

**CONTRIBUTION TO THE IDENTIFICATION OF  
SOLUTION FOR THE REMEDIATION OF SALT-  
AFFECTED SOILS OF THE HIGH VALLEY OF  
COCHABAMBA IN BOLIVIA: USE OF GYPSUM,  
SULPHUR AND *SUAEDA FOLIOSA***

**CLAIRE DE FROIDMONT**

**TRAVAIL DE FIN D'ÉTUDES PRÉSENTÉ EN VUE DE L'OBTENTION DU DIPLÔME DE  
MASTER BIOINGÉNIEUR EN SCIENCES ET TECHNOLOGIES DE L'ENVIRONNEMENT**

**ANNÉE ACADÉMIQUE 2017-2018**

**PROMOTEUR: GILLES COLINET**

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## Abstract

Salt-affected soils can be found throughout the continents and are a major issue for agriculture. In order to evaluate the potential of rehabilitation of saline-sodic soils in the High Valley of Cochabamba in Bolivia, three different remediation techniques were tested: two by chemical amendments, gypsum and sulphur, and one by phytoremediation with the native halophyte *Suaeda foliosa*. An experiment was carried out in pots containing saline-sodic soil in a greenhouse to evaluate the effect of these management techniques on the saline properties of the soil. Two different doses of the gypsum requirement (GR) and sulphur requirement (SR) were tested, 100% of the GR/SR and 50% of the GR/SR. As for the phytoremediation treatment plants of 1-2 months and of 3-4 months were put to trial. Treatments with chemical amendments managed to decrease Exchangeable Sodium Percentage (ESP) from 70% down to 27%, 43%, 52% and 58% for the 100% - GR, 50% - GR, 100% - SR and 50% - SR respectively. For all the chemical treatments, the electrical conductivity (EC) was reduced by more than 50% and the pH was significantly reduced. Even though, EC, pH and ESP was reduced after the chemical amendment treatments, they are still above the threshold values for the soil to be characterised as non-saline-sodic. The results for the phytoremediation treatments weren't as promising with increased values of ESP and EC. A longer-term experiment should be carried out to continue the evaluation of the reclamation by chemical amendments and the phytodesalination capacity of *Suaeda foliosa*.

## Résumé

Les sols affectés par le sel peuvent être trouvés sur tous les continents et constituent un problème majeur pour l'agriculture. Afin d'évaluer le potentiel de réhabilitation des sols salins-sodiques dans la Haute Vallée de Cochabamba en Bolivie, trois techniques d'assainissement différentes ont été testées: deux par amendements chimiques, gypse et soufre, et une par phytoremédiation avec l'halophyte natif *Suaeda foliosa*. Une expérience a été réalisée sous serre dans des pots contenant un sol salin-sodique afin d'évaluer l'effet de ces techniques de réhabilitation sur les propriétés salines du sol. Deux doses différentes du besoin en gypse (GR) et du besoin en soufre (SR) ont été testées, 100% du GR / SR et 50% du GR / SR. En ce qui concerne le traitement par phytoremédiation, des plantes de 1 à 2 mois et de 3 à 4 mois ont été testées. Les traitements avec des amendements chimiques ont permis de réduire le pourcentage de sodium échangeable (PSE) de 70% à 27%, 43%, 52% et 58% pour les doses 100% - GR, 50% - GR, 100% - SR et 50% - SR respectivement. Pour tous les traitements chimiques, la conductivité électrique (CE) a été réduite de plus de 50% et le pH a été significativement réduit. Même si la CE, le pH et le PSE ont été réduits après les traitements avec les amendements chimique, ils sont toujours supérieurs aux valeurs seuils pour qualifier le sol de non salin-sodique. Les résultats des traitements de phytoremédiation avec des valeurs accrues de PSE et de CE sont eux moins prometteurs. Une expérience à plus long terme devrait être menée pour poursuivre l'évaluation de la réhabilitation des sols par des amendements chimiques et la capacité de phytodésalinisation de *Suaeda foliosa*.

## List of abbreviations

BD	Bulk Density
CEC	Cation Exchange Capacity
D	Depth
EC	Electrical Conductivity
EC <sub>e</sub>	Electrical conductivity of the saturated paste extract
ESP	Exchangeable Sodium Percentage
F	Calcium-Sodium exchange efficiency factor
GR	Gypsum Requirement
LR	Leaching Requirement
NRCS	Natural Resources Conservation Service
SAR	Sodium Adsorption Ratio
SR	Sulphur Requirement
USDA	United States Department of Agriculture
USSL	United States Salinity Laboratory

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# 1. Introduction

Salt affected soils can be found throughout different continents covering almost 10% of earth's land area, or 954 million of hectares (FAO, 1992). South America, North and Central Asia and Australasia are the most impacted, with 75% of the salt affected lands being spread out over these continents (Pessarakli & Szabolcs, 1999). This degradation process mainly occurs in arid and semi-arid regions, where precipitations are scarce and therefore limit the leaching of the salts to the groundwater and back to the ocean through streams. This is one of the biggest issues regarding crop production (Richards *et al.*, 1954, Rengasamy 2010).

Salt-affected soils can be described as soils having an excessive amount of ions such as potassium ( $K^+$ ), magnesium ( $Mg^{2+}$ ), calcium ( $Ca^{2+}$ ), chloride ( $Cl^-$ ), sulphate ( $SO_4^{2-}$ ), carbonate ( $CO_3^{2-}$ ), bicarbonate ( $HCO_3^-$ ) or sodium ( $Na^+$ ). Salt accumulation is a major environmental issue due to its impacts on the ecosystems. The destruction of the soil structure, deterioration of hydraulic properties, imbalances in plant nutrition are some of the consequences of excessive salt content in soils. (Richards *et al.*, 1954, Bresler *et al.*, 1982)

This type of soil degradation can firstly result from a high salt concentration in the parental material, such as marine deposits from previous geologic periods, or the rise of the water table (primary salinity). Secondly, human intervention such as inappropriate irrigation techniques used by farmers often worsen these poor soil conditions (secondary salinity) (Qadir & Schubert, 2002).

Classical solutions to this problem are the application of amendments and leaching of excess salts. New techniques include phytoremediation with salt accumulating plants.

A total salt-affected area of 5 949 thousand hectares was accounted for in Bolivia representing about 5.5% of the country's total area (Massoud, 1977). This is a big issue in the high valley of Cochabamba where the area of farmland has decreased because of the presence of excess salts. We hypothesized that in the region of Cochabamba (Bolivia), phytoremediation is a promising solution to improve the soil condition while being environmentally friendly compared to the use of chemical amendments.

