with detail, from Khalid et al. (2017).

Physical remediation Physical treatments are cited among the types of remediation and could seem to be beneficial alternatives, from the environmental point of view, to the classic technologies of contamination removal. Such methodologies, however, only allow the separation of contaminants from the solid/liquid matrix and if implemented alone entail high disposal costs in landfill for contaminated material. Often the combination with chemical treatments, with the use of special reagents, allows the stabilization or destruction of contaminants. Physical remediation treatment can also be applied to highly contaminated sites.

- *Soil replacement:* consisting of partial or total replacement of contaminated soil, through excavation operations and off-site disposal. The addition of clean material allows the dilution of contaminants. This technique was the most used before the 90s because it is well suited to highly contaminated soils with a small area but is expensive given the high labor effort;

- *Soil isolation:* this technology prevents the movement of HMs or at least tends to limit it within a bounded area preventing further contamination (Zhu et al., 2012). The intervention mainly aims at the protection of groundwater through the installation of barriers surrounding the site with vertical extension up to the waterproof layers. The downside is that such interventions are only temporary and are carried out only when other methodologies cannot be economically or physically installed.

- *Vitrification:* this treatment, through the contribution of high temperatures, allows the separation of contaminants by desorption/ volatilization, the destruction of them by pyrolysis or immobilization by fusion of the solid matrix. This methodology can be applied in different situations and is carried out through the entry into the ground of electric current through electrodes. This remediation technique can be applied both in-situ and ex-situ, but cost analysis prefers the first option. Some critical aspects are related to possible gaseous emissions, the creation of intermediate contaminants and the impact of temperature in the soil matrix. The main limitations are related to soil conditions, to determining factors such as humidity and the presence of alkali and to the possibility of use only in small scale for heavy metals (Khalid et al., 2017).

- *Electrokinetic remediation:* this new physical methodology allows the separation of contaminant by electrophoresis, electrical infiltration or electromigration (Yao et al., 2012). The Electrokinetic technique is often associated with other processes such as chemical, biological, with permeable barriers and others. A dissolution operation with specific electrolytes is previously required to treat heavy metals, but efficiency depends on various environmental factors to be determined on a case-by-case. Being an economic intervention in relation to efficiency, the total costs increase considering the preliminary operations

Chemical remediation Chemical treatments reduce the toxic effect of contamination through a series of chemical reactions. The main types of reactions used are oxidation, reduction and neutralization (Evanko and Dzombak, 1997). In cases of heavy metal pollution, chemical treatments are intended to reduce the availability and mobility of metals through specific reagents. The problems of such treatments may result from the possible creation of more toxic intermediate substances than the original ones or from the fact that the reagents can themselves be a source of contamination (Evans and Furlong, 2003).

- *Immobilization:* during this treatment, the addition of chemical agents allows the immobilization of contaminants through reduced mobility, bioavailability and bioaccessibility of heavy metals. The compounds used are mainly organic and inorganic and allow the modification of soil properties, such as pH, producing the precipitation of metals and above all create complex bonds able to immobilize heavy metals (Khalid et al., 2017).

- *Encapsulation:* To limit contamination, the Encapsulation method can also be applied by mixing contaminated soil with products such as asphalt, lime or concrete in order to immobilize harmful compounds. Concrete is the most widely used material in this process but this technology is still used to a limited extent (Pandey et al., 2012).

- *Soil washing:* the washing technique uses reagents and extracting solutions to allow the leaching of HMs from the contaminated soil. The matrix with heavy metal is removed through excavation operations and then mixed with the extraction solution, defined case by case according to the type of metal and soil. During washing, the contaminants pass into the liquid phase from which they will then be separated. After washing operations, the soil, to be reused or stored on the site, must respect certain established parameters. This technique has proved to be very efficient in terms of extraction and environmental impact, while in economic terms, preliminary operations can impact on intervention costs (Khalid et al., 2017).

Biological remediation Biological remediation methods consist of the use of micro-organisms or plants in order to degrade and transform toxic compounds. This innovative solution, considered economic and permanent, allows the recovery of the soil through mineralization and transforming heavy metals into less toxic or less bioavailable compounds. Biological treatments have a high potential in terms of recovery and intervention costs, operating directly on site and are non-invasive. The main disadvantages are the design difficulties associated with environmental conditions and the relative slowness of the process. The main techniques used, as explained above, are:

- *Phytoremediation*;
  - Phytovolatilization
  - Phytostabilization
  - Phytoextraction
  - Phytoextraction by genetically modified plants
  - Chelate assisted phytoremediation
- *Microbial assisted phytoremediation.*

In conclusion, after other remediation methodologies have been shown, the need to restore the damaged landscape and the high portions of contaminated land shifted the focus on more environmentally friendly types of treatment such as biological. The considerable costs and the high alterations of the substrate by the classic chemical-engineering techniques have pushed an acceleration in the research and development of new biological techniques.

Chapter 3

Feasibility study

3.1 Site description

The site is located in O'Higgins region (region VI), about fifteen kilometres (15 km) north-west of the city of Rancagua and around ninety kilometres (90 km) south of the capital of Chile, Santiago. The tailing dam, site of the study, lies at Latitude 34°04'42" S and Longitude 070°48'48" W and it is composed of two deposits. The owner of the deposit is "Compania Exploradora Y Explotadora Minera Chileno Rumana SA" and the material comes from copper processing in a nearby mine of the Chancon complex.

As reported by SERNAGEOMIN (2020), Chancon 1 and Chancon 2 deposits contain sterile material of 345 100 and 44 457 tons respectively but are currently inactive. The total cover area is about 46 411.96 m^2 and the shape is easily detectable at the foot of the mountains.

Figure 3.1. Satellite photo of Chancon tailing site, from Earth Explorer (2020).

The image extracted from the satellite, showed in Figure 3.1 allows us to understand the high extent of the storage area of the material. In addition, as can be identified, the storage is dry as the material has already completed its evaporation cycle.

Figure 3.2. Photo from the top of Chancon tailing dam deposit.

Figure 3.3. Photo from the top of Chancon tailing dam deposit.

The Chancon tailing deposits are showen in Figure 3.2 and Figure 3.3. The first photo, taken from the top of the deposit, shows the high extension of the tailing dam. The second photo was taken from the bottom and clearly shows the contrast and the visual impact created in relation to the surrounding environment and nature: a disturbance on the mountain landscape.

3.2 Meteorological condition of the site

The meteorological characterization of the site under study is a fundamental preliminary phase. This study is also very important in cases, such as this one, of the design of phytoremediation and phyextotraction plants. Plants have a high sensitivity to weather conditions and need certain conditions to develop and grow over time.

The study aimed to analyse the different variables detected in two weather stations treating the data of temperature, precipitation, irradiation and wind. This chapter will also deal with the evapotranspiration of soil using the Thornthwaite method. In order to evaluate the seasonal changes, it is important to know that there are four seasons in Chile: summer (December to February), autumn (March to May), winter (June to August), and spring (September to November).

Through Vismet portal (www.vismet.cr2.cl/) it was possible to identify several meteorological stations in the vicinity of the site, as indicated in the Figure 3.4, and a selection was made based on the closer proximity to the site and the possibility of access to historical data. The two meteostations chosen are Graneros Norte and Punta Cortes, property of "Red Agroclimática Nacional - Agromet".

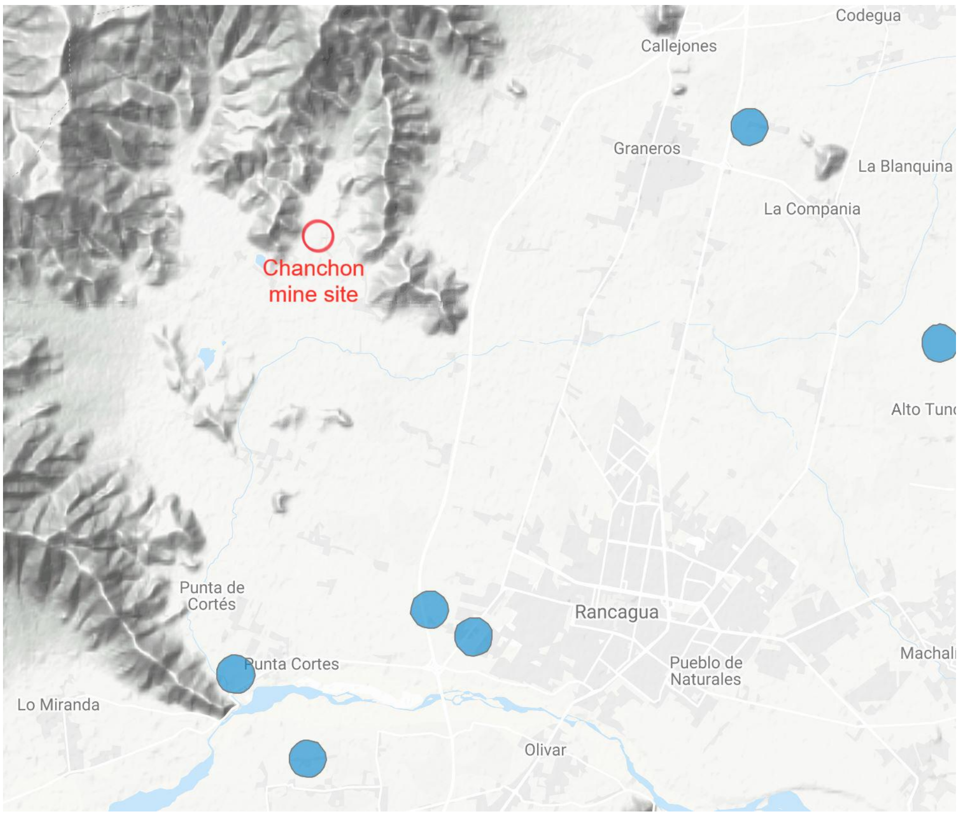

Figure 3.4. Map of available weather stations in the proximity of the site, from VisMet (2020).

The raw data was found through the Agromet portal (www.agromet.cl/) and relates to the period 2010-2019 for the station Graneros Norte and the period 2013-2019 for the station Punta Cortes. This information shall include:

- Minimum, maximum and average temperature, expressed in °C;
- Hourly precipitation, expressed in mm;
- Relative humidity, expressed in %;
- Maximum solar radiation, expressed in W/m^2;
- Maximum wind speed, expressed in m/s, and direction of wind.

The two weather stations are about 14.35 km apart and are located to the north east and south east of the site respectively, equidistant about 10 km. The difference in altitude is 18 metres. The Satellite photo, shown in Figure 3.5, indicates the linear distance. In the Table below the principal information, such as identification code, coordinates and altitude of weather stations, are indicated.

Table 3.1. General information regarding weather stations, from VisMet (2020).

Meteo Station	Graneros Norte (Graneros)	Punta Cortes (Rancagua)
ID station	AG62	AG65
Confidence data	100%	100%
Latitude	-34.061	-34.169
Longitude	-70.705	-70.791
Altitude	494 m	476 m

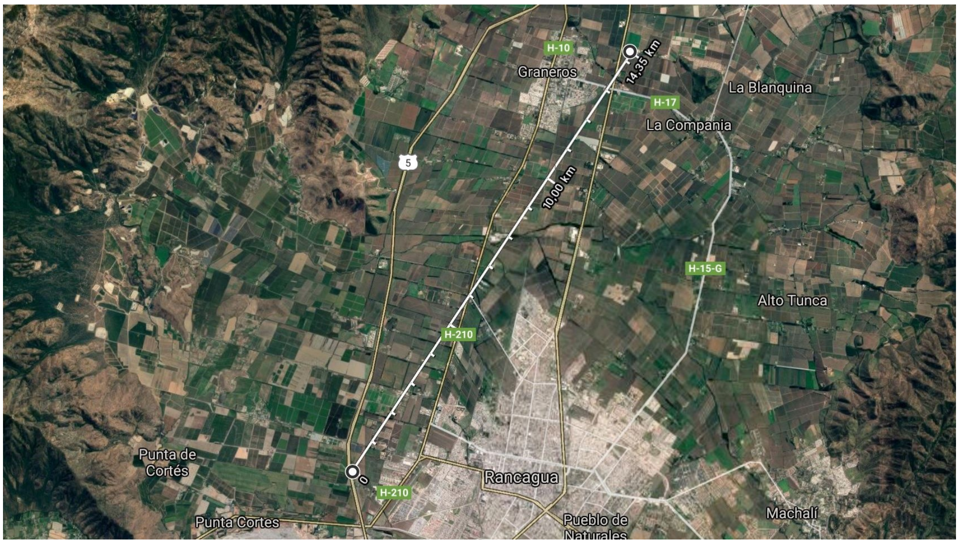

Figure 3.5. Satellite map with indication of air distance between two stations, from Google Maps (2020).

3.2.1 Temperature

Historical temperature data obtained from the Agromet site have been corrected by eliminating abnormal values due to malfunctions or errors in the recording and are reported in the tables below.

Table 3.2. Monthly Average Temperature - Graneros Norte weather station.

	Year	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sept	Oct	Nov	Dec
Average Temperature (°C)	2010	20.6	19.0	17.6	11.8	10.1	7.2	5.2	8.7	11.4	13.6	17.3	17.7
	2011	20.6	19.5	16.4	12.3	9.2	6.5	6.0	8.2	12.3	14.2	17.0	20.3
	2012	20.7	20.5	18.5	13.4	11.0	8.9	6.0	8.4	12.0	13.3	17.4	18.1
	2013	21.0	20.2	16.8	12.5	9.4	7.2	7.2	8.4	10.9	14.5	17.1	20.9
	2014	20.5	19.3	17.1	11.5	10.5	6.0	7.2	10.5	11.8	15.9	17.3	19.2
	2015	21.6	20.0	19.1	14.0	10.3	7.4	8.7	10.8	12.3	13.3	17.0	19.9
	2016	21.0	20.7	17.9	13.0	12.1	7.3	9.0	13.0	23.0	15.5	18.7	19.6
	2017	22.6	20.7	18.0	13.5	10.2	6.7	7.9	8.9	11.7	13.7	17.7	20.5
	2018	20.9	20.8	17.3	14.2	10.8	7.3	7.4	9.4	12.2	14.7	18.6	19.8
	2019	21.4	21.7	17.9	14.8								

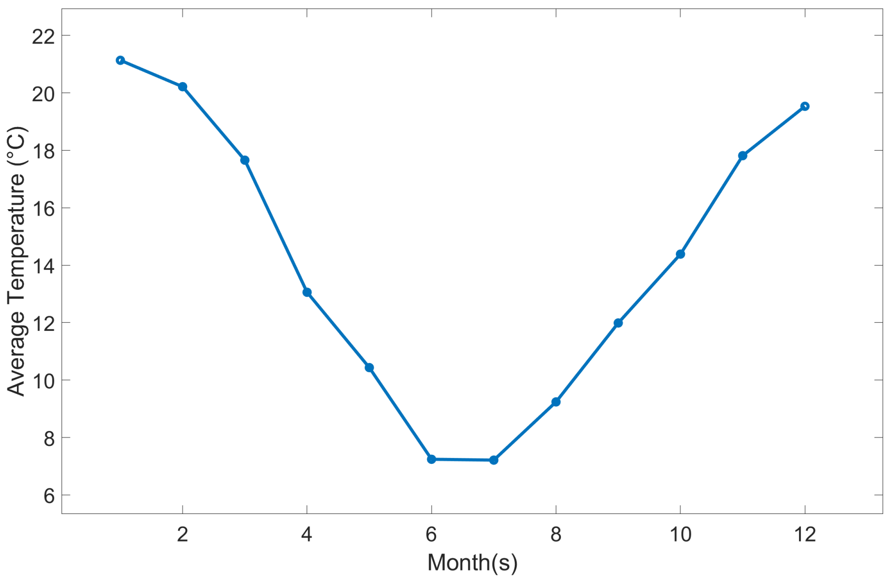

Figure 3.6. Graph with indication of average temperature in Graneros Norte station.