The experimental part of this study was realised at the faculty of Agronomical and Animal Sciences of the Universidad Mayor de San Simon, Cochabamba, Bolivia. The aim was to test and compare different solutions, chemical amendments and phytoremediation in controlled conditions, as a means to remediate salt-affected soils. The soils were sampled from the high valley of Cochabamba. They are representative of the main salt-affected soil types of the region, as described by Weber (2018).

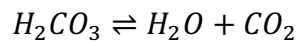
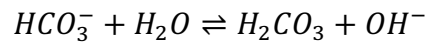
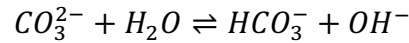
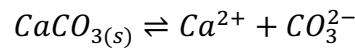
## 2. State of the art

### 2.1 Description of salt affected soils

#### 2.1.1 General information

Different types of salt-affected soils exist, saline, saline-sodic and sodic soils with or without alkalisation and can be characterised as such:

Sodicity can be described as the presence of fixed sodium on the exchange complex of the soil, whereas salinity is characterised by a high concentration of salts, commonly the cations calcium ( $\text{Ca}^{2+}$ ), magnesium ( $\text{Mg}^{2+}$ ) and sodium ( $\text{Na}^+$ ) and the anions chloride ( $\text{Cl}^-$ ) and sulfate ( $\text{SO}_4^{2-}$ ), in the soil solution. Less frequently a high concentration of potassium ( $\text{K}^+$ ), and nitrate ( $\text{NO}_3^-$ ) can also characterise a saline soil. A third process can also be mentioned: alkalisation which can occur in saline and sodic soil. These soils typically have a high pH due to the high concentration of carbonates ( $\text{CO}_3^{2-}$ ) and bicarbonates ( $\text{HCO}_3^-$ ) (Richards *et al.*, 1954, Scianna, 2002, Legros, 2007, Weil & Brady, 2017). The process of alkalisation can be explained by the following equations:



The carbon dioxide found in the soil solution can either come from the atmosphere or biological activities. These reactions more often occur due to the alteration of the parent material containing carbonated minerals (Weil & Brady, 2017).

#### 2.1.2 Assessment of salinity and sodicity

##### A. Sodium Adsorption Ratio (SAR) and Exchangeable Sodium Percentage (ESP)

Soil sodicity is usually assessed through the sodium adsorption ratio (SAR) or the exchangeable sodium percentage (ESP) and soil salinity through the electrical conductivity (ECe).

SAR is defined as follows:

$$SAR = \frac{[Na^+]}{\sqrt{\frac{[Ca^{2+}] + [Mg^{2+}]}{2}}}$$

where the concentrations of the cations are measured in mmol<sub>c</sub>/L

To determine the ESP, the cation exchange capacity (CEC), describing the soil's capacity to withhold cations, has to be previously calculated.

ESP can be calculated with the following formula:

$$ESP = \frac{Exchangeable_{Na}}{CEC} * 100$$

where both the CEC and the exchangeable sodium are expressed in mmol<sub>c</sub>/kg.

However, to determine ESP it has been discussed that it can be better to use the sum of only the main exchangeable cations (Calcium, Magnesium, Potassium and Sodium), called the effective CEC instead of the CEC (Hazelton & Murphy, 2007).

SAR and ESP both express the amount of sodium present but are not identical. ESP is often used to determine the amount of gypsum needed to remediate an excess sodium problem. As for SAR, it is used to characterise the presence of sodium in the irrigation water and the soil solution. (Horneck *et al.*, 2007)

The following equations, presented in table 1, defining the relationship between SAR and ESP can be found in scientific literature (Qadir & Schubert, 2002).

Table 1: Various relationships between ESP and SAR of soils. (Qadir & Schubert, 2002)

Equation	Number of soil samples	Reference
$ESP = [100 (-0.0126 + 0.01475 SAR)] / [1 + (-0.0126 + 0.01475 SAR)]$	59	US Salinity Laboratory (1954)
$ESP = [100 (0.0063 + 0.0124 SAR)] / [1 + (0.0063 + 0.0124 SAR)]$	15	Franklin and Schmehl (1973)
$ESP = [100 (0.1149 + 0.0109 SAR)] / [1 + (0.1149 + 0.0109 SAR)]$	150	Paliwal and Gandhi (1976)
$ESP = (1.95 SAR_{1:5}) + 1.8$	138	Rengasamy <i>et al.</i> (1984)
$ESP = [100 (-0.0867 + 0.02018 SAR)] / [1 + (-0.0867 + 0.02018 SAR)]$	180	Ghafoor <i>et al.</i> (1988)
$ESP = [100 (-0.0268 + 0.02588 SAR)] / [1 + (-0.0268 + 0.02588 SAR)]$	144	Ghafoor <i>et al.</i> (1988)

ESP, Exchangeable sodium percentage; SAR, Sodium adsorption ratio of saturated soil paste extract; SAR<sub>1:5</sub>, Sodium adsorption ratio of soil to water extract of 1:5.

## B. Electrical conductivity

The electrical conductivity measures the ability of a soil to conduct a current. Since water isn't a good conductor compared to soluble salts, there is a clear positive correlation between concentration of soluble salts in the soil solution and the electrical conductivity (El Oumri & Vieillefon, 1983). The electrical conductivity is usually measured in decisiemens per meter (dS/m) or microsiemens per cm ( $\mu\text{S}/\text{cm}$ ). It is usually determined either with the saturated paste extract method (ECe) or the 1:1, 1:2 or 1:5 soil to water extract method (EC). The main difference between the two methods is that for the first one, natural soil saturated conditions are tentatively simulated whereas in the second one not. For this reason, the saturated paste extract method is usually preferred, although reliable and precise results can also be obtained through the 1:1 soil to water extract (Zhang *et al.*, 2005). Several relationships between the ECs found with soil to water extract and the saturated paste extract can be found in the scientific literature (Richards *et al.*, 1954, Zhang *et al.*, 2005, He *et al.*, 2013).

### 2.1.3 Classification of salt affected soils

For a rehabilitation purpose, salt affected soils can be divided into three main categories according to the Natural Resources Conservation Service (NRCS) of the United States Department of Agriculture: saline, saline-sodic or sodic (Richards *et al.*, 1954).

Typically, in a saline soil, the predominant cations are calcium and magnesium in the soil solution. A saline soil would have an electrical conductivity of the saturated paste extract over 4 dS/m, a Sodium Adsorption Ratio (SAR) below 13 and an Exchangeable Sodium Percentage below (ESP) 15%. For sodic soils a high concentration in exchangeable sodium can be found, typically over 15%, a SAR above 13 and an ECe below 4 dS/m. As for saline-sodic soils, SAR, ESP and ECe are high confirming a high level of both sodium and other salts. Table 2 shows the thresholds for ECe, SAR, ESP and the typical soil structure corresponding to the salt-affected soils categories provided by the NRCS.

Furthermore, a visual indicator can also help to categorise the soil; saline soils have a visible deposit of salt on the surface, contributing to a whiten coloured soil. Whereas, sodic soils are usually black due to the dispersion of organic matter resulting of the destruction of the soil's structure. As for saline-sodic soils, they will tend to have a greyish colour (Horneck *et al.*, 2007, Sonon *et al.*, 2015).

Table 2: USDA classification of salt-affected soils depending on the electrical conductivity, SAR, ESP and pH.

<b>Salt-affected soil classification</b>	<b>Electrical conductivity (ECe) [dS/m]</b>	<b>Sodium Adsorption Ratio (SAR)</b>	<b>Exchangeable Sodium Percentage (ESP) [%]</b>	<b>pH</b>	<b>Typical soil structure</b>
None	< 4	< 13	< 15	< 8.0	Flocculated
Saline	> 4	< 13	< 15	< 8.5	Flocculated
Sodic	< 4	> 13	> 15	> 8.5	Dispersed
Saline-sodic	> 4	> 13	> 15	< 8.5	Flocculated

#### 2.1.4 Processes contributing to salinity

Three processes can lead to salinity:

- (1) Groundwater salinity: occurs in drylands where evaporation is high, groundwater rises to the soil surface bringing with it dissolved salts.
- (2) Transient salinity: occurs when the infiltration rate is very low, creating a saturated zone in the subsoil. Whether salts come from rainfall or soil weathering, it accumulates in the root zone when water evaporates during the dry season. Transient salinity varies with rainfall.
- (3) Irrigation salinity: occurs when irrigation water has a high soluble salt content and insufficient leaching. Low-lying lands close to the coast are even more subject to salinity when irrigation or rainfall water are mixed with seawater. (Rengasamy, 2006, Sonon *et al.*, 2015).

Processes of salinization can also be divided into two categories: primary salinity, which is due to natural processes such as hydrologic, geologic, topographic or climatic factors and secondary salinity which is due to anthropogenic activities. The origin of the salinity in the High Valley of Cochabamba hasn't yet been deeply studied and identified. Although according to Metternicht (1996), the main causes come from the composition of the parental material, topographic position and the dry climate affecting the leaching of soluble salts, corresponding to a primary salinization. For further information, please refer to Metternicht's PhD thesis (1996) where a complete diagnose of the region of the High Valley of Cochabamba can be found.

## 2.1.5 Impacts of sodicity and salinity on soil properties

### A. Sodicity

Colloids exposed to a fluid, such as water in soils, usually have a net negative charge. This negative charge generates the double diffuse layer around colloids. Two layers can be distinguished (Figure 1):

- (1) The dense layer or Stern's layer where cations balance out the negative charge and are strongly adsorbed to the colloid,
- (2) the diffuse layer or Gouy-Chapman layer, where counter ions tend to diffuse away from the colloid where a lower concentration of ions can be found.

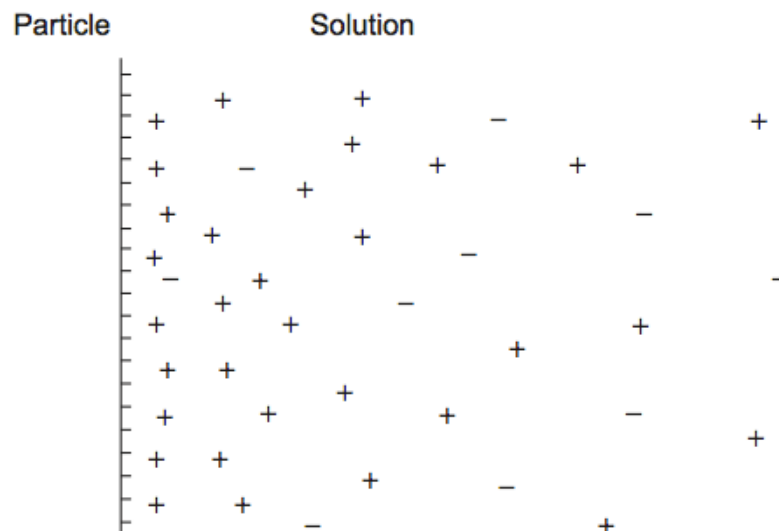


Figure 1: Diffuse double layer according to Gouy. (Van Olphen, 1977).

The cations are bonded to the colloid by electrostatic forces, whose strength depends upon the charge, the distance between the cation and the negative surface of the colloid and the valence of the cations. Furthermore, the thickness of the double diffuse layer is strongly dependent upon the type of ions and their concentration (Van Olphen, 1977).

As two colloids approach each other, electrical repulsion forces, known as swelling pressure, will exist due to their positive diffuse layer. As they get even closer, the exchangeable cations on their surface will act as a link between the two colloids, strongly binding them together and thus improving the stability of the soil. Hence, the type of exchangeable cations present will determine the strength of the bond between two colloids. For example, sodium being a monovalent cation has a poor clay-bonding ability. Whereas, calcium and magnesium being bivalent cations, their clay-binding ability

is higher. The clay-binding ability can also be expressed in terms of the cations capacity to be hydrated. Indeed, sodium and magnesium are strongly hydrated when exposed to water, creating a layer of water around them and thus, increasing the distance between two colloids. Calcium is a poorly hydrated ion. With that and its bivalent quality, calcium has the best clay-binding ability when compared to sodium and magnesium. Soils having a high concentration of sodium are therefore subject to higher swelling when exposed to water, which results in the slaking and dispersion of aggregates and thus a lower stability (Caenn *et al.*, 2017). It has been observed that swelling was positively correlated with ESP when it exceeded 15. Indeed, as explained, sodium has a much larger ionic size in water compared to calcium and magnesium which explains the higher swelling (Van Olphen, 1977, Weil & Brady, 2017).