Table 3.3. Monthly Average Temperature - Punta Cortes weather station.

	Year	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sept	Oct	Nov	Dec
Average Temperature (°C)	2013	20.5	20.4	17.4	13.5	10.0	8.0	-	10.1	11.2	15.0	17.6	20.0
	2014	20.6	19.6	17.9	12.7	11.3	7.0	8.3	10.1	11.8	16.1	17.4	18.8
	2015	21.6	20.4	20.4	15.4	11.3	8.1	9.0	10.8	12.2	13.2	16.9	19.5
	2016	20.6	21.0	18.4	13.4	12.3	7.6	9.0	10.4	13.5	15.2	18.6	19.4
	2017	22.9	21.7	18.8	14.7	10.5	7.7	8.1	8.6	6.7	14.3	17.8	20.2
	2018	20.8	20.8	17.8	14.3	10.7	8.0	7.8	9.6	12.3	14.3	19.0	18.7
	2019	21.2	21.3	18.6									

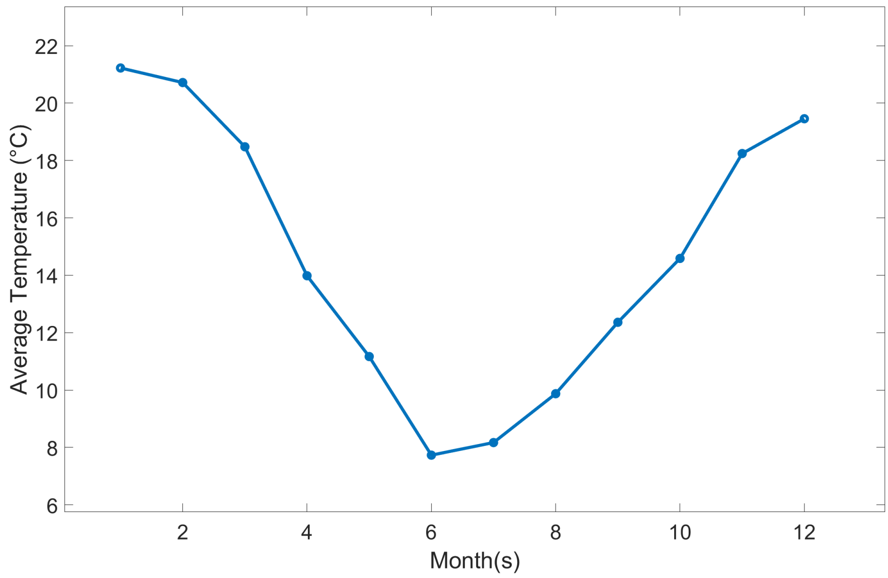

Figure 3.7. Graph with indication of average temperature in Punta Cortes station.

The temperature trend throughout the year is similar in the two weather stations, showing that June and July are the coldest months while January has an average temperature which is always above 20 degrees. Also, in this study, it is really important to determine the appearance related to the minimum temperature in the colder months. Unfortunately, in works that use and implement plants, temperature values below zero degrees Celsius (0°C) cause considerable problems in the management of the plant and in the correct operation. Analyzing the temperature data over time of both weather stations, the average daily temperature never drops below zero degrees. Focusing the analysis on an hourly scale results in a maximum value of 20 hours within a total 720, with temperatures below zero, during the coldest months.

3.2.2 Precipitation

As with other data, the precipitation values were extracted from the Agromet portal. It was necessary to correct the data from the reported outliers, as they could have influenced subsequent calculations. In the following Tables, 3.4 and 3.5, the total monthly values of millimeters of rain for the Graneros Norte station and for the Punta Cortes station are reported.

Table 3.4. Monthly Total Precipitation - Graneros Norte weather station.

	Year	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sept	Oct	Nov	Dec
Precipitation (mm)	2010	0.5	1.6	0.2	1.2	33.4	118.5	76.1	11.8	13.5	3.2	28.4	0.4
	2011	0.2	1.7	1.7	18.3	1.4	106.7	101.2	83.9	6.4	16.6	0	0
	2012	0	0.4	0	20.9	55	131	9.2	51	8.8	29.4	0.8	32.2
	2013	3	0	0	2.2	140.2	47	22.2	42.8	17.8	0.8	0	0.2
	2014	0	0	0.8	0.8	72	143.3	30.8	33.9	63.7	3.6	15.7	5.4
	2015	0	0.6	28.6	1.5	7.5	0.6	38.2	178.8	45.8	79.7	19	0
	2016	7.9	0	1.4	194.2	23.4	29	54.8	-	-	-	22.3	60.8
	2017	0	-	0	-	140.7	111.1	28.9	83.3	17.2	22.8	7.7	10.5
	2018	0	0	13.5	0.6	17.9	58.8	114.8	32.6	53.8	33.1	11.8	0.6
	2019	0.8	0.5	1.4	0.3								

Table 3.5. Monthly Total Precipitation - Punta Cortes weather station.

	Year	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sept	Oct	Nov	Dec
Precipitation (mm)	2013	0	0	0	0.2	93.8	22.4	-	-	13	1.2	0	0
	2014	0	0.2	0.4	0	26.8	51	12.6	34.6	24.6	1.4	6.2	5.4
	2015	0	1.9	25.1	0	1.6	0.4	19.2	74.9	31.4	39.1	18.1	0
	2016	4.1	0	0	44.2	5.4	7.8	7.6	1	0.2	176.1	6.9	1.8
	2017	0	0	0.2	23.4	36.8	46.9	23.5	80.9	0.4	2.4	7.6	0
	2018	0	0	10.1	0	20.2	39.7	88	53.6	41.8	14.2	1	0
	2019	0	0	0.1									

Looking and analyzing the data in the tables it is possible to conclude that the locality of Graneros Norte is more rainy. Despite that, the rainfall rate remains relatively low, never exceeding 200 mm of rain in the rainiest months. In Graneros Norte station some monthly data is missing due to maintenance and data logger recording problems.

3.2.3 Evapotranspiration

The Potential evapotranspiration (PET) is "the amount of water transpired in a unit time by a short green crop completely shading the ground, of uniform height, and never short of water", definition given by Penman (1948). In order to calculate the PET, in 1955, Thornthwaite and Mather developed an estimation method that is based exclusively on air temperature. This method involves the calculation of three different factors:

- Heat Index I, function of monthly average temperatures;
- The factor a, calculated with the formula shown below.

The process that allows the determination of the quota of the evapotraspirazione begins with the calculation of the Annual Heat Index (I), with the formula:

$$PET_{nc} = 16 \cdot \left(10 \cdot \frac{T}{I}\right)^a \tag{3.1}$$

where:

- T is the Mean Monthly Temperature, in Celsius grade (°C);
- The Monthly Heat Index (i) is:

$$i = \left(\frac{T}{5}\right)^{1.514} \tag{3.2}$$

- The Annual Heat Index (I) is:

$$I = \sum_{i=1}^{12} i \tag{3.3}$$

- The factor a is:

$$a = 0.49 + 0.079 \cdot I - (7.71 \cdot 10^{-5}) \cdot I^2 + (6.75 \cdot 10^{-7}) \cdot I^3 \tag{3.4}$$

In first approximation, the Potential Evapotranspiration (PET) estimations are based upon a 12-hour day (theoretical sunshine hours per day) and a 30-day month (Pascual-Ferrer and Candela, 2015). The values that are obtained should be corrected according to the real length of the month and the theoretical sunshine hours for the latitude of interest, with the formula:

$$PET_c = PET_{nc} \cdot \frac{N}{12} \cdot \frac{d}{30} \tag{3.5}$$

where N are the theoretical sunshine hours for each month and d number of days for each month. The chart shown in Figure 3.8 shows the average number of sunshine hours related to the Rancagua city.

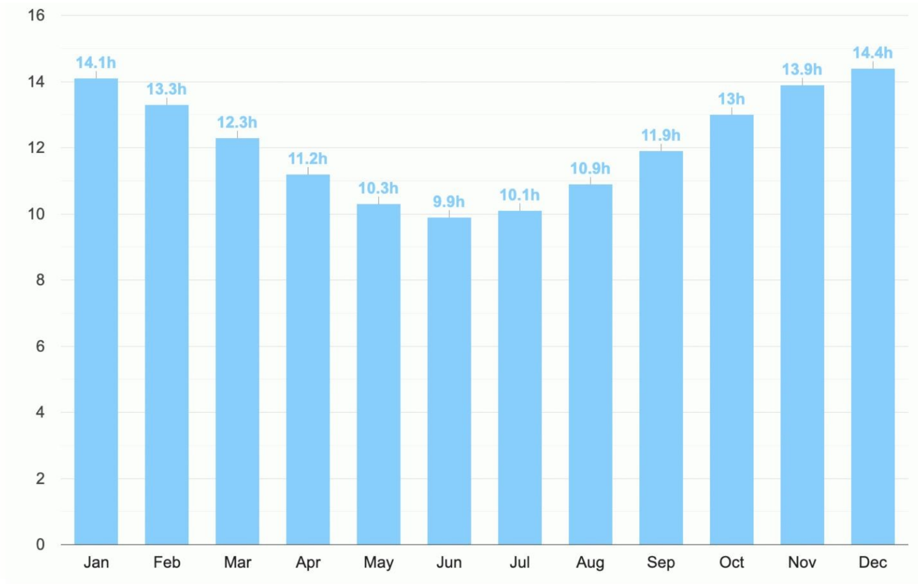

Figure 3.8. Average daylight hours in Rancagua, Chile, from Weather Atlas (2020).

Some bibliographical resources propose, in alternative, a method of correction of the values of evapotraspiration through the determination of the Correction factor L_i, function of the latitude of the site. It is a tabulated value, determined for each month and has the same function as the procedure shown above.

In Figure 3.9 the values of the Correction Factor as a function of latitude are reported, obtained from (Teegavarapu, 2012). These values are indicated for latitudes from 0 to 50 South and North in multiples of 10. A linear interpolation has been performed to identify the relative values to the latitude 34.061 South, related to the weather station Graneros Norte.

Latitude	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
0	1.04	0.94	1.04	1.01	1.04	1.01	1.04	1.04	1.01	1.04	1.01	1.04
10 N	1.00	0.91	1.03	1.03	1.08	1.06	1.08	1.07	1.02	1.02	0.98	0.99
20 N	0.95	0.90	1.03	1.05	1.13	1.11	1.14	1.11	1.02	1.00	0.93	0.94
30 N	0.90	0.87	1.03	1.08	1.18	1.17	1.20	1.14	1.03	0.98	0.89	0.88
40 N	0.84	0.83	1.03	1.11	1.24	1.25	1.27	1.18	1.04	0.96	0.83	0.81
50 N	0.74	0.78	1.02	1.15	1.33	1.36	1.37	1.25	1.06	0.92	0.76	0.70
10 S	1.08	0.97	1.05	0.99	1.01	0.96	1.00	1.01	1.00	1.06	1.05	1.10
20 S	1.14	1.00	1.05	0.97	0.96	0.91	0.95	0.99	1.00	1.08	1.09	1.15
30 S	1.20	1.03	1.06	0.95	0.92	0.85	0.90	0.96	1.00	1.12	1.14	1.21
40 S	1.27	1.06	1.07	0.93	0.86	0.78	0.84	0.92	1.00	1.15	1.20	1.29
50 S	1.37	1.12	1.08	0.89	0.77	0.67	0.74	0.88	0.99	1.19	1.29	1.41

Figure 3.9. Tabulated correction factor L_i in function of latitude, from Teegavarapu (2012).

In Table 3.6 the calculation values made to obtain the values of evotranspiration (ETP) for the weather station of Punta Cortes are shown. The two correction methods have given the same results.

Table 3.6. Evapotranspiration (ETP) calculations with Thorntwaite methos.

	Jan	**Feb**	**Mar**	**Apr**	**May**	**Jun**	**Jul**	**Aug**	**Sept**	**Oct**	**Nov**	**Dec**
Mean P (mm)	0.59	0.30	5.13	9.69	26.37	24.03	25.15	40.83	15.91	10.00	5.69	1.03
Mean T (°C)	21.22	20.72	18.48	13.98	11.17	7.73	8.17	9.87	12.36	14.59	18.24	19.46
T/5 (°C)	4.25	4.14	3.70	2.80	2.23	1.56	1.63	1.98	2.47	2.92	3.65	3.90
i (°C)	8.92	8.60	7.24	4.74	3.38	1.93	2.10	2.80	3.94	5.06	7.10	7.82

I	63.64
a	1.4909

ETP (mm)	99.60	86.77	81.01	51.74	38.24	21.38	23.99	31.82	43.05	56.95	76.91	87.48
P-ETP (mm)	-99.02	-86.47	-75.88	-42.06	-11.87	2.64	1.16	9.01	-27.14	-46.95	-71.23	-86.46

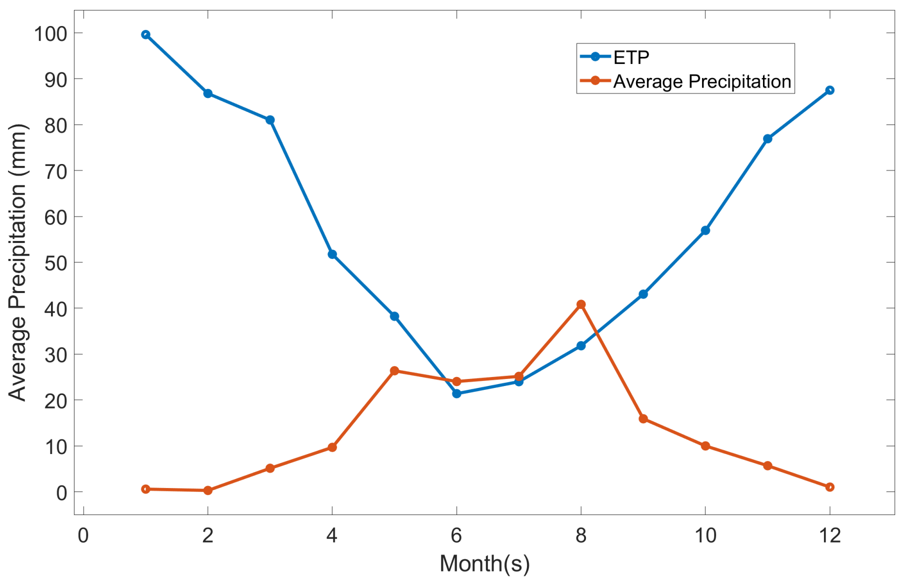

Figure 3.10. Graph with indication of contribution of Evapotranspiration and Precipitation.