This is one of the major issues in sodic soils. It is common to see crust formation at the soil surface, as a consequence of the rearrangement of soil particles upon drying, fine particles being washed into the pores and thus filling them up. This phenomenon called slaking, is enhanced by the mechanical impact of rainfall, known as splash, and amplified when soil stability is low. This degradation process significantly reduces the infiltration rate as shown in figure 2, increases erosion and makes it harder for seeds to emerge (McIntyre, 1958; Moore & Singer, 1990; Oster *et al.*, 1996). Furthermore, this hard layer limits roots proliferation reducing water and nutrient supply for crops (Barraclough & Weir, 1988). With swelling and dispersion, the water movement through the soil characterised by the hydraulic conductivity in unit of distance per unit of time and water entry in the soil characterised by the infiltration rate, also measured in unit of distance per unit of time, have been found to be also impacted. Quirk and Schofield (1955) found that a sodium saturated soil had a clear decrease in hydraulic conductivity mainly due to the pores being filled by the dispersed particles.

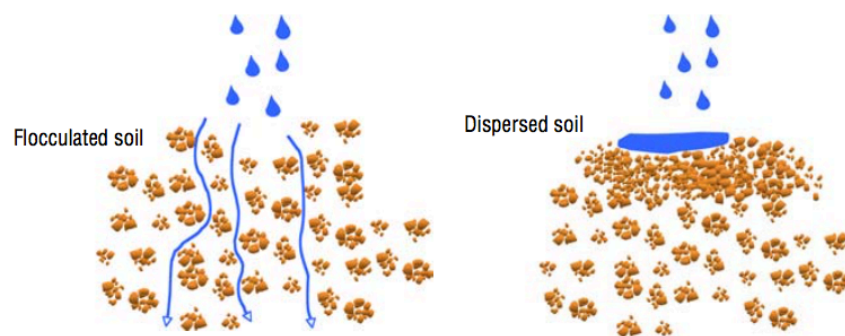


Figure 2: Flocculated soil compared to dispersed soil due to sodification. (Horneck *et al.*, 2007).

United States Salinity Laboratory (USSL) has fixed a value for hydraulic conductivity of 0.1 cm/h for which irrigation and leaching is considered to be seriously affected. McIntyre (1979) has found that for an ESP value above 15 the hydraulic conductivity can drop up to three times less than the limit fixed by the USSL.

In figure 3, a positive correlation can be seen between the quantity of exchangeable sodium and the air:water permeability ratio. The higher the ratio the higher the ESP is and therefore the higher



the deterioration of the soil structure. The deterioration of the soil is also related to the total specific area of the soil (Richards *et al.*, 1954). To understand the relationship between the total specific area of the soil and the deterioration of the soil structure a few concepts have to be introduced.

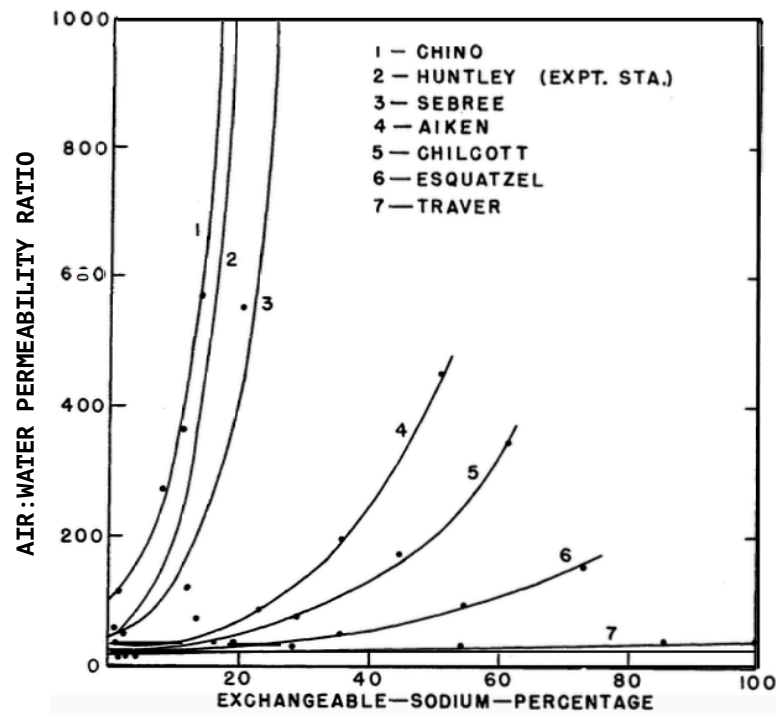


Figure 3: Effect of exchangeable sodium percentage on air:water permeability ratio. (Richards, 1954).

The interlayer distance of these clays will influence the ability of clays to swell as shown in figure 4. Indeed, where the interlayer space is bigger, for example montmorillonite also known as swelling clays,  $H_2O$  molecules and exchangeable cations are able to enter this interlayer space increasing the swelling effect when exposed to water (Al-Ani *et al.*, 2008, Weil & Brady, 2017). This can be explained by the clay mineral's specific surface area which can vary from 200-800, 65-100 and 10-20  $m^2/g$  for montmorillonite, illite and kaolinite respectively (Mitchell & Soga, 2005).

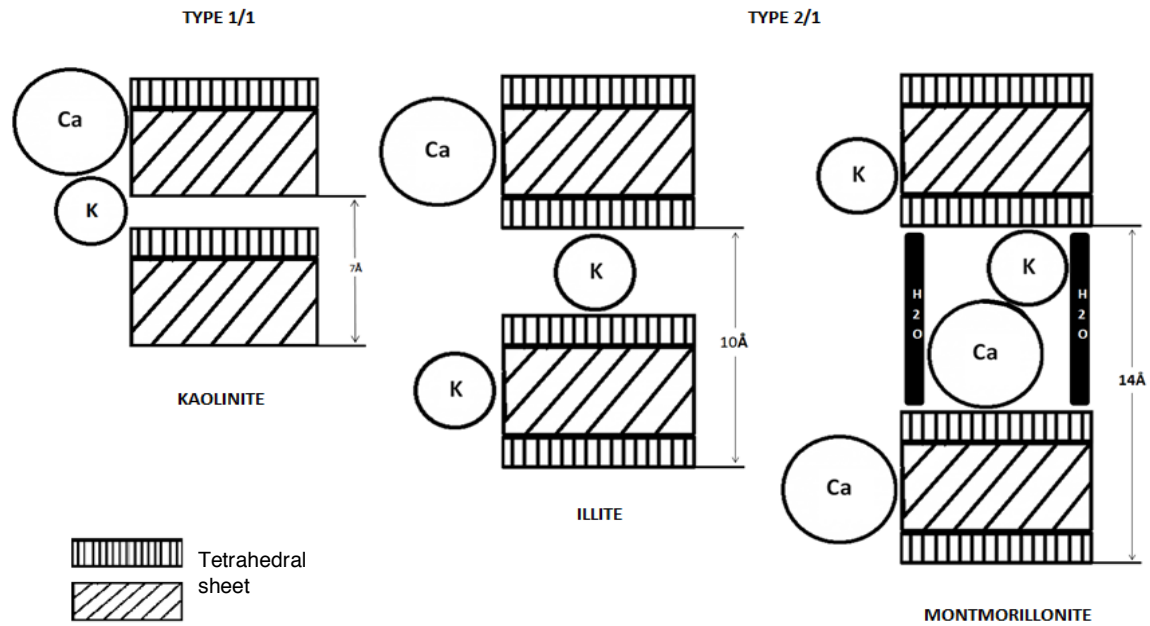


Figure 4: Structure of kaolinite, illite and montmorillonite adapted from Weil & Brady, 2017 and Bock, 2015.

Different results have been found, for example Oster *et al.*, (1980) studied the effect of the addition of sodium on illite and montmorillonite on the flocculation values of the two clays. As SAR increased, the flocculation value increased. This is shown in figure 5 for montmorillonite's and illite's flocculation value with different SAR values. Indeed, for SAR values of 5, 10 and 20, flocculation values for montmorillonite were of 3, 4 and 7 mmol/L respectively as for illite they corresponded to 6, 10 and 18 mmol/L. This effect was even more obvious with Na-montmorillonite and Ca-montmorillonite, with flocculation values of 12 mol/m<sup>3</sup> and 0.25 mol/m<sup>3</sup>, respectively, proving the negative impact of excess sodium.

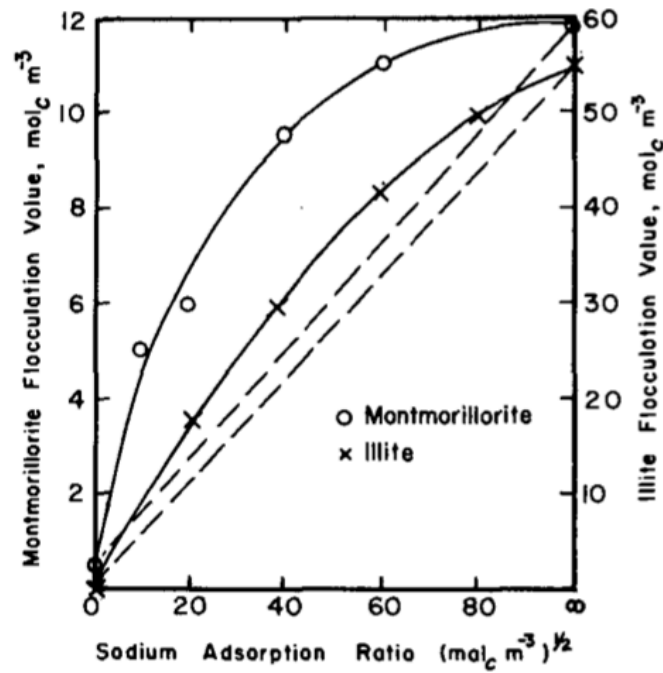


Figure 5: Evolution of montmorillonite and illite flocculation value expressed in  $\text{mol}_c \text{m}^{-3}$  with Sodium adsorption ratio expressed in  $(\text{mol}_c \text{m}^{-3})^{1/2}$ . (Oster *et al.*, 1980).

In arid and semiarid regions, the dominant mineral is montmorillonite, although kaolinite and illite can also be found (Schainberg & Letey, 1984). The problem with montmorillonite, as said previously, is that it is more subject to swelling. Studies show that from an ESP value of 15% and above, swelling increased in montmorillonite soils whereas soils containing mostly kaolinite were barely affected by variations of sodium content (McNeal & Coleman, 1966, Schainberg *et al.*, 1970).

Sodic soils are also characterised by a high pH, generally above 8.5. Gupta *et al.* (1984) studied the effect of pH on clay dispersion of soils and concluded that high pH will induce clay dispersion. Three different soils were tested and pH above 9.5, 9.2 and 7.4, respectively for the three soils, increased clay dispersion significantly.

### B. Salinity

In order to grow, crops need nutrients. On one hand there are the macro-nutrients which are essential such as carbon (C), hydrogen (H), oxygen (O), nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg) and sulphur (S). On the other hand, there are the micro-nutrients such as iron (Fe), manganese (Mn), zinc (Zn), copper (Cu), boron (B), molybdenum (Mo), chlorine (Cl), and (Ni). Carbon, hydrogen and oxygen are taken by the crop through air and water, but the remaining elements have to be absorbed from the soil solution through plant roots (Fageria *et al.*, 2011).

In salt-affected soils, excessive concentrations of soluble salts can lead to toxicity or deficiency of other nutrients for plants. In non-saline conditions, deficiency in nutrients showed to be

the most limiting factor whereas, in saline conditions, toxicity due to salinity has a higher negative impact on plant growth than nutrient deficiency (Grattan & Grieve, 1992).

Osmose is a phenomenon occurring between two liquid solutions having different concentrations of solutes. The solvent will have a tendency to flow from the lowest concentration in solutes to the highest one to reach equilibrium. The osmotic pressure is defined as the minimum pressure needed to prevent the solvent to flow from the low solute concentration solution to the high solute concentration solution (Weil & Brady, 2017). This mechanism occurs in salt-affected soils inhibiting water uptake and significantly reducing water availability to plants, therefore reducing plant growth (Rengasamy, 2010).

Thresholds for the electrical conductivity measured with the 1:5 soil to water extract method to characterise salinity of a soil depending on its clay content have been proposed (Table 3). In this table, Shaw (1999) described the plant response to a given electrical conductivity.