Analyzing the graph and the data of the average precipitation together and the evapotranspiration, the average monthly precipitation is scarce especially in the summer months (from October to March) and reach its maximum peak of 40.83 mm during the month of August, while the ETP has an inverse course. Only during the months of June and August the contribution of precipitation is greater than the ETP from the ground.

3.2.4 Relative humidity

The water content of the air is an important factor for the success of HM phyto-extraction plants from contaminated soils. The amount of water transpired from the ground is dependent on the humidity of the air and the transpiration gradient is the driving force of this process. The study of this relative humidity variable is necessary as gross errors could be committed by the underestimation of it. In a practical sense, considering all the other factors equal, the higher the drying conditions of the air, the greater the water transpired by the plant, involving a greater translocation of contaminants.

Firstly, the dataset from Punta Cortes station was analysed, treating relative humidity data on a monthly basis. The values of monthly average relative humidity were plotted in a graph, shown in Figure 3.11, to better appreciate seasonal fluctuations.

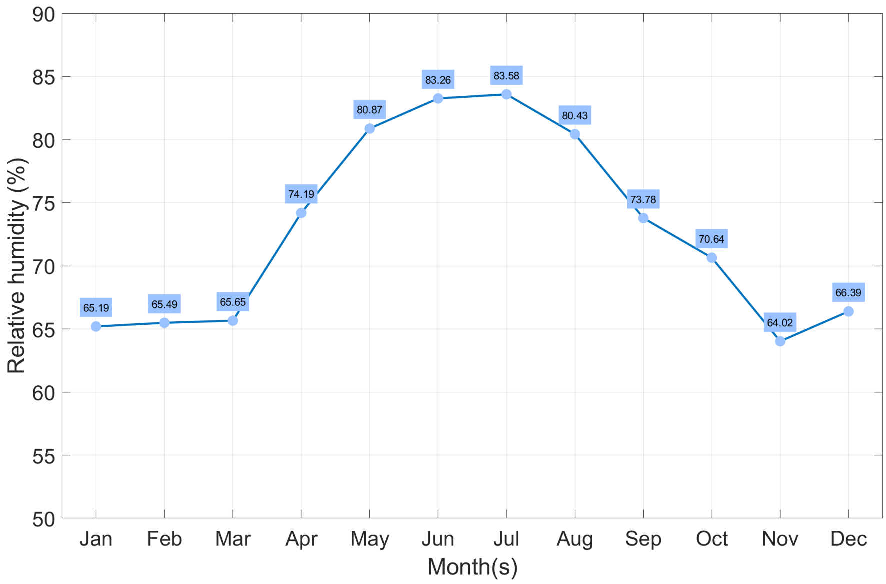

Figure 3.11. Monthly Average Relative Humidity of the air in Punta Cortes.

The analysis of the data shows that the relative humidity of the air in the first three months of the year is similar, that is around 65 percent. The major step occurs between the following months, when the average relative humidity jumps to 74.2% in April. The trend of the curve grows, reaching its peak in July, with a relative humidity of 83.6%. In the second half of the year the curve shows a decreasing trend, falling to 64% in November. The curve studied has a shape similar to the distribution of precipitation throughout the year.

3.2.5 Solar radiation

The measurement of solar radiation on the ground is rather delicate compared to other measurements more commonly carried out at meteorological stations (for example the temperature) but if carried out correctly it represents a radiometric measurement of excellent accuracy. The radiation power per unit area is measured in W/m^2 (Watt per square metre).

Solar radiation provides the energy for transpiration and this atmospheric variable affects plant growth and photosynthesis. This means that with less light the growth rate will be lower and consequently the photosynthesis process will be slower. Landmeyer (2011) and Mccutcheon and Schnoor (2004) studies demonstrated that there is a strong relationship between solar radiation and plant transpiration.

In the graphs below the Solar Radiation, determined in Punta Cortes station, are presented divided by season.

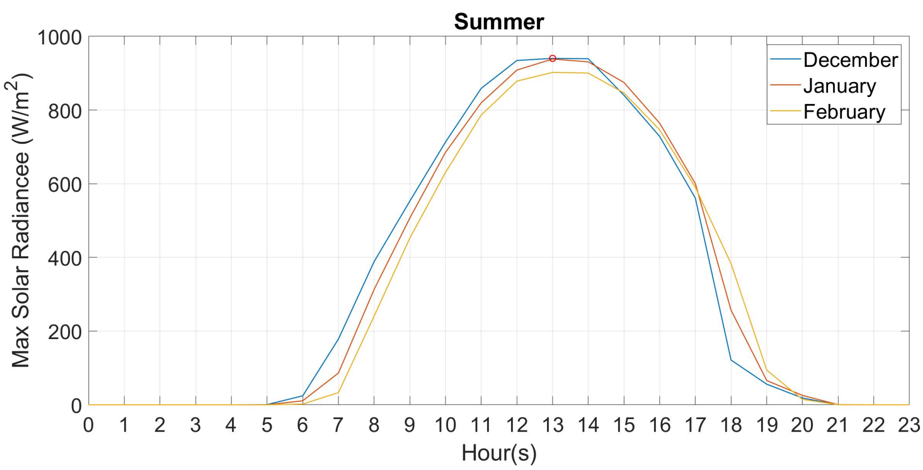

Figure 3.12. Comparison of Daily Average Solar Radiation for Summer months.

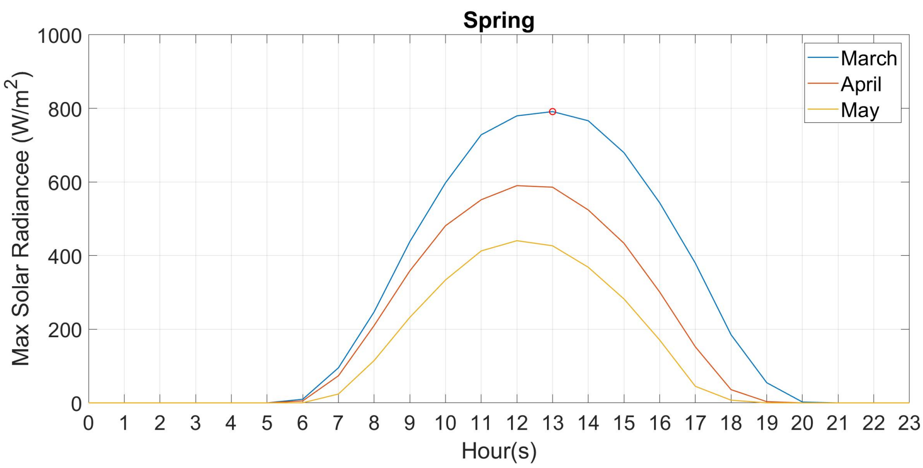

Figure 3.13. Comparison of Daily Average Solar Radiation for Spring months.

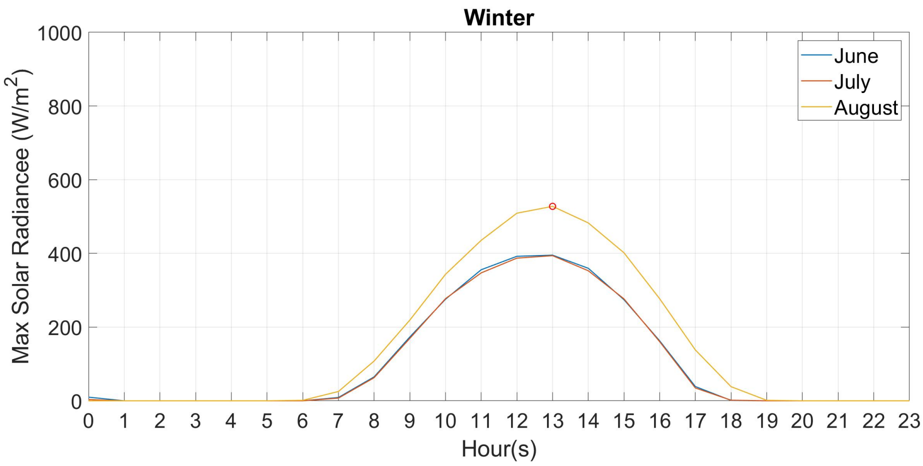

Figure 3.14. Comparison of Daily Average Solar Radiation for Winter months.

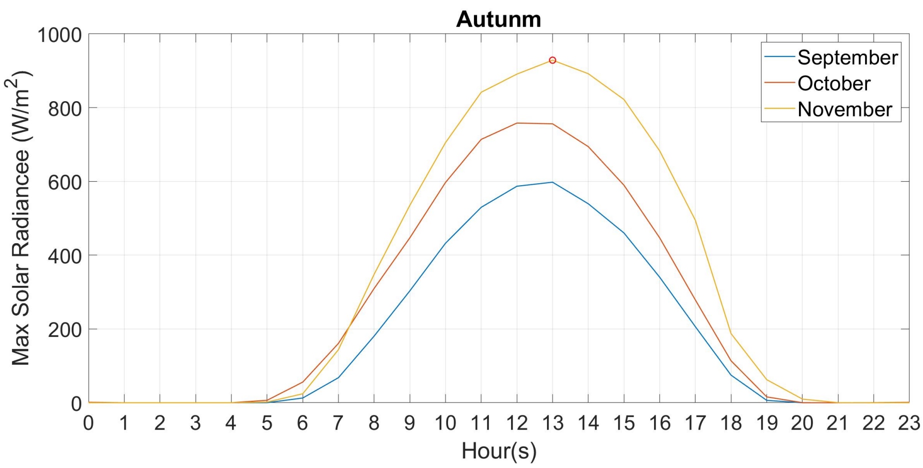

Figure 3.15. Comparison of Daily Average Solar Radiation for Autumn months.

Looking at the data, the daily trend of Solar Radiation looks similar for all seasons, having the characteristic parabolic shape with the daily maximum value at around mid day, that which changes greatly season by season is the value. During the summer, Solar Radiation reaches values of about 900-1000 W/m^2 while during colder months it has values below 600. The Spring and Autumn are transition seasons from the other two.

3.2.6 Wind

The analysis of wind speed and direction is necessary in Phytoremediation studies because wind affects the rate of transpiration and plant health. The effect of wind occurs mainly on windy days, as removing air in balance with the leaf in surface, increased transpiration occurs. However, wind gusts in combination with precipitation and high soil humidity can cause root damage until complete eradication of the plants (Landmeyer, 2011). Through the Agromet portal, as for the other meteorological variables, the Wind direction, expressed in °, and Wind max speed, expressed in m/s, were obtained.

The analysis was carried out by processing both sets of data on Matlab to obtain a Wind Rose diagram. It allows wind conditions, direction and speed to presented graphically over a period of time in a specific position, identified by our study in the weather station of Punta Cortes. The observation period runs from January 2012 to March 2019.

The diagram shown in Figure 3.16 includes 8 radiant rays representing the directions of the wind in terms of the cardinal directions of the wind (North, South, East and West) and their intermediate directions. The colours provide details on the speed, in m/s, of the wind from each direction while the percentage numbers indicated, for each concentric circle, represent the cumulative distribution of wind speed for each direction.

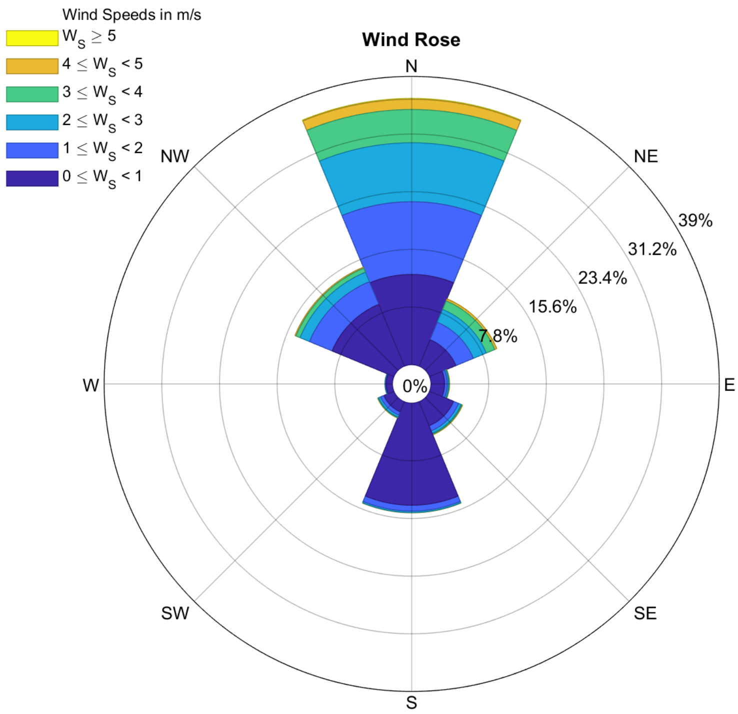

Figure 3.16. Wind Rose diagram of Punta Cortes weather station.

For a better understanding and analysis of the data shown in the Wind Rose diagram above, it is necessary to highlight some points:

- The main direction for the wind turns out to be the North. About 37% of the time the wind blows in this direction, reaching peaks of less than 5 m/s;
- The north adjacent directions, north-east and north-west, have winds with speeds of less than 5 m/s, extending respectively for about 10% and 15% of the hours;
- For the southern direction, the wind blows for about 15% of the time period but does not reach high speeds, remaining below 2 m/s;
- The wind blows in East and West directions only for a small percentage of time in the period and the speeds are less than 1 m/s.

In order to obtain an objective evaluation of the winds that characterize the site, the Beaufort scale was used (showed in Figure 3.17). It is an empirical measurement of the wind force and represent the main instrument of classification. The winds in Punta Cortes station reach maximum speeds of 5 m/s. These winds, called Light air, Light breeze and Gentle breeze, in increasing order of speed, cause at least light movements of the leaves and smaller twigs.

Beaufort number *BN*	Description	Limits of speed		Approximate wave height (m)
		knots	m/s	
0	Calm	1	0.3	–
1	Light air	1–3	0.3–1.5	–
2	Light breeze	4–6	1.6–3.3	0.7
3	Gentle breeze	7–10	3.4–5.4	1.2
4	Moderate breeze	11–16	5.5–7.9	2.0
5	Fresh breeze	17–21	8.0–10.7	3.1
6	Strong breeze	22–27	10.8–13.8	4.0
7	Near gale	28–33	13.9–17.1	5.5
8	Gale	34–40	17.2–20.7	7.1
9	Strong gale	41–47	20.8–24.4	9.1
10	Storm	48–55	24.5–28.4	11.3
11	Violent storm	56–63	28.5–32.6	13.2
12	Hurricane	64 and over	32.7 and over	–

Figure 3.17. Beafourt wind scale, from Molland et al. (2017).