*Table 3: Salinity characterisation depending on electrical conductivity measured with the 1:5 soil to water ratio and clay content and plant response depending on salinity rating. (Shaw, 1999).*

Soil salinity rating	Electrical conductivity ratings (1:5, dS/m) for a range of soil clay contents				Plant response
	10–20% clay	20–40% clay	40–60% clay	60–80% clay	
Very low	<0.07	<0.09	<0.12	<0.15	no effect
Low	0.07–0.15	0.09–0.19	0.12–0.24	0.15–0.30	moderately sensitive crops affected
Medium	0.15–0.34	0.19–0.45	0.24–0.56	0.30–0.70	moderately tolerant crops affected
High	0.34–0.63	0.45–0.76	0.56–0.96	0.70–1.18	tolerant crops affected
Very high	0.63–0.93	0.76–1.21	0.96–0.53	1.18–1.87	very tolerant crops affected
Extreme	>0.93	>1.21	>1.53	>1.87	generally too saline for crop growth

## 2.2 Management of salt affected soils

To ameliorate saline, sodic and saline-sodic soils, both have to go through a leaching process to remove the excess soluble salts. A previous step can be done for sodic soils to leach the excess sodium, by providing an external source of  $\text{Ca}^{2+}$  to replace the  $\text{Na}^+$  present on the exchange complex. The choice of a remediation technique for salt affected soils depends upon many factors such as, the thickness of soil to be treated, the depth to groundwater, the soil physicochemical and biological properties, the quantity of available water and more.

In order to ameliorate soils for an agricultural purpose by leaching, a depth of at least 0,3 m should be considered as it represents the main zone of root investigation. Indeed, the irrigation should

be able to go in and out of the root zone for the leaching of the salt to be effective (Reeve *et al.*, 1948, Sonon *et al.*, 2015).

### 2.2.1 Leaching without amendments

As previously explained, salinity can occur when evapotranspiration is higher than irrigation or rainfall. Hence, a solution would be applying more water for excess salts to flow down to deeper zones of the soil, avoiding the negative impacts on crops. Several equations can be found in scientific literature to determine the amount of water needed for leaching, called “Leaching Requirement”. The easiest LR calculation is the one used by the USDA (1954) and can be expressed as follows:

$$\frac{D_{dw}}{D_{iw}} = LR = \frac{EC_{iw}}{EC_{dw}}$$

where  $D_{dw}$  and  $EC_{dw}$  are the depth and electrical conductivity of the drainage water and  $D_{iw}$  and  $EC_{iw}$  are the depth and electrical conductivity of the irrigation water. Another widely used way to calculate LR, can be done with the following equations (Ayers & Westcot, 1985):

Surface irrigation:

$$LR = \frac{EC_w}{5 * EC_e - EC_w}$$

Sprinkler or drip irrigation:

$$LR = \frac{EC_w}{2 * (maxEC_e)}$$

where  $EC_w$  is the electrical conductivity from the water analysis and  $EC_e$  is the maximum electrical conductivity of the soil for a given crop to have a certain degree of yield reduction. Further information on the  $EC_e$  of the soil for a given crop yield reduction, are available in the Food and Agriculture Organisation of the United Nations (FAO) report written by Ayers and Westcot (1985). However, these methods have been known to have their limits since they are based on steady-state conditions. In other words, salt concentration, water and solute flow and water content is supposedly constant over time, which isn't true in field conditions (Letey *et al.*, 2011). A more appropriate approach would be to use models using transient-state conditions. Indeed, Corwin *et al.* (2011) has compared steady-state and transient-state models and concluded that with a transient-state model, water quantities applied could be reduced by 125 Mm<sup>3</sup>. Other studies have calculated the precise leaching requirement depending on the crop grown in order to have a full crop production capacity (Hoffman *et al.*, 1979, Jobes *et al.*, 1981). Lastly, studies particularly the ones of experimentations in

columns have used the pore volume of soil to calculate the amount of water to be applied to obtain adequate leaching of the soil columns (Kahlon *et al.*, 2013, Ahmad *et al.*, 2015).

Once the LR is calculated, the type of leaching method has to be chosen to maximise efficiency. Several leaching methods have been studied to ameliorate saline soils with decreasing efficiencies as followed; intermitting ponding > sprinkler irrigation > continuous ponding. Oster *et al.* (1972) have confirmed that for all the three types of leaching mentioned, the time needed to reduce salinity by half was the same, but the quantities of water applied for each technique were different. Hence, for intermitting ponding, sprinkler irrigation and continuous ponding the water quantities applied were 301.2 cm, 349.4 cm and 465.8 cm respectively (Oster *et al.*, 1972).

For sodic soils, a source a  $\text{Ca}^{2+}$  is required to leach the excess  $\text{Na}^+$ . This is possible in gypsiferous and calcareous soils without any supplies of amendments, in other situation chemical amendments are sometimes imperative to the reclamation of sodic soils.

## 2.2.2 Chemical amendments

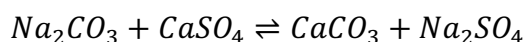
Reclamation of sodic soils requires application of some chemical amendments, more precisely a source of  $\text{Ca}^{2+}$ , to replace the exchangeable  $\text{Na}^+$  present in the root zone. Several types of amendments exist: soluble calcium salts, acids or acid forming substances (FAO, 1988). Gypsum ( $\text{CaSO}_4 \cdot 2\text{H}_2\text{O}$ ) acting as an external source of  $\text{Ca}^{2+}$  to reduce the amount of  $\text{Na}^+$  in sodic soils as well as sulphuric acid ( $\text{H}_2\text{SO}_4$ ), are the most commonly used since it has a relatively low cost and is easily obtained (Qadir *et al.*, 2001).

### A. Principle

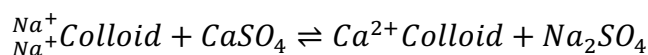
Gypsum is a common evaporite mineral, its occurrence comes from sedimentary rock deposits in lagoons, lakes or on the coast. Water rapidly evaporates, leaving an excess of precipitated salts, sulphate and calcium in the case of gypsum (Tucker, 2001). When gypsum is grounded and added to a sodic soil, the sodium will associate with the sulphate making it leachable as shown is the following reactions (Richards *et al.*, 1954, FAO, 1988, Weil & Brady, 2017):



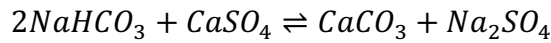
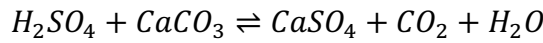
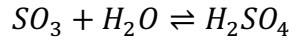
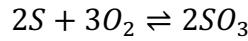
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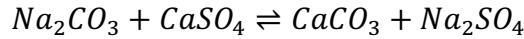
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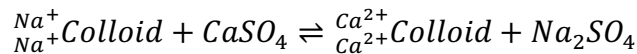
Sulphuric acid on the other hand, is not found naturally on earth, it is essentially an industrial product. Nevertheless, sulphuric acid can be found by applying sulphur (S) directly to the soil. Sulphur is commonly obtained through the extraction of natural gases and petroleum. Huge quantities of sulphur deposit, as combined states or free states, can be found all around the world (Tucker, 2001, Weil & Brady, 2017). When sulphur, as a yellow powder, is applied to a sodic soil, it will be oxidised by microbiological activities, thus forming sulphuric acid. Sulphuric acid will then dissolve the calcite if it's present in the soil, producing the needed source of calcium to remove the exchangeable sodium. The sulphuric acid can also directly react with carbonated sodium present in the soil. Of course, since sulphur needs to first react with the micro-organisms present in the soil to form sulphuric acid, a warm and aerated environment is required. Furthermore, the use of this amendment is slower compared to applying sulphuric acid directly (Hanson *et al.*, 2006). These reactions can be expressed as follows (FAO, 1988, Qadir *et al.*, 2007, Richards *et al.*, 1954, Weil & Brady, 2017):



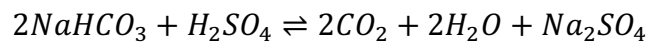
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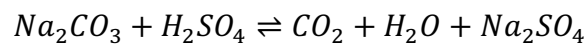
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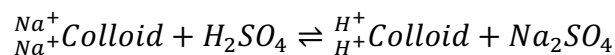
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or



or



Many studies on the effects of gypsum have been carried out but there are still some discussions about the quantity of gypsum needed and how it actually influences the leaching of cations (Bresler *et al.*, 1982). Gypsum requirement is often calculated with the following equation (Hoffman, 2007, Lebron *et al.*, 2002, Richards *et al.*, 1954):

$$GR = (0,0086 * F * BD * D * CEC * (ESP_i - ESP_f))$$

where GR is the gypsum requirement, F is a Ca-Na exchange efficiency factor, BD is the bulk density expressed in g/cm<sup>3</sup>, D is the depth of soil to be reclaimed expressed in cm, CEC is the Cation Exchange Capacity expressed in meq/100 g and ESP<sub>i</sub> and ESP<sub>f</sub> are the initial and final Exchangeable Sodium Percentage expressed in %. The Sulphur requirement can be evaluated by dividing the gypsum requirement by 5.38 (Richards *et al.*, 1954).

### B. Literature review

With a determined GR dose, positive effects on aggregation, exchangeable cations, infiltration rate, hydraulic conductivity, EC, ESP have been reported. Hamza and Anderson, (2003) found improvement in physical properties such as, infiltration rate, exchangeable Ca and water-stable aggregates with the application of gypsum. Infiltration rate after 4 years of treatment had increased by 68%, exchangeable Ca increased from 49 to 64% and water-stable aggregates increased by 34% after gypsum application on a sandy loam.

Abdel-Fattah (2012), has tested 3 different treatments, gypsum, water hyacinth compost and rice straw compost combined with or individually. At the end of the leaching process, EC, pH and ESP had decreased although the 100% GR in combination with the rice straw compost was the most effective for all the indicators. The initial ESP value of 39.55% was decreased at least up to 4.99%, a value under the sodic threshold value. As for the pH and the EC it decreased from 8.36 dS/m to at least 8.06 dS/m and 20.34 dS/m to at least 6.50 dS/m respectively.

Different levels of gypsum application, 0%, 25%, 50%, 75%, 100% and 200% of the gypsum requirement to reclaim a salt-affected soil grown under rice. Wheat crop was then grown on the same reclaimed soils without any gypsum application. No significant effect was found on wheat yield between treatments of 75%, 100% and 200% GR. Yield increased by 75.5% with both 75% and 100% GR and by 80% with 200% GR. This shows that a 75% GR, a more economical solution, could be sufficient to remediate salt-affected soils (Khattak *et al.*, 2007).

Application of sulphuric acid to remediate salt affected soils can also be applied and has shown its effectiveness. As previously explained, the sulphuric acid requirement is lower than the gypsum requirement, and studies have shown its equivalent effectiveness if not better. Overstreet *et al.*, (1951) compared the impact on yield of pasture plants of sulphur, gypsum and sulfuric acid, with doses of 1.86 tons/acre, 10 tons/acre and 5.70 tons/acre respectively. The mean yields reported with sulphur, gypsum and sulphuric acid application were of 1.71, 2.46, 3.47 of fresh weight/acre. Not only did sulphuric acid give the most promising result but the dose needed compared to gypsum is almost reduced by half. With these promising results for sulphuric acid a second experiment was carried out. With a dose of 1.42 tons/acre of H<sub>2</sub>SO<sub>4</sub> and 2.50 tons/acre of gypsum, yield was still



almost twice as much for sulphuric acid treatments compared to gypsum. Mean yields for alfalfa were of 8.03 tons/acre with a sulphuric acid treatment and of 4.90 tons/acre with a gypsum treatment.

Worku *et al.*, (2016) observed that 100% of gypsum (doses from the FAO guidelines) gave an average yield of 2.29 tons/ha and 100% H<sub>2</sub>SO<sub>4</sub> gave an average yield of 2.63, which isn't a significant difference. But the combination 50% gypsum with 50% H<sub>2</sub>SO<sub>4</sub> proved to be significantly higher than the two latter treatments with an average yield of 3.13 tons/ha. An ESP and EC<sub>e</sub> reduction of 43.71% 60.3% respectively with the 50% gypsum combined with 50% H<sub>2</sub>SO<sub>4</sub> reflected to be the best treatment compared to 100% gypsum and 100% H<sub>2</sub>SO<sub>4</sub>.

Sulphuric acid has also proven to increase the infiltration rate by crusting prevention up to 52.7 mm/h compare to the control which had a 21.1 mm/h infiltration rate. A faster reaction was also observed with acid amendment compared to gypsum amendments. An SAR value of 1 was obtained in less than one hour compared to more than 2 hours for the gypsum (Amezketta *et al.*, 2005).