3.3 Soil chemical analysis

The chemical analysis of soil can be a good starting point for the analysis of phytoremediation interventions. The analysis provides information about chemical component that are into the tailings soil and it is provided and updated each year by (SERNAGEOMIN, 2020).

SERNAGEOMIN is a Chilean government agency which has the role of providing information and regulating the mining industry in Chile. The Department of Mining Project Evaluation has created a national program, a characterization program for tailings deposits, with the purpose of monitoring of deposits of mining wastes. The last report was released on 13th January, 2020.

The tailings deposit, analysed in this study, is divided into two parts, called Chancon 1 and 2. The geochemical characterization of the samples allows to evaluation of the concentration of different elements:

- 12 main elements, rock-forming minerals, expressed as oxides in %. The elements are silicon (SiO_2), aluminium (Al_2O_3), titanium (TiO_2), iron (Fe_2O_3), calcium (CaO), magnesium (MgO), manganese (MnO), sodium (Na_2O), potassium (K_2O), phosphorus (P_2O_5) and sulphate (SO_3).
- Sulphur content (S), expressed in %;
- 30 common trace elements (less than 1%), expressed in g/t. The elements are showed in Table 3.1.
- 14 rare metals, expressed in g/t. The metals are Lanthanum (La), Cerium (Ce), Praseodimium (Pr), Neodymium (Nd), Samarium (Sm), Europium (Eu), Gadolinium (Gd), Terbium (Tb), Eysprosium (Dy), Holmium (Ho), Erbium (Er), Thulium (Tm), Ytterbium (Yb) and Lutetium (Lu).

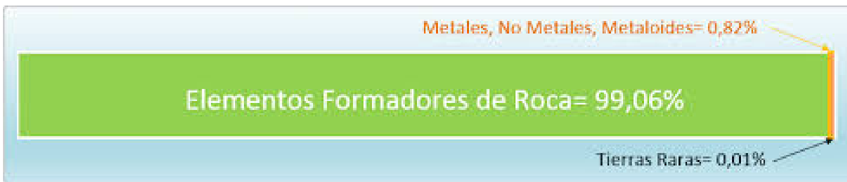

Figure 3.18. Components of the soil analysed, from SERNAGEOMIN (2020).

As can be seen from Figure 3.18, the elements that form the rock comprise the predominant percentage while the elements metals, non-metals and metalloids along with rare metals are a small percentage. However, the importance of their determination derives from the fact that the data provide concentrations in grams per tonnes (g/t), which is equivalent to parts per million (ppm), but when multiplied by the mass of a deposit, they show the real quantitative importance of each element and the impact that results from it.

The chemical study of the site of interest focuses on the analysis of the chemical components present and their amounts expressed in g/t. The values for both deposits are given in the Table 3.7.

Table 3.7. Chemical Analysis of Tailing dam, Chancon 1 and Chancon 2, data elaborated from SERNAGEOMIN (2020).

S.No.	Chemical Component	Result (g/t)	
		Chancon 1	Chancon 2
1	Copper (Cu)	983	1166
2	Vanadium (V)	91	97
3	Chromium (Cr)	71	65
4	Cobalt (Co)	<5	<5
5	Nikel (Ni)	6	8
6	Zinc (Zn)	6324	8795
7	Rubidium (Rb)	82	80
8	Strontium (Sr)	192	203
9	Yttrium (Y)	17	16
10	Zirconium (Zr)	312	185
11	Niobium (Nb)	7	6
12	Barium (Ba)	668	592
13	Lead (Pb)	829	919
14	Scandium (Sc)	6	19
15	Carbon monosulfide (Cs)	3.16	3.42
16	Hydrogen fluoride (Hf)	5.85	3.35
17	Tantalum (Ta)	1.57	1.27
18	Thorium (Th)	5.24	4.23
19	Uranium (U)	2.07	1.65
20	Arsenic (As)	501.62	567.33
21	Molybdenum (Mo)	<5	<5
22	Antimony (Sb)	<10	<10
23	Tin (Sn)	<20	<20
24	Silver (Ag)	2.02	11.39
25	Cadmium (Cd)	9.18	15.49
26	Bismuth (Bi)	<10	<10
27	Tungsten (W)	<10	<10
28	Gold (Au)	0.02	0.03
29	Mercury (Hg)	0.01	0.07

After the acquisition of the concentration data of the various chemical elements it is necessary to establish the actual degree of soil contamination. As previously presented in the chapter on legislation, there is currently no legislation in Chile on maximum permitted concentrations of toxic elements in soils. It is therefore necessary to use an international approach to legislation.

In order to carry out this assessment, Gonzalez et al. (2008) proposes to use of the guide values provided by the Swedish Environmental Protection Agency (www.internat.naturvardsverket.se) as a reference point. Around the year 2000, this agency carried out a collection of values, differentiated according to the type of contaminated matrix, with which to compare the concentrations measured at the site. For soil, the guide values for metals are reported in Table 3.8. It should also be noted that the values reported for contaminated sites can be considered contamination levels below which there is no short/long-term risk of adverse effects on man or the environment.

Table 3.8. Classification of current conditions for contaminated soil in terms of mg/kg *dw* based on guideline values for contaminated soil, retrieved from Swedish Environmetal Protection Agency (2002).

Chemical Element (Metals)	Slightly serious (Grade 1)	Moderately serious (Grade 2)	Very serious (Grade 3)	Extremely serious (Grade 4)
Arsenic (As)	15	15-45	45-150	150
Lead (Pb)	80	80-240	240-800	800
Cadmium (Cd)	<0.4	0.4-1.2	1.2-4	4
Copper (Cu)	100	100-300	300-1000	1000
Chromium (Cr)	120	120-360	360-1200	1200
Chromium (CrVI)	5	5-15	15-50	50
Mercury (Hg)	1	1-3	3-10	10
Nikel (Ni)	35	35-105	105-350	350
Vanadium (V)	120	120-360	360-1200	1200
Zinc (Zn)	350	350-1050	1050-3500	3500

In order to have a complete picture of the level of contamination and the importance of remediation, it is important to identify and classify chemicals according to their hazardousness. As reported by the Swedish Environmetal Protection Agency (2002), which sets out a classification table of the hazardous properties of chemicals in order to facilitate risk assessment, elements such as arsenic, lead, cadmium, mercury, and chromium (VI) are classified as "Extremely hazardous" while elements such as copper, nickel and vanadium are classified as "Very dangerous". Only zinc is classified as "Moderately dangerous".

The hazardousness can also be expressed through a weight assigned to each individual contaminant chemical, as reported in the study carried out by Lam Esquenazi et al., 2018 (2) that identifies the importance of intervening in a given site assigning a relative weight for the various elements. These numbers are shown in Table 3.9.

Table 3.9. Assigned weights to each element for the calculation of the weighted intervention ranking, modified from Lam Esquenazi et al., 2018 (2).

Chemical Element	Assigned weights
Arsenic (As)	3.0
Lead (Pb)	3.0
Cadmium (Cd)	3.0
Copper (Cu)	1.0
Chromium (Cr)	2.0
Mercury (Hg)	2.0
Nikel (Ni)	3.0
Vanadium (V)	-
Zinc (Zn)	1.0

Table 3.10. Grade of contamination of Tailing Dams soil by HMs, classified from Grade 1 (Slightly serious) to Grade 4 (Extremely serious).

Chemical Component	**Chancon 1** (g/t)	**Chancon 1** Grade of Contamination	**Chancon 2** (g/t)	**Chancon 2** Grade of Contamination
Arsenic (As)	501.62	Grade 4	567.33	Grade 4
Lead (Pb)	829	Grade 4	919	Grade 4
Cadmium (Cd)	9.18	Grade 4	15.49	Grade 4
Copper (Cu)	983	Grade 3	1166	Grade 3
Chromium (Cr)	71	Grade 4	65	Grade 4
Mercury (Hg)	0.01	Grade 1	0.07	Grade 1
Nikel (Ni)	6	Grade 1	8	Grade 1
Vanadium (V)	91	Grade 1	97	Grade 1
Zinc (Zn)	6324	Grade 4	8795	Grade 4

Following these observations, considerations can be made regarding the level of contamination at the site under study, assessing the concentrations of contamination in the tailing dams "Chancon 1" and "Chancon 2". As can be seen from the Table 3.10, the concentrations of arsenic (As), lead (Pb) and cadmium (Cd) are the most worrying, having a high degree of contamination and a higher relative weight. Chromium (Cr) and zinc (Zn) concentrations also have the highest degree, however, respectively a lower relative weight, 2.0 for Cr and 1.0 for Zn. Copper (Cu) needs attention because it falls within the degree 3 of contamination but despite this it does not have a high weight. The other elements have low levels of contamination, are "Slightly serious" and do not require a remediation focused on them.

Chapter 4

Case Study

4.1 Introduction

In the previous section the site under study was presented and a characterization was developed from a meteorological and chemical point of view. This allowed us to highlight the Chilean environmental situation and then go on to choose the species that could adapt to this situation. The analysis of this case study aims to highlight and study the possibility of treating tailing dams, characterized by high concentrations of heavy metals and trace elements, through the development of a plant for phytoremediation.

The case study focuses mainly on the use of the Phytoextraction methodology, suitable for deposits of materials in which the presence of water is poor and where it is not possible to implement a flow of water circuit. The objective remains to establish the basis for a future planting, identifying at this stage the plants, endogenous and not, suitable to carry out such tasks. Initially the bibliographic study will allow to find similar researches in the matter, analyzing first the situation at Chilean level, then with the study of the native and hyper-accumulative species, present in the southern nation and then moving with the global view of methodology. The aim remains to take into account all the factors that affect the success of the treatment and to obtain a design targeted to the case study.

During the study, the other phytoremediation methodology suitable for the treatment of heavy metals (HMs), phytostabilization, can also be considered but that only allows the stabilization in existence and re-population by plant species of the site of interest. The perspectives of this methodology are already shown in some experimental studies but the negative aspect remains that this operation does not allow the return of land to activities such as agriculture or the reuse of land for other purposes.

4.2 Bibliography review

An important point of the identification of the species and the definition of the plant suitable for the site concerns the study of the bibliographic sources. First of all, it is necessary to analyse the research carried out in Chile and its effects in this field in order to obtain a specific picture of the situation in which the work will be carried out by identifying the autochthonous species which have already shown positive effects in this field. In addition, the international bibliography will also be deepened to define the design methods and have a complete picture of the situation in countries more advanced and in which research has made progress. Obviously in the choice of the plants, as previously indicated, the environmental factor will prevail taking into account the climatic conditions to which the species must live.

4.2.1 Endogenous point of view

An analysis of the bibliography on Phytoremediation, focused in the Chilean country allows to obtain a general view and also provides an excellent starting point for our study. Environmental characteristics similar to our case study and research of the possible effects of hyperaccumulation of endemic species allow to carry out a more accurate plant design and a selection aimed at the goal of remediation. In recent decades, as a result of environmental advances in the field and the spread of new remediation technologies such as Phytomining, the hyperaccumulator species have gained more importance and have been the subject of studies to determine their potential. Generally, the search for hyperaccumulative or heavy metal tolerant species starts from the characterization of the plant communities diffused in the polluted environments.

Although the research on Phytoremediation has been going on since the 1990s, in South American countries, it has not been expanded for the reasons of lack of legislation and because very few native species suitable for the practice were found. As reported by Ginocchio and Baker (2004), hyperaccumulator plants are rare in the plant kingdom and by the year 2000 represented less than 0.2% of angiosperms, distributed between different families and selective for different types of contaminant. South America, despite being rich in natural mineral deposits and having a mining industry well rooted in the territory for some time, has a limited number of plants resistant to metals and metal hyperaccumulators compared to other areas of the world. The study also reported a list of metallophytic or hyperaccumulative plants in South America, highlighting that in the Chilean nation were identified only 6 plant species and classified as metallophytes against copper as they showed no hyperaccumulation but only resistance to contaminated soils.

The study of Ginocchio and Baker (2004) showed the importance of filling the void created by the lack of knowledge about metallophytes and hyperaccumulators throughout Latin America. Industrial and extractive activity also creates a potential risk to the country's biodiversity, whose damage must be mitigated by going to preserve and study the endogenous flora that inhabits the abandoned sites. As of 2006, only a few species resistant to metals and hyper-metals, only 172, had been identified in Latin America. This is because only a few scientific studies on soil characterization in the vicinity of mining areas had been published, thus creating difficulties in passing on the necessary information.

In 2008, a study conducted by Gonzalez et al. (2008) aimed to identify plant species with hyperaccumulator characteristics in Chilean environmental conditions. The investigation was conducted in two areas contaminated by various toxic elements including high concentrations of copper, in the proximity of a foundry. Research and species identification was carried out in 2005 and 2006. Only species with biomass suitable for a phytoextraction process were sampled within the test areas, excluding smaller species such as herbaceous plants.

During the study, as many as 22 species were identified with an above-normal degree of copper accumulation. From the chemical analysis of the foliar apparatus, however, unfortunately, no species has been found to be hyperaccumulative, with maximum accumulation that was 614 mg/kg twhile the limit for hyperaccumulator is 1000 mg/kg. This implies that the species with medium or low accumulation are metallophytic, that is able to tolerate heavy metals but do not have the characteristics suitable for phytoextraction plants. The species of major interest is *Oenothera affinis*, metallophytic species but with a high accumulation of metal therefore a possible candidate for works of extraction of contaminants. It was concluded that, despite the classification in the category of high accumulation and not hyperaccumulative metallophyte, it could have promising developments in the field of phytoextraction. The paper also sets the basis for new studies, emphasizing the need to understand the dynamics of absorption, accumulation and translocation within plant structures to increase the efficiency of processes and determine the best stage in which to collect plants.

In the following years very little progress in phytoextraction was made, because, despite the great biodiversity of Chile and the neighbouring states of South-America, endemic plants have developed mechanisms of resistance that allow them to survive in hostile places such as tailing dams but do not yet have good ability to translocate contaminants from the root to the shoot. The national studies, concentrated in the regions with great mining development, have only taken into account the phytostabilization technology, in order to repopulate the abandoned dams but not being able to return such land for other activities.

In 2011, a study carried out by Diaz et al. (2011) in the region of Antofagasta, aimed to assess the absorption of heavy metal by three native plants (*Pluchea absinthioides, Atriplex atacamensis and Lupinus microcarpus*). Dry soil conditions and soil contamination with levels of arsenic well beyond the normal limit result in unfavourable conditions for the development of vegetation. The analysis of the obtained data and the evaluation of the bioconcentration and translocation factors have shown how all three autochthonous plants selected concentrated the contaminant and transfer most of it into the leaves rather than the roots. The species *Lupinus microcarpus* had both the highest bioconcentration factor and the transport index of arsenic in the leaves. Obviously the potential of the species must be evaluated in relation to the environment in which they must go to act.

Some studies, such as that conducted by Lam Esquenazi et al., 2018 (1), have tried to identify new species, and assess their potential. The species *Adesmia ataca-*

mensis, endemic to Chile, has been found to be overaccumulating of copper as it has been shown to be able to accumulate and move large quantities of metal. It will be explained in detail in the next chapter.

4.2.2 Exogenous point of view

At a global level, studies on the various techniques of Phytoremediation have been varied and concern both practical applications and classification of hyperaccumulative species. Hampered by the lack of systematic screening of plant species and research, the discovery of hyperaccumulative plants has been directed only towards certain specific regions of the world.

The study of the Phytoremediation technique involves the participation of several experts in the field so the passage of information is essential to the good design of a plant. The behavior of the plants is studied both in the engineering field, then modelling the plant under the hydraulic, geotechnical and operational aspect, both in the chemical and genetic field, characterizing the behaviours of the various species and the mechanisms of tolerance and detoxification used. Worldwide, several studies have dealt with these aspects of the technique, finding it difficult to relate them in different territorial areas, given the high site-specificity of the subject. The search for over-accumulating, accumulating or contaminant tolerant plants (in the present case, heavy metals) is limited to a territorial field where the environmental conditions are homogeneous or similar, including water inputs, irradiation and soil conditions which influence remediation.

Several studies have dealt with this discipline going to investigate the potential applied to the various territorial cases. The technique has been applied, in addition to extreme cases of soil contamination such as landfills or in mining, also for the purification of contaminated water and waste water. Obviously the investigated plants have different characteristics according to the contaminated matrix and the objective of reclamation. The study of the global scope of the discipline can provide several useful information and a general vision but needs to be deepened with a site-specific characterization.

In addition to the study of the technology itself, other related aspects with the aim of identifying limitations and testing possible solutions must be considered. As stated in the chapter on phytoremediation, the main limitations of the technology concern the slow growth of plants and the limited production of biomass, thus affecting the efficiency of remediation of sites. Several solutions, in terms of treatment of plants and soil, have been applied, comparing over time, through field experiments, its effects with and without treatment. In addition, to consider, there is also the degree of availability for absorption, namely the phytoavailability of metals in the soil. This factor is influenced by numerous soil variables, such as cationic exchange capacity, pH and organic matter content.

In this respect, micro-organisms and amendments, such as chelating agents, which increase the bioavailability of metals in soil, may be added to improve metal accumulation capacities and plant uptake rates. Several agents such as persistent aminocyclic (APCA), such as ethylenediaminetetraacetic acid (EDTA) have been used in various phytotraction experiments. Recently, biodegradable APCA, disuccinate ethylenediamine (EDDS) and nitrilatriacetic acid (NTA) have been proposed as an alternative to EDTA and other persistent APCA (Evangelou et al, 2007). The application of EDTA soil has been the most efficient to improve phytoavailability. These new assisted treatment methods make it possible to take into account not only hyperaccumulative plants but also species that are only accumulative but with important characteristics in terms of biomass production and translocation from roots to leaves.

Over the years, with the aim of broadening the knowledge of this new tool for the reclamation of contaminated sites, some scholars have tried to create a global database for hyperaccumulative and metallophytic species, in order to be able to explore the different species that have shown scientific evidence on the subject and possibly test or explore them in different places. For the first time in 2012, Professor Famulari in collaboration with the expert Kyla Witz created a website (http://www.steviefamulari.net/phytoremediation) dedicated to this. The database is a collection and research of plant species that help to capture contaminants, including heavy metals and other materials such as organic, biodiesel, oils and radionuclides. The aim was to make this technology more accessible and easy to handle even by a non-technical audience. The classification of the plants, together with the description and source of the study, is based on the name of the contaminant and the type of contaminant.

Subsequently, also Reeves et al. (2018) highlighted the need for a detailed and accessible database, useful to provide information to any researcher intending to investigate the matter. In 2017, the database contained 721 species of which 523 for nickel, 53 for copper, 42 for cobalt, 5 for arsenic, 1 for chromium, 8 for lead, 20 for zinc and the remaining rare earth species manganese, selenium, thallium and cadmium. Also, the study provided a broad view on the distribution of hyper-accumulating plants globally, identifying the most represented families as that of the Brassicaceae (83 species) and the Phyllanthaceae (59 species).

4.3 Plant used on Phytoextraction treatment

As previously treated, plants suitable for phytoremediation treatments can be varied although their performance depends on the specific area in which they develop and the environmental conditions. These plants show detoxification mechanisms targeted for certain contaminants. Selectivity can be linked to a contaminant or show small accumulations for other heavy metals. The fundamental pre-eminence to be taken into account concerns not only the accumulation but also the tolerance to other elements in soils characterized by multi-contamination. Later, some species that have shown scientific evidence in the accumulation of contaminants and that could be excellent candidates for the phytoextraction system will be exposed in detail.

4.3.1 Plant focused on Arsenic accumulation

Arsenic is ubiquitous in all environments and its native concentrations in the soil generally range from 1 to 50 mg/kg, with average concentrations of 5 mg/kg. It may occur mainly in inorganic forms such as arsenite (Aso_3^{3-}) or arsenate (Aso_4^{3-}) and plant uptake is similar to phosphate. Obviously arsenic-contaminated sites usually have unfavourable and hostile soil conditions due to poor soil structure, low organic content, and pH alteration. Arsenic toxicity interferes with sulfhydric groups in the cells of most plants, showing toxic symptoms such as decreased plant growth and crop development. In general, much of the arsenic in the soil is not directly available to plants but in order to be absorbed by the root system arsenic must be in a bioavailable form. The following are some bibliographical studies of promising plants for the treatment of arsenic (Bondada and Ma, 2003).

Table 4.1. Hyperaccumulator plants suitable for Arsenic contamination soils.

Scientific and Common name of plant	Contaminant type	Notes
Pteris vittata L. (Chinese Brake Fern)	Arsenic, Chromium, Copper, Lead, Mercury, Nickel, Zinc	Hyperaccumulator, uses for Arsenic uptake
Agrostis castellana (Highland Bent Grass)	Arsenic, Lead, Manganese, Zinc	Origin in Portugal, As-accumulating proprieties
Atriplex atacamensis (Cachiyuyo)	Arsenic	Native of northern chile Adaptability in arid conditions

Pteris vittata L. The plant *Pteris vittata L.* belongs to the Pteridaceae family and is of Pteridophyte type. This type of plants are seedless vascular plants having a herbaceous behaviour, with underground stem and rooting apparatus which develops up to a maximum of 30 cm. The fern can reach heights of up to 50 cm, with alternate leafy leaves, with finely serrated edges. As can be seen from the Figure 4.1, the top of the leaves are dark green while on the bottom the color is lighter with evident ribs. This family is big and varied and has about 280 species distributed all over the world, but mostly in the tropical belt, adapting to live in warm-humid climates.

Figure 4.1. Pteris vittata L. species.

Since 2001, when its potential to accumulate large amounts of toxic substances such as Mercury, Lead and Arsenic was discovered, this plant is described in the literature as a hyperaccumulative plant. In addition, its characteristics make it an excellent candidate for phytotraction processes, as it is a perennial plant, having good development of biomass and a high efficiency of the processes of accumulation and transport from the roots to the foliar apparatus.

In 2001, Ma et al. (2001), in a study carried out in Central Florida at a site contaminated by Arsenate, discovered the effectiveness of this species in the extraction of As. *Pteris vittata L.* was the first known hyperaccumulative plant of As and the first fern to function as a hyperaccumulator. During this study large amounts of arsenic were recorded in the species, reaching concentrations of over 7000 mg/kg of As in fronds. This, when compared with the concentration values in normal and uncontaminated soils, having about 3 mg/kg of As in the leaves, gives an account of the ability of the species to tolerate and accumulate the pollutant.

Since the following years, as justified by the studies of Chen et al. (2002) and An et al. (2006), researches have been carried out on the accumulation of As by the fern. The species *Pteris vittata L.* has been identified also in China, both in the central and in the southern regions, studying the accumulation, the resilience in hostile environments and the tolerance also to other contaminants. In fact, in addition to recording concentrations of Arsenic of 5000 mg/kg in the leaves (Chen et al., 2002) and a high adaptation to the environment, in 2006 the accumulation of Zinc (Zn) reaching 737 mg/kg concentrations during the field study in the Chinese provinces of Guangxi and Hunan was also highlighted. For normal plants, total concentrations of Zn in soil between 70 and 400 mg/kg are toxic, so even if the accumulation by *Pteris vittata L.* does not reach the limit of 1000 mg/kg to be considered as hyperaccumulative, it could be very good for the reclamation of soils contaminated with Zn and As (An et al., 2006).

Danh et al. (2014) analyzed the critical aspects of fern use and introduced some practices to address these deficiencies. With regard to the accumulation of arsenic he analyzed the values of BCF and TF finding very variable values from site to site and highliting the greater affinity of the absorption system with phosphate compared to arsenate. The application of phosphate can therefore improve the efficiency of phytoremediation by effectively reduce the absorption and toxicity of arsenic in plants. As far as biomass production is concerned, in general most hyperaccumulators are wild plants with slow growth and low biomass. Such a feature for *P. vittata* can be greatly improved with the addition of fertilizers and shrubby mycorrhizal fungi. For the adaptability of the species, often the difficulty comes from the fact that the soils are multi-contaminated, but in the case of *P. vittata* the tolerance is high (the values are shown in Table 4.2).

With regard to the root system, the study carried out by Liao et al. (2003) highlighted the fact that the roots of this plant can penetrate up to 30 cm into the soil and thus efficiently extract contaminants. The higher density of roots is found in the first 10 cm so it is good for treating elements such as arsenic which are more concentrated on the surface. The cultivation of the species must take place in the first phase in the plant nursery, given the low rate of germination in field, and then transplanted in the field.

Table 4.2. Maximum concentration values tolerated by the plant species *Pteris vittata L.* The boxes marked with '-' means that there is no data available. Retrived from Danh et al. (2014).

Chemical Element (Metals)	**Maximum Tolerated Concentration (mg/kg)**
Arsenic (As)	100 000 mg/kg
Lead (Pb)	94 300 mg/kg
Cadmium (Cd)	284 mg/kg
Copper (Cu)	2 781 mg/kg
Chromium (Cr)	-
Mercury (Hg)	-
Nikel (Ni)	57.8 mg/kg
Vanadium (V)	-
Zinc (Zn)	84 700 mg/kg

To be taken into account in plant purification plants are the times of reclamation. It is still difficult to estimate this value, but by making some simplifications one can draw some considerations. The time required for recovery is a function of:

- Contaminant concentration in soil;
- Biomass production and yield;
- Number of plants that survive;
- Estimating the right time for collection.

Low biomass production can be a limiting factor in the application of *P. vittata* to the recovery of As-contaminated land on a large scale (Danh et al., 2014). Despite this, however, the species remains an important candidate for arsenic treatment plants by studying accessory applications of phosphate or other treatments to improve the efficiency of the process.

Atriplex atacamensis The family Atriplex, which includes over 250 species including *A. Atacamenis*, is very diverse and consists of annual or perennial species of herbs or shrubs. In this family many species are halophytic and develop in dry environments with salty soils. Nevertheless, this genus is distributed almost all over the world, starting from the subtropical regions up to the temperate and subartic ones. Chile, like all of America, is rich in this species, along with other countries such as Australia and Eurasia. Among the 23 native or endemic species of the South American country the shrub *Atriplex atacamensis* appears. It usually populates the beds of dry rivers and the overlap of this species with the areas of mining interest is clear. This shrub has a high branches, can measure up to 3 m in height and is able to produce high amounts of biomass. The leaves are of the order of 15 mm while the roots develop well also in depth, with a good degree of intake of substances from the soil. The shrub is shown in Figure 4.2.

Figure 4.2. Atriplex atacamensis species.

The first researchers to investigate the potential of the species were Diaz et al. (2011) and Tapia et al. (2013). The studies have been directed to the determination of the accumulation and translocation by the shrubs, also comparing the behaviors with other autochthonous plants of the same species. In 2011, Diaz et al. (2011) sampled the presence of arsenic, total and bioavailable fraction, in soil and the accumulation by native plants in the pre-andine area in northern Chile. Included among the plants was the species *A. Atacamensis*, the analysis of which showed no particular signs of toxicity or inhibition to growth due to the contaminant. The Arsenic concentration data collected, respectively 6.3 mg/kg in the leaves, 2.5 mg/kg in the stems and 1.7 mg/kg in the roots, in plants located in a soil containing 54 mg/kg and a pH of 8.3 if compared with other research indicates that the translocation takes place but is slower. This is because 90-day analyses showed low translocation and significantly higher concentrations in the roots.

Tapia et al. (2013) study showed that both *A. atacamensis* and *A. halimus* are resistant to Arsenic contamination, but the second species showed a greater total accumulation of As and a better translocation to the leaves. Nevertheless, analysis of BCF and TF values has shown that both species accumulate arsenic in moderate concentrations mainly in the roots.

In 2016, Vromman et al. (2016) conducted a specific study for the accumulation of arsenic within the species. The experiment took place in the north of Chile, precisely in the region of Antafogasta, a place characterized by high mining activity aimed at the extraction of copper. Chemical analysis of the only river in the area, the Rio Loa, from the Andes mountains showed an average concentration of arsenic of 440 µg per liter, due to natural effects such as volcanic and geothermal activity but also to human activities such as mining. This concentration is 100 times higher than the limit for drinking water. From the chemical analysis of the soils and from the studies of concentrations carried out on the plants, analyzing both the foliar and the radical apparatus, it has been shown that the species *Atriplex atacamensis* is able to survive and grow in difficult environmental conditions, with a high concentration of As and other heavy metals. For these reasons, therefore, the species has proved to be an excellent candidate for the reclamation of contaminated soils characterized by arid conditions.

In the same year, Fernandez et al. (2016) studied the species Atriplex, comparing the subspecies *A. Halimus*, *A. Nummularia* and *A. Atacamensis*. The study area, located in the Atacama Desert in northern Chile, is also characterised by concentrations of arsenic in the soil that require remediation. The climate is extremely arid, with very little rain and presence of winds of moderate to high intensity. The experiment lasted 5 months and involved the planting of species in quantities of 27 plants in total, with a random distribution in an area of 14 square meters. The analysis carried out through the determination of translocation factors and bioconcentration has highlighted the excellent phytostability capacity of the species Atriplex also showing the limits for phytotraction due to the low production of biomass in environments characterized by lack of water and high salinity. In addition, it has been shown that the species *A. halimus* and *A. nummularia* are preferable as the production of biomass was 30% by weight, higher than *A. atacamensis*.

4.3.2 Plant focused on Copper accumulation

Copper, expressed with the chemical symbol Cu, is a chemical element naturally present in the earth's crust and in all environmental media such as soil, air and water. It plays a vital role for plants, being an essential micronutrient for the development of the plant, going to favor the various processes of photosynthesis, respiration and protein production. Concentrations of Cu from 5 to 30 mg/kg within plant tissues are normal. Copper deficiency in plant tissue results in inhibition of growth, compromises the development and production of biomass while high concentrations may affect the morphology of the plant. In soils where the degree of contamination is high, with concentrations above 60-125 mg/kg, this metal becomes toxic, affecting the biological processes of non-tolerant plants and causing them to die (Napoli et al., 2019).

The high release of Cu and contamination can result from the development of anthropogenic activities such as mining operations (extraction and processing), agriculture (fertilizers, pesticides and fungicides), the disposal of solid waste (urban and industrial) and from treatment facilities. Copper released into the environment tends to stick to organic material and minerals, but when it dissolves, if it comes into contact with surface water it can be transported in the form of copper compounds or as free copper, or bonded to particles. (ATSDR, 2004) The highest contact and exposure routes of the population result from contamination of water and food, while the share taken through inhalation is much less significant. To date, worldwide, about 34 overaccumulating copper species have been discovered and a promising species for the present case study will be described.

Adesmia atacamensis This plant species, shown in the Figure 4.3, woody and branched type, is a plant endemic to Chile, precisely from the north and central Chile.

Figure 4.3. Adesmia atacamensis species.

It has been identified as the plant population of some copper extraction sites and this makes it adaptable in soil conditions with high contamination, in semi-arid climates such as those of the northern regions of the South American country. This species, besides tolerating drought situations, is adaptable also in soils with high salinity. This species, of wild type, is an hyperaccumulator but has a slow growth and a low production of biomass, characteristic common to several hyperaccumulative species.

The study carried out by Lam Esquenazi et al., 2018 (1) was aimed at evaluating the potential of the endemic species as an application in the phytoremediation processes of copper mine tails. In the study, carried out in the region of Antofagasta, in northern Chile, the concentrations of different heavy metals within the species were evaluated and analyzed. The environmental conditions, characterized by a semi-arid climate and the site under investigation, a deposit of mine tailings, allow to make a similarity with the subject of this study. The area in which the study was conducted presents unfavourable conditions for the growth of vegetation, due to the high solar radiation incidence and the absence of water. The ecosystem is defined as xeromorph, having high temperatures during the day, low rainfall and wide temperature fluctuation.

From the characterization of the flora in the analyzed mining areas it has been deduced that the vegetal species *Adesmia atacamensis* and *Cistanthe salsoloides* were present and common to the four analyzed mining areas. These species have been characterized and their average size been determined but the study continued only for *A. atacamensis* for reasons of availability. The study led to these conclusions:

- by analysing the values of BCF, A atacamensis appears to be an exclusion of Pb, Fe and Zn, therefore not suitable for the phytotraction of these metals, but with regard to Cu, the results suggest a capacity of hyperaccumulator;
- by analysing the TF values, the species A atacamensis has values greater than 1, a sign of the transfer of substances to the foliar apparatus, for metals such as Fe, Pb and Zn, while for copper, despite the high quantities accumulated, TF values remain always less than 1.

In conclusion, the species *A. atacamensis* was found to be excellent for phytoremediation porpouse. The high values of BCF have testified this but analyzing the values of TF it would seem to be more suitable for phytostabilization rather than phytoextraction. It has, therefore, the power to extract metals and translocation them but it is specified that the high values of metal present in the soil can influence the translocation from roots to leaves so such study and application requires a previous chemical characterization of the substrate of the site.

4.3.3 Plant focused on Lead accumulation

Lead (Pb) appears to be a chemical element among heavy metals of environmental interest as it causes environmental alteration and high phytotoxicity in cultivated areas. This metal is one of the most toxic and is widely spread on Earth. A small amount of lead is present in the environment due to natural causes but contamination of soil and water, both groundwater and surface, occurs as a result of anthropogenic industrial and mining activities. Pb remains in the environment for a long time and high levels of exposure can cause toxic biochemical effects in humans.

From the biochemical point of view, the mechanism of toxic activity of Pb derives from the strong affinity of the metal cation for sulphur. Its presence at low concentrations in all plant tissues has no deleterious effects but high concentrations within plants inhibit physiological processes and compromise some biochemical reactions essential for plant metabolism. Photosynthesis is one of the most sensitive processes to Pb contamination and toxicity symptoms are shown as growth reduction and leaf appearance. The contamination can then migrate from the soil to animals, mainly herbivores in which Pb is accumulated in various organs when contaminated plants are ingested (Liu et al., 2000).

Brassica Juncea L. The plant *Brassica juncea L.*, also known as Indian mustard, is a spontaneous annual plant and belongs to the Brassicaceae family. The stem of this plant is of ascending type and the flowers that grow are characterized by yellow petals, as can be seen in Figure 4.5. This plant can be annual or biennial when cultivated, and plants with a height of more than one metre and roots which penetrate to a depth of 100 cm can be produced. The species is native to Central Asia and has spread over the years to the Americas.

Figure 4.4. Brassica juncea L. (Indian mustard) species.

One of the important features of the *Brassica juncea L.* is the fact that the plant is very adaptable. This is demonstrated by the fact that it tolerates annual precipitation ranging from 500 to 4200 mm, average annual temperatures ranging from 6 to 27 degrees centigrade and pH from 4.3 to 8.3 degrees. It is used in some Asian countries as a fresh product and is also found in processed products. In other environments, on the other hand, it is used to bring organic substance to the soil or to carry out biofumigation operations. In addition, B. juncea has also been studied for its ability to absorb selenium, sulfur, lead, chromium, copper and zinc.

Indian mustard can be successfully cultivated in sites polluted by heavy metals and offers great potential in the reclamation of these lands through phytoextraction. It presents high concentrations of metal in the above ground portions in relation to lower concentrations in soil, indicating a transport of substances to the foliar apparatus. This species is able to metabolize heavy metals, especially Zn, Pb and Cd, accumulating them in less toxic forms through defense mechanisms (Rathore et al., 2017).

As early as 1995, Nanda Kumar et al. (1995) had identified several crops of Indian mustard that could effectively accumulate Pb and other heavy metals. Also in 1998, the study done by Begonia et al. (1998) reported the behavior of the plant species towards lead. It has been shown that the lead present on the growth soil has not had a significant deleterious effect on the total foliar area per plant. The reduction of the foliar area, in relation to control plants, was around 25%, increasing in relation to the concentration of contaminant in the soil. Mustard buds have accumulated different amounts of lead in relation to the concentration applied in the soil. The data showed that the application of 100, 250 and 500 µg/ml Pb in the growth medium determined the uptake of the Pb bud for 72, 149 and 258 µg/g of dry biomass, respectively. In addition, it is important to note that the translocation of Pb from roots to buds is normally very low.

As for the phytoextraction of lead, the efficiency of the process always remains linked to the low mobility and bioavailability of heavy metal in the soil. In addition, high concentrations of contaminants can inhibit the growth of the plant species or lead to a reduction in the biomass produced. So studies on the phytoextraction of Pb focused on the use of fast-growing crops, as in the case of Brassica Juncea, having good biomass yields. Relatively low mobility of metals in the subsoil can act, as Lim et al. (2004) studies suggest, through the use of EDTA chelating substances that allow increased mobility and thus greater absorption. Obviously the application of EDTA requires an accurate dosage to achieve greater efficiency. The study carried out in the presence of contamination by lead and arsenic also treated the application of an electric field around the plant, obtaining positive results in the reduction, in terms of time, intervention and doubling the repair rate compared to the application of EDTA only.

As mentioned above, in addition to the accumulation of lead, it should be noted how efficient it is in purifying soils from other contaminants such as cadmium or zinc. This allows to open preferential scenarios in which the choice falls on plants that despite having less accumulation can, however, treat multi-contaminate soils. The study of Szczygłowska et al. (2011) highlighted the fact that *B. Juncea* showed an high capacity to accumulate Cd, mainly in buds, reaching a level of over 1400 g of Cd per gram of dry weight. Other studies have also compared the effectiveness of removing zinc from soil in relation to *Thlaspi caerulescens*, a zinc hyperaccumulator. The positive response in the field was due to the fact that *B. Juncea* produces ten times more biomass than *T. Cearullescens* so allow a better remediation.

4.3.4 Plant focused on Zinc accumulation

Zinc is a naturally occurring element in environmental matrices such as air, water and soil, but also within living beings. It plays an essential role in the growth and development of plants thanks to its structural, catalytic and activating properties. Generally, in cultures, the concentrations that allow a correct growth are around 15/20 mg per kg of dry weight. Naturally, the pedogenetic processes of leaching of the mother rocks cause a release of Zinc, along with eruptive phenomena, while a small part is transported by normal erosion processes. Environmental background concentrations are linked to these processes while abnormal amounts of this metal are linked to anthropogenic activities due to industrial sites, mining and activities such as coal combustion, waste and steel processing.

High concentrations can lead to toxicity to fauna, humans and flora, leading to morphological, biochemical and physiological disturbances. The toxic effect of zinc appears to be related to its bio-availability in the soil, obviously function of factors such as pH, microbial community, chemical composition and mineralogical and the organic matter present in the soil. The toxicity of Zn increases with its bio-availability. Despite this, however, some plants have shown survival in soils with high Zn concentrations, developing not only detoxification mechanisms but also accumulation and translocation within the tissues. By 2020, 28 species of zinc hyper-accumulators were identified, most belonging to the family Brassicacee (Balafrej et al., 2020).

Noccaea caerulescens *Noccaea caerulescens*, formerly also called *Thlaspi caerulescens*, is a flowering herbaceous plant that is part of the family of the Cruciferae (Brassicaceae). It is a perennial or biennial plant, and generally presents with rosette leaves with white flowers. The species, however, may have variations in the forms in relation to the environment in which it grows.

Figure 4.5. Thlaspi caerulescens or Noccaea caerulescens species.

This species, besides playing the role of bioaccumulator in ecosystems or in the environments it occupies, is identified as extremophile as it is one of the few to be able to survive on "extreme" soils which are characterised by the natural presence of certain metals such as zinc or cadmium. Tolerance to these metals is much higher than for other metallophytic species and therefore involves a promising scenario for its ability to repair cadmium and/or zinc-polluted soils through extraction and accumulation in its airborne parts.

For the first time, in 1983, Reeves and Brooks (1983) highlighted the unusual absorption of Zinc and Nikel by the species. Going to quantify it, the accumulation amounted to levels higher than 1000 g/g in the dry mass of the plants for Zn and levels of 1000-30,000 g/g, appear on a dry basis, for Ni. This study has opened a new field with respect to the species attracting the attention of other scholars, who have also examine in depth the behavior of the species in relation to multicontaminate soils.

With regard to this, as reported by Marschner (1995), the species shows a high sensitivity to the toxicity of Cu, to be considered when the soil contamination is mixed with this contaminant. Cu, even if it is an essential element for plants, can be potentially toxic if the concentrations are high, so to make an effective phyextotraction a low availability of Cu is advisable. Some studies, such as the one proposed by Keller and Hammer (2004), identify this toxicity in the Iron deficiency for plants, introducing the need for remedial measures such as treatment with Fe, exerted in lower biomass production and lower extractive capacity.

Chapter 5

Conclusion

The mining sector and its associated industry represent, in some countries, a priority and fundamental activity for economic development, despite fluctuations due to changing demand over time and other factors such as productivity and technological innovation. This obviously entails not only economic benefits for the countries affected by this activity but also high environmental liabilities which affect the quality of life in the vicinity of mining sites. The growing awareness of the environment has gradually spread worldwide and even the least progressive states in the sector have started to follow protocols and practices of environmental protection, drafting codes and laws to limit environmental contamination.

Looking at the copper extraction sector in detail, growth is linked to factors such as the growing demand for energy and the evolution in the telecommunications and electronics sectors. If we tie this aspect to the decrease of the degree of the mineral in the soil it is easy to deduce how much the amount of waste material, usually of fine type, turns out to be obtained after the operations of shredding and flotation. This resulting material is transported through piping systems or mechanical transport and stored in large deposits called "tailing dams". They are often the focus of interest as they contain material with high concentrations of heavy metals and therefore require insulation from groundwater, environmental protection and recovery. This must be supported by legislation that protects human health in the vicinity of polluted sites. In Chile, although belatedly, the issue of the environmental sustainability of mining sites was introduced in 1994 by the Environmental Foundation Act (Act No. 19 300) and subsequently updated and extended. This has therefore gradually put the spotlight on soil remediation technologies, bringing to the fore the need to identify contaminated sites and plan correct management, closure and remediation operations.

Phytoremediation was presented as a whole, identifying the mechanisms and strategies of treatment utilised to clean up contaminated sites. The technology identified for the treatment of heavy metals was that of phytoextraction, a technique that allows the accumulation of contaminants in the roots of plants and then the translocation of the same in the foliar apparatus. The elements to be treated, heavy metals, have special characteristics because they persist in the soil without degrading and tend to create complex bonds. In addition, it is important not only to determine the total soil concentration of these compounds, but above all the bioavailable part, as this part can be absorbed by the plants. The species used in phytoextraction must have the characteristics of tolerance, resistance and translocation of the high concentrations present in the soil but it is also desirable that the growth is rapid and with the development of a high biomass. These plant species are classified according to the concentrations absorbed in relation to the concentrations in the soil, and subdivided into excluder, accumulator and hyperaccumulator. For the construction of a plant of phytoextraction it is desirable to use over-accumulating plants or possibly large accumulators with good characteristics in terms of growth and biomass production.

Choosing the right species is an important step and determines whether or not the site's remediation is successful. The decision-making process takes into account several factors and information that have been analyzed in this thesis. Bioconcentration factor (BCF) and translocation factor (TF) criteria shall be used for the evaluation of the intervention. From the comparison with other traditional treatment methods, it is clear that the classic methods of physical reclamation allow to completely remove heavy metals but in addition to having a high cost have a destructive for the environment, whereas chemical remediation methods do not safeguard the environment from the release of intermediate contaminated sites, despite the fact that such treatments are cheap and easy to apply. To this end it is necessary to investigate new methodologies, such as phytoremediation, which is eco-friendly and cost-effective.

The meteorological characterization of the site in question was necessary to define the environmental conditions in which the plants would have to grow and develop. To determine the atmospheric variables, two weather stations in the vicinity of the site were taken into account, extracting the available historical data. First, the temperature data of the two stations were extrapolated. From the analysis of the graphs and the monthly average values shown, the temperature follows the usual annual trend, having average maximum values around 20°C during the month of January and reaching the annual lows during the month of August, down to a maximum of average temperatures around 6°C. In this phase it was also important to determine the icy days as temperatures below zero can cause problems in the management of the plant and in the correct growth of the species.

The analysis of the precipitation, evapotraspiration and humidity variables showed the presence of a dry-arid climate, with rainfall hardly ever exceeding 120 mm/month in the rainiest months. The evapotraspiration, studied through the model of Thornthwaite, has an annual course opposite to that of the precipitations. The share of meteoric water fails to compensate for the values of ETP, except in a few winter months, like June, July and August, in which there is water infiltration. The relative humidity of the air follows a bell pattern throughout the year, more or less related to the trend of precipitation. The highest values of humidity were reached during the winter months, reaching values of over 80% during the month of August.

For plants solar radiation is a source of energy and a stimulus for development as well as a stress factor. It has a great impact on the actual yield of plants, influencing the processes of photosynthesis and transpiration of plants. In the study site, although the daily trend is similar for all seasons, during the summer months it reaches values of over 900 W/m^2 in the daily peak but also in the winter months the values remain relatively good, with values of about 400 W/m^2 during the peaks.

Another parameter such as wind needs attention because high speeds can compromise the construction of plants leading to grubbing from the ground. The Wind Rose, made for the weather station of Punta Cortes, shows that the north is the preferred wind direction with speeds that never exceed 5 m/s. The winds that blow on the site can be classified as Light Breeze or Gentle Breeze and do not present problems for the development of the plant.

The chemical analysis of soil has a high importance in the design of a phytoextraction plant as it establishes the starting point in the determination of contamination and in the subsequent selection of the species to be used. The analysis of the tailing dam Chancon 1 and Chancon 2 were provided by SERNAGEOMIN, the Chilean state body that regulates mining activity and provides annual updates on the state of the landfills of fine material, with the aim of monitoring environmental performance and proper management. As many as 29 chemical components have been classified in the two sites, including high values of elements such as arsenic (As), lead (Pb), cadmium (Cd), copper (Cu), chromium (Cr) and zinc (Zn). In order to carry out a risk analysis, using the global and Swedish guidelines, they have been classified according to their hazardousness and a table has been presented indicating the degree of danger in relation to the concentration present in the soil. By interfacing this information it was possible to identify the degree of contamination in the two dam. From what has emerged, heavy metals such as arsenic (As), lead (Pb) and cadmium (Cd) are the ones giving the greatest concern, with concentrations of 500, 900 and 10 g/t respectively. Other heavy metals such as chromium (Cr) and zinc (Zn) have the same high concentrations but have a lower degree of danger. In the case of copper (Cu), even if the danger is less than the others, the concentrations indicate that the contamination is "Extremely serious".

With the evidence of the most dangerous and important contaminants to be treated, a bibliographical review has been carried out in this matter analyzing both the endogenous and exogenous point of view. In Chile, the studies carried out in this area are modest and there are still shortcomings in research. As of 2006, in Latin America, only 172 species resistant to heavy metals have been classified. The species identified, such as *Oenothera affinis*, are not hyperaccumulator but fall into the category of metallophytes, however, presenting a high accumulation. These species can only be investigated if they are resilient to multi-contamination and have good biomass growth. Other endogenous/native species such as *Atriplex atacamensis* and *Adesmia atacamensis* have shown scientific evidence of Arsenic and Copper accumulation in the territories of the Antofagasta region in northern Chile. In the global context, the need was highlighted to scale the problem according to the climatic and environmental situation, evaluating alternatives provided by the methods assisted with micro-organisms and amendments.

The research on plants used in phytoextraction was carried out by dividing it by contaminants and reporting the species that could be the right candidates for experimental planting. The various species have been described and different scientific research has been analyzed for each one, reporting results and analyzing the boundary conditions to determine the applicability also in the area under study. For arsenic, the species *Pteris vittata L.* and *Atriplex atacamensis* have been identified. The first plant species, in addition to resisting in environments affected by multi-contamination, is an excellent candidate as it has a good development of biomass as well as a high efficiency of accumulation processes and in the translocation of contaminants from roots to shoot apparatus, and has shown its potential in several scientific studies. The species *Atriplex atacamensis*, besides being native to the northern regions of Chile, is able to survive and grow in difficult environmental conditions contaminated by As and other heavy metals.

In addition to the previous species, also *Adesmia atacamensis*, endogenous of Chile, reported good prospects, showing excellent values of accumulation and translocation for copper, but the analysis of the values of BCF has shown that it acts as an exclusor for other elements such as Pb, Fe and Zn. This therefore requires a deepening of the bio-availability of these elements to determine the possible application in the field. The species *Brassica Juncea L.* is not native to America but has been established in the continent for some time. This species, of the family of the Brassicaceae, is very adaptable and resists in places with annual precipitations between 500 and 4200 mm, average annual temperatures between 6 and 27 degrees centigrade and soils with pH between 4.3 and 8.3. *Brassica Juncea L.* has shown effectiveness of accumulation of Zn, Cd and Pb, accumulating them in less toxic forms but showing for very high concentrations of lead some limitations for the growth and the foliar development. As for the main zinc-accumulating plants, the species *Noccaea caerulescens* has been identified; a plant that can survive on soils characterized by the presence of metals such as zinc, cadmium and nickel, accumulating in quantities of more than 1000 g per gram of dry mass. However, it is very sensitive to multi-contamination with the presence of copper.

Based on what has been investigated, the phytoremediation technique, specifically phytoextraction, can be used to clear deposits of waste material from mining operations. The plants identified have shown prospects in the feasibility of decontamination work thanks to studies carried out in similar environments or climatic conditions. As a site-specific technique, a future investigation should be aimed at determining the bioavailable share of the individual elements present in the tailing dam and the study of the individual species in the field, with the realization of an experimental planting. The climatic conditions and the characterization carried out highlighted a warm and temperate climate, with monthly rainfall over 100 mm only in the winter months. The species investigated allow growth and accumulation even in soils with little water, although it will be a factor to be investigated in the on-site study.

Bibliography

Adriano, D.C. (2001). Trace Elements in Terrestrial Environments: Biogeochemistry, Bioavailability and Risks of Metals. *Springer, New York*. DOI: http://dx.doi.org/10.1007/978-0-387-21510-5.

Agua royal spring, Engineering Mining Solution. (2020). Flat Pipe Heap leaching. Retrieved from: https://www.aquaroyalspring.com/ [Accessed: 2.04.2020].

Ayres, R. U., Ayres, L. W., & Råde, I. (2002). The Life Cycle of Copper, its Co-Products and By-Products. *International Institute for Environment and Development*.

An, Z., Huang, Z., Lei, M., Liao, X., Zheng, Y., Chen, T. (2006). Zinc tolerance and accumulation in Pteris vittata L. and its potential for phytoremediation of Zn- and As-contaminated soil. *Chemosphere, 62*, (5), 796-802. DOI: https://doi.org/10.1016/j.chemosphere.2005.04.084.

Agency for Toxic Substances and Disease Registry (ATSDR). 2004. Toxicological profile for Copper. *Atlanta, GA: U.S. Department of Health and Human Services, Public Health Service.*

Aurubis – a leading worldwide provider of non-ferrous metals. (2020). Copper – a unique metal. Retrieved from: https://www.aurubis.com/en/about-aurubis [Accessed: 22.05.2020].

Baker, A. & Brooks, R.. (1989). Terrestrial Higher Plants Which Hyperaccumulate Metallic Elements, A Review of Their Distribution, *E. Ecol. Phytochem. Biorecovery, 1.* DOI:

Baker, A.J.M., Proctor, J. (1990). The influence of cadmium, copper, lead, and zinc on the distribution and evolution of metallophytes in the British Isles. *Pl Syst Evol, 173*, 91–108. DOI: https://doi.org/10.1007/BF00937765.

Balafrej, H., Bogusz, D., Triqui, Z. E. A., Guedira, A., Bendaou, N., Smouni, A. & Fahr, M. (2020). Zinc Hyperaccumulation in Plants: A Review. *Plants, 9*(5), 562.

Barceló, J. & Poschenrieder, C. (2003). Phytoremediation: Principles and perspectives. *Contributions to Science, ISSN 1575-6343, 2*,(3), 333-344.

: Begonia, G. B., Davis, C. D., Begonia, M. F. T., & Gray, C. N. (1998). Growth Responses of Indian Mustard [Brassica juncea (L.) Czern.] and Its Phytoextraction of Lead from a Contaminated Soil. *Department of Biology, Jackson State University, Jackson, USA. Bull. Environ. Contam. Toxicol.*

Bhanbhro, Riaz. (2014). Mechanical Properties of Tailings: Basic Description of a Tailings Material from Sweden. DOI: https://doi.org/10.13140/2.1.1338.2082.

Bondada, B.R., Ma, L.Q. (2003) Tolerance of Heavy Metals in Vascular Plants: Arsenic Hyperaccumulation by Chinese Brake Fern (Pteris Vittata L.). *Chandra S., Srivastava M. (eds) Pteridology in the New Millennium. Springer*. DOI: https://doi.org/10.1007/978-94-017-2811-9_28.

Chen, T., Wei, C., Huang, Z., Huang, Q., Lu, Q. & Fan Z. (2002). Arsenic hyperaccumulator Pteris Vittata L. and its arsenic accumulation. *Chin. Sci. Bull. 47*, 902–905. DOI: https://doi.org/10.1360/02tb9202.

COCHILCO, Chilean Copper Commission. (2017). Chilean Copper Mining Cost. Retrieved from: https://www.cochilco.cl/Presentaciones%20Ingls/Chilean%20Copper%20Mining%20Costs.pdf [Accessed: 28.06.2020].

Copper Alliance & The Association for Science Education. (2020). Copper Mining and Extraction. Retrieved from: https://copperalliance.eu/knowledge-base/education-resources/ [Accessed: 28.03.2020].

Crawford, C.B. & Quinn, Brian. (2017). Microplastic Pollutants.

Cristaldi, A., Gea, O. C., Jho, E. H., Zuccarello, P., Grasso, A., Copat, C., & Ferrante, M. (2017). Phytoremediation of contaminated soils by heavy metals and PAHs. A brief review. *EnvironmentalTechnology&Innovation*. 8. 309-326. DOI: https://doi.org/10.1016/j.eti.2017.08.002.

Danh, L. T., Truong, P., Mammucari, R. & Foster N. (2014). A Critical Review of the Arsenic Uptake Mechanisms and Phytoremediation Potential of Pteris vittata. *International Journal of Phytoremediation, 16*(5), 429-453. DOI: https://doi.org/10.1080/15226514.2013.798613.

Diaz, O., Tapia, Y., Pastene Olivares, R., Montes, S., Núñez, N., Vélez, D. & Montoro, R. (2011). Total and Bioavailable Arsenic Concentration in Arid Soils and its Uptake by Native Plants from the Pre-Andean Zones in Chile. *Bulletin of environmental contamination and toxicology, 86*, 666-9. DOI: https://doi.org/10.1007/s00128-011-0269-0.

Dobson, M.C. & Moffat, A.J. (1995). A Re-Evaluation of Objections To Tree Planting On Containment Landfills. *Waste Management and Research, ISWA, 13*(6), 579-600. DOI: https://doi.org/10.1177/0734242X9501300607.

U.S. Earth Explorer (2020). Retrieved from: https://earthexplorer.usgs.gov/ [Accessed: 5.04.2020].

Encyclopedia Britannica (2020). Roasting, smelting, and converting. Retrieved from: https://www.britannica.com/technology/copper-processing/Roasting-smelting-and-converting [Accessed: 1.04.2020].

Evanko C. & Dzombak D.A. (1997). Remediation of metals-contamined soils and groundwater. *GWRTAC (Ground Water Remediation Technologies Analysis Center)*

Evans G.M. & Furlong J.C. (2003). Contaminated land and bioremediation. *Environmental Biotechnology: theory and application, John Wiley Sons Ltd*, 89–111.

Evangelou, M.W.H., Ebel, M., Schaeffer, A. (2007). Chelate assisted phytoextraction of heavy metals from soil. Effect, mechanism, toxicity, and fate of chelating agents, *Chemosphere, 68*(6), 989-1003. DOI: https://doi.org/10.1016/j.chemosphere.2007.01.062.

Favas, P., Pratas, J., Varun, M., D'Souza, R., & Paul, M. S. (2014). Phytoremediation of Soils Contaminated with Metals and Metalloids at Mining Areas: Potential of Native Flora. *Environmental Risk Assessment of Soil Contamination*. 10.5772/57469.

Famulari, S. & Witz, K. (2015). A User-Friendly Phytoremediation Database: Creating the Searchable Database, the Users, and the Broader Implications. *International journal of phytoremediation, 17*, 737-744. DOI: https://dx.doi.org/10.1080/15226514.2014.987369.

Fernández, Y. T., Diaz, O., Acuna, E., Casanova, M., Salazar, O., Masaguer, A. (2016). Phytostabilization of arsenic in soils with plants of the genus Atriplex established in situ in the Atacama Desert. *Environmental monitoring and assessment, 188*(4), 235. DOI: https://dx.doi.org/10.1007/s10661-016-5247-x.

FISA, GBR. (2018). Chile Mining 2018: EXPOMIN's Official Investment Guide. Retrived from https://www.gbreports.com/files/pdf/_2018/Chile_Mining_2018_-_Book_Preview.pdf [Accessed: 22.06.2020].

Garside, M. (2020). Global copper reserves by country 2019. Retrieved from: https://www.statista.com/statistics/273637/copper-reserves-by-country/. [Accessed: 21.05.2020].

Ginocchio, R. & Baker, A. J. M. (2004). Metallophytes in Latin America: a remarkable biological and genetic resource scarcely known and studied in the region. *Revista chilena de historia natural, 77*(1), 185-194. DOI: https://dx.doi.org/10.4067/S0716-078X2004000100014.

Gleeson, D. (2020). Metso books comminution equipment order from Codelco at El Teniente. *im-mining.com*. Retrieved from: https://im-mining.com/2019/04/26/metso-books-comminution-equipment-order-codelco-el-teniente/ [Accessed: 28.03.2020].

Gonzalez, I., Muena, V., Cisternas, M., Neaman, A. (2008). Acumulación de cobre en una comunidad vegetal afectada por contaminación minera en el valle de Puchuncaví, Chile central. *Revista chilena de historia natural, 81*(2), 279-291. DOI: https://dx.doi.org/10.4067/S0716-078X2008000200010

Google Maps. (2020). Retrieved from: www.googlemaps.com. [Accessed: 1.05.2020].

Hazrat A., Ezzat K. & Muhammad A. S. (2013). Phytoremediation of heavy metals — Concepts and applications. *Chemosphere, 91*(7), 869-881. DOI: https://doi.org/10.1016/j.chemosphere.2013.01.075.

Hinchman, R. R., Negri, M. C. & Gatliff, E. G. (1995). Phytoremediation: using green plants to clean up contaminated soil, groundwater, and wastewater. *Argonne National Laboratory Hinchman, Applied Natural Sciences, Inc.* Retrieved from: http://www.treemediation.com/Technical/Phytoremediation1998.pdf.

Interstate Technology and Regulatory Council (ITRC). (2001). Phytotechnology Technical and Regulatory Guidance Document.

Jamasmie, C. (2020). This is what the world's largest underground mine looks like. *Mining.com*. Retrieved from: https://www.mining.com/chiles-codelco-boost-production-el-teniente-copper-mine/ [Accessed: 27.03.2020].

John, D.A. & Leventhal, J.S. (1995). Bioavailibility of Metals in Preliminary compilation of descriptive geoenvironmental mineral deposit models. *U.S. Department Of The Interior, U.S. Geological Survey, Edward A. du Bray Editor.*

Keller, C. & Hammer, D. (2004). Metal availability and soil toxicity after repeated croppings of Thlaspi caerulescens in metal contaminated soils. *Environmental Pollution, 131*(2), 243-254. DOI: https://doi.org/10.1016/j.envpol.2004.02.030.

Khalid, S., Shahid, M., Niazi, N. K., Murtaza, B., Bibi, I., Dumat, C. (2017). A comparison of technologies for remediation of heavy metal contaminated soils. *Journal of Geochemical Exploration, 182*(B), 247-268. DOI: https://doi.org/10.1016/j.gexplo.2016.11.021.

Lam Esquenazi, E., Keith Norambuena, B., Montofré, I. & Galvez, M. (2018). Copper Uptake by Adesmia atacamensis in a Mine Tailing in an Arid Environment. *Air, Soil and Water Research.* DOI: https://doi.org/10.1177/1178622118812462.

Lam Esquenazi, E., Keith Norambuena, B., Montofré, I. & Galvez, M. (2018). Evaluation of soil intervention values in mine tailings in northern Chile. *PeerJ, 6.* DOI: https://doi.org/10.7717/peerj.5879.

Landmeyer, J. (2011). Introduction to Phytoremediation of Contaminated Groundwater: Historical foundation, hydrologic control, and contaminant remediation. *Springer Science & Business Media.* Retrieved from: https://books.google.it/books?id=Yca0hVWTdv0C.

Leonida C. (2020). Froth Flotation for the 21st Century. *Engineering and Mining journal.* Retrieved from: www.e-mj.com/features/froth-flotation-for-the-21st-century/ [Accessed: 28.03.2020].

XY Liao, XY Xiao & TB Chen. (2003). Effects of Ca and As addition on As, P and Ca uptake by hyperaccumulator Pteris vittata L. under sand culture. *Acta Ecologica Sinica, 23*(10), 2057-2085.

Lim, J., Salido, A. L., Butcher, D. J. (2004). Phytoremediation of lead using Indian mustard (Brassica juncea) with EDTA and electrodics. *Microchemical Journal, 76*(1–2), 3-9. DOI: https://doi.org/10.1016/j.microc.2003.10.002.

Liphadzi, M.S. , Kirkham, M.B. & Musil, C.F. (2005). Phytoremediation of soil contaminated with heavy metals: a technology for rehabilitation of the environment. *South African Journal of Botany, 71*(1), 24-37. DOI: https://doi.org/10.1016/S0254-6299(15)30145-9.

Liu, D., Jiang, W., Liu, C., Xin, C. & Hou, W. (2000). Uptake and accumulation of lead by roots, hypocotyls and shoots of Indian mustard [Brassica juncea (L.)]. *Bioresource Technology, 71*,(3), 273-277. DOI: https://doi.org/10.1016/S0960-8524(99)00082-6.

Loch Arellano, B. (2017). Evaluación del uso de atriplex nummularia con la aplicación de enmiendas húmicas para la fitoestabilización de relaves mineros. Retrived from: http://repositorio.uchile.cl/handle/2250/146093 [Accessed: 19.06.2020].

López, P., Ainzúa, S., Zolezzi, C., Vasconi, P. (2003). La Minería y su Pasivo Ambiental. *Fundación Terram, Series APP, 23.*

Ma, L. Q., Komar, K., Tu, C., Zhang, W., Cai, Y. & Kennelley, E. D. (2001). A fern that hyperaccumulates arsenic. *Nature, 409*, 579. DOI: https://doi.org/10.1038/35054664.

Macnair, M. (1993). Tansley Review No. 49. The Genetics of Metal Tolerance in Vascular Plants. *The New Phytologist, 124*(4), 541-559. Retrieved from: www.jstor.org/stable/2557972 [Accessed: 26.05.2020].

Marschner, H. (1995). Mineral nutrition of higher plants 2nd edition. *Academic, Great Britain.*

Mccutcheon, Steven & Schnoor, Jerald. (2004). Phytoremediation Transformation and Control of Contaminants. *Environmental Science and Pollution Research, 11* (1). DOI: https://doi.org/10.1007/BF02980279.

Minello, F.(2010). Tesi di Dottorato - Fitorisanamento di suoli contaminati da metalli pesanti e metalloidi. *Università Ca' Foscari Venezia Dottorato di Ricerca in Scienze Ambientali, Scuola di Dottorato in Scienze e Tecnologie.*

Ministerio de Minería - División de Desarrollo Sostenible. (2019). Plan Nacional de Depósitos de Relaves para una Minería Sostenible

Molland, A., Turnock, S. & Hudson, D. (2017). Components of Hull Resistance. *In Ship Resistance and Propulsion: Practical Estimation of Ship Propulsive Power*, 12-69. Cambridge: Cambridge University Press. DOI: https://doi.org/10.1017/9781316494196.005.

Muszyńska, E. & Labudda, M. (2019). Dual Role of Metallic Trace Elements in Stress Biology—From Negative to Beneficial Impact on Plants. *International Journal of Molecular Sciences, 20*(13), 3117. DOI: https://doi.org/10.3390/ijms20133117.

Nanda Kumar, P. B. A., Dushenkov, V., Motto, H., & Raskin, I. (1995). Phytoextraction: The Use of Plants To Remove Heavy Metals from Soils. *Environmental Science Technology, 29*(5), 1232-1238. DOI: https://doi.org/10.1021/es00005a014.

Napoli, M., Cecchi, S., Grassi, C., Baldi, A., Zanchi, C.A., Orlandini, S. (2019). Phytoextraction of copper from a contaminated soil using arable and vegetable crops. *Chemosphere, 219*, 122-129. DOI: https://doi.org/10.1016/j.chemosphere.2018.12.017.

NRCan. (November 27, 2019). Distribution of copper consumption worldwide in 2018, by end use. *Statista*. Retrieved from: https://www.statista.com/statistics/605463/distribution-of-copper-consumption-worldwide-by-end-use/ [Accessed: 21.05.2020].

Ogundiran, M.B. & Osibanjo, O. (2009). Mobility and speciation of heavy metals in soils impacted by hazardous waste. *Chemical Speciation & Bioavailability, 21*(2), 59-69. DOI:https://doi.org/10.3184/095422909X449481.

Pandey, B., Kinrade, S. D., Catalan, L. J. (2012). Effects of carbonation on the leachability and compressive strength of cement-solidified and geopolymer-solidified synthetic metal wastes. *Journal of environmental management, 101*, 59–67. DOI: https://doi.org/10.1016/j.jenvman.2012.01.029.

Pascual-Ferrer, J. & Candela, L. L. (2015). Water balance on the Central Rift Valley - Appendix 1. Retrived from: http://hdl.handle.net/2117/89152.

Peijnenburg, W.J.G.M., Zablotskaja, M., Vijver, M.G. (2007). Monitoring metals in terrestrial environments within a bioavailability framework and a focus on soil extraction. *Ecotoxicology and Environmental Safety, 67*(2), 163-179. DOI: https://doi.org/10.1016/j.ecoenv.2007.02.008.

Pietrelli, L., Ciferri, F., Giancanelli, V., Menegoni, P., Farneti, T., Modestia, F. & Salluzzo, A. (2004). Accumulo di metalli pesanti in piante erbacee spontanee. *AcquaAria, 9*, 24–29.

Plette, A.C.C., Nederlof, M.M., Temminghoff, E.J.M. & van Riemsdijk, W.H. (1999). Bioavailability of heavy metals in terrestrial and acquatic system: a quantitative approach. *Environmental Toxicology and Chemistry*,18, 1882–1890. DOI: https://doi.org/10.1002/etc.5620180903.

Radulescu, C., Stihi, C., Barbes, L., Chilian, A. & Chelarescu, E. (2013). Studies Concerning Heavy Metals Accumulation of Carduus nutans L. and Taraxacum officinale as Potential Soil Bioindicator Species. *Revista de Chimie -Bucharest- Original Edition-, 64*, 754-760.

Rajesh k., Rohit, M., Vani, M., Afifa, Q., Anand, P., Shashi, S., Madhu, P., Ashutosh, P. & Anupam, D. (2016). Detoxification and Tolerance of Heavy Metals in Plants. DOI: https://doi.org/10.1016/B978-0-12-803158-2.00013-8.

Rathore, S., Shekhawat, K., Dass, A., Kandpal, B. & Singh, V. (2017). Phytoremediation Mechanism in Indian Mustard (Brassica juncea) and Its Enhancement Through Agronomic Interventions. *Proceedings of the National Academy of Sciences, India Section B: Biological Sciences, 89*. DOI: https://doi.org/10.1007/s40011-017-0885-5.

Reeves, R.D. & Brooks, R.R. (1983). European species of Thlaspi L. (Cruciferae) as indicators of nickel and zinc. *Journal of Geochemical Exploration, 18*(3), 275-283. DOI: https://doi.org/10.1016/0375-6742(83)90073-0.

Reeves, R.D., Baker, A.J.M., Jaffré, T., Erskine, P.D., Echevarria, G. & van der Ent, A. (2018), A global database for plants that hyperaccumulate metal and metalloid trace elements. *New Phytol, 218*, 407-411. DOI: https://doi.org/10.1111/nph.14907.

SERNAGEOMIN, per Servicio Nacional de Geología y Minería. (2020). Geochemistry of surface samples of tailings deposits in Chile. https://www.sernageomin.cl/datos-publicos-deposito-de-relaves/

Sharma, P. & Pandey, S. (2014). Status of phytoremediation in world scenario. *International Journal of Environmental Bioremediation and Biodegradation, 2*, 178-191. DOI: https://doi.org/10.12691/ijebb-2-4-5.

Spitz, K., & Trudinger, J. (2009). Mining and the environment: From ore to metal. *Boca Raton: CRC Press.*

Subhashini, V., & Swamy, A. (2013). Subhashini and Swamy Phytoremediation of Pb and Ni Contaminated Soils Using Catharanthus roseus (L .). *Universal Journal of Environmental Research and Technology, 3*, 465-472.

Swedish Environmetal Protection Agency. (2002). Methods for Inventories of Contaminated Sites, Environmental Quality Criteria, Guidance for data collection. Report 5053

Szczygłowska, M., Piekarska, A., Konieczka, P. & Namieśnik, J. (2011). Use of brassica plants in the phytoremediation and biofumigation processes. *International journal of molecular sciences, 12*(11), 7760–7771. DOI: https://doi.org/10.3390/ijms12117760.

Szymanowski, J. (1996). Copper hydrometallurgy and extraction from chloride media. *Journal of Radioanalytical and Nuclear Chemistry.*

Tapia, Y., Diaz, O., Pizarro, C., Segura, R., Vines, M., Zúñiga, G. & Moreno-Jiménez, E. (2013). Atriplex atacamensis and Atriplex halimus resist As contamination in Pre-Andean soils (northern Chile). *Science of The Total Environment, 450–451*, 188-196. DOI: https://doi.org/10.1016/j.scitotenv.2013.02.021.

Teegavarapu, Ramesh S. V. (2012). Hydrologic modeling and design. *Floods in a Changing Climate: Extreme Precipitation, International Hydrology Series*, 225–240. DOI: 10.1017/CBO9781139088442.010.

Toledo, T. Y. (2016). Situacion de los pasivos ambientale - Mineros en Chile. El caso de los depositos de relaves. *Fundacion Terram*. Retrived from: www.terram.cl [Accessed: 24.06.2020].

U.S. Congress, Office of Technology Assessment. (1988). Copper: Technology and Competitiveness.

U.S. Environmental Protection Agency. (1994). Extraction and beneficiation of ores and minerals, vol. 4.

National Risk Management Research Laboratory and Office of Research and Development, U.S. Environmental Protection Agency. (2000). Introduction to Phytoremediation.

US Geological Survey. (February 7, 2020). Total copper mine production worldwide from 2006 to 2019 (in 1,000 metric tons). *Statista*. Retrieved from: https://www.statista.com/statistics/254839/copper-production-by-country/ [Accessed: 21.05.2020].

US Geological Survey. (February 7, 2020). Global copper reserves as of 2019, by country (in million metric tons). *Statista*. Retrieved from: https://www.statista.com/statistics/273637/copper-reserves-by-country/ [Accessed: 21.05.2020].

VisMet portal, Visulisador meteorologico. (2020). Retrieved from: http://vismet.cr2.cl/ [Accessed: 30.04.2020].

Vromman, D., Paternostre, B., Briceño, M., Teixeira-Cardoso, C., Flores-Bavestrello, A., Vanhecke, N., Kumar, M., Martínez, J. & Lutts, S. (2016). Arsenic Distribution in Shoots of the Halophyte Plant Species Atriplex atacamensis Growing in an Extreme Arid Mining Area from Northern Chile. *J Biorem Biodegrad, 7*(3). DOI: https://doi.org/10.4172/2155-6199.1000349.

Weather Atlas, Global Weather and Climate data. (2020). Retrieved from: www.weather-atlas.com/en/chile/rancagua-climate [Accessed: 10.05.2020].

Jacques V. Wirtez. (1999). Mining and metallurgical waste management in the chilean copper industry. *Departamento de Ingenieria de Minas, Universidad de Chile.*

Wójcik, M., Gonnelli, C., Selvi, F., Dresler, S., Rostański, A., Vangronsveld, J. (2017). Chapter One - Metallophytes of Serpentine and Calamine Soils – Their Unique Ecophysiology and Potential for Phytoremediation. *Advances in Botanical Research, 83*, 1-42. DOI: https://doi.org/10.1016/bs.abr.2016.12.002.

Yao, Z., Li, J., Xie, H., Yu, C. (2012). Review on Remediation Technologies of Soil Contaminated by Heavy Metals. *Procedia Environmental Sciences, 16*, 722-729. DOI: https://doi.org/10.1016/j.proenv.2012.10.099.

Yilmaz, E. (2011). Advances in reducing large volumes of environmentally harmful mine waste rocks and tailings. *Department of Applied Sciences, University of Québec at Abitibi-Temiscamingue (UQAT)*. Retrieved from: https://min-pan.krakow.pl/wp-content/uploads/sites/4/2017/12/yilmaz.pdf [Accessed: 20.04.2020].

Zacchini, M., Pietrini, F., Mugnozza, G., Iori, V., Pietrosanti, L. & Massacci, A. (2008). Metal Tolerance, Accumulation and Translocation in Poplar and Willow Clones Treated with Cadmium in Hydroponics. *Water Air and Soil Pollution, 197*, 23-34. DOI: https://doi.org/10.1007/s11270-008-9788-7.

Zhu, L., Ding, W., Feng, L., Kong, J., Xu, Y., Xu, J., Yang, X. (2012). Isolation of aerobic denitrifiers and characterization for their potential application in the bioremediation of oligotrophic ecosystem. *Bioresource Technology, 108*, 1-7. DOI: https://doi.org/10.1016/j.biortech.2011.12.033.