As much as sulphuric acid has proven its effectiveness, little research has been done on the effects of sulphur and the quantity needed in the particular case of salt-affected soils reclamation. Still, Ahmed *et al.*, (2016) compared sulphur (S) to gypsum using the soil gypsum requirement (SGR) to calculate the doses of sulphur to apply. The efficiency of the different treatments in terms of improvement of yield and reduction of soil pH, EC<sub>e</sub> and SAR were evaluated as gypsum with 100% SGR = sulphur with 125% of SGR = sulphur with 100% of SGR > sulphur with 75% of SGR > sulphur with 50% of SGR > sulphur with 25% of SGR > control. With the gypsum treatment EC<sub>e</sub> and SAR were decreased by 44.09% and 60.04% respectively. As for the sulphur with 125% of SGR and with 100% of SGR, which showed no significant difference with the gypsum treatment, EC<sub>e</sub> was lowered by 44.26% and 43.27% and SAR by 55.36% and 54.45% respectively. Grain yield has shown to increase with the application of sulphur. With doses of 25, 50 and 75 kg of S/ha grain yield was of 3.60, 4.04 and 3.62 t/ha compared to the control which had a grain yield of 3.2 t/ha, a significant difference according to Ali *et al.* (2012). Unfortunately, the effect on physical and chemical properties of a salt-affected soil are poorly analysed.

Chemical amendments could be a solution although leaching through precipitation or irrigation is mandatory to evacuate the excess salts from the root zone. In the region of Cochabamba, where an arid climate can be found, access to water isn't always easy (Ayala Flores *et al.*, 2009). A solution to this issue could be the use of halophytes to desalinise without the necessity to leach.

### 2.2.3 Phytoremediation

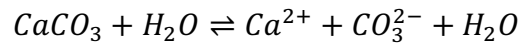
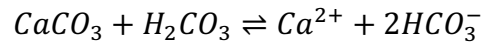
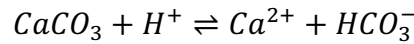
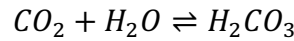
#### A. Principle

Phytoremediation is well known for its potential to remediate contaminated by toxic metals soils and for its technological and economic advantages to do so (Flathman & Lanza, 1998). Phytoremediation to desalinise salt-affected soils is an emerging plant-based solution using

halophytes which are species tolerant to salt. Four factors happening via the phytoremediation process contribute to the desalinisation of a soil: partial pressure of CO<sub>2</sub> in the root zone, improvement of soil aggregation and hydraulic properties of the root zone, accumulation of Na<sup>+</sup> in the shoots and release of protons (H<sup>+</sup>) in the root zone (Qadir & Oster, 2004). These different mechanisms are represented in figure 6 and are explained below:

- Partial pressure of CO<sub>2</sub> in root zone

As it is well known, plant respiration is essential for their growth. One of the by-product of plant respiration is CO<sub>2</sub>, extracted from the atmosphere and released in the root zone (Evert & Eichhorn, 2013). As CO<sub>2</sub> is released through the roots it can react with calcite to release the needed Ca<sup>2+</sup> to replace the exchangeable Na<sup>+</sup>. This reaction is the result of different processes, firstly CO<sub>2</sub> will be hydrated and secondly three different reactions can occur to release the Ca<sup>2+</sup> through the dissolution of calcite. These processes are defined by the following equations (Dreybrodt *et al.*, 1996, Robbins, 1985):



- Soil structure

Plant roots have many beneficial effects on the structure of the soil and thus improving its properties. Firstly, as roots grow they have a capacity to move soil particles around, creating pores needed for water movements. Secondly, roots are a natural habitat for many micro-organisms which contribute to the stability of soils among other activities. (Gregory, 2006, Oades, 1993).

- Accumulation of Na<sup>+</sup>

As previously said, halophytes are defined as plants capable of growing in saline environments. A mechanism that enables halophytes them to grow in saline environments, is to accumulate salts in their shoots via a compartmentalisation of the ions in the vacuole (Fageria *et al.*, 2011, Evert & Eichhorn, 2013).

- Release of H<sup>+</sup> in root zone

It is well known that as root uptake cations, H<sup>+</sup> are released to the environment. One of the problems of saline-sodic soils is the increasing pH, through the alkalinisation process

as previously explained. Thus, plants by their everyday activity of cation uptake, could help decrease the soil pH (Overstreet & Jacobson, 1952). Another positive effect that release of protons by roots would do, is as explained for the respiration process,  $H^+$  facilitates the dissolution of calcite, releasing the needed calcium.

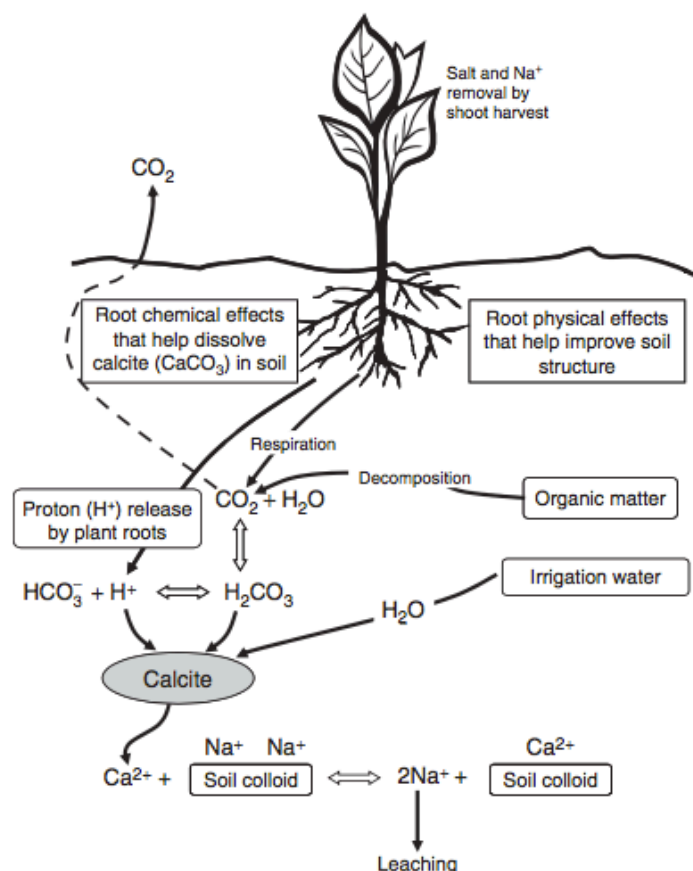


Figure 6: Schematic representation of the beneficial effects of phytoremediation. (Qadir *et al.*, 2007).

## B. Literature review

Results close to the ones achieved with chemical amendments can be found. Qadir *et al.*, (1996) compared the reclamation of saline-sodic soils by 2 different doses of gypsum and *Leptochloa fusca*, also known as kallar grass. The total amount of sodium removed by leaching was of 1884.2, 1300.7 and 1022.7 mmol<sub>c</sub> for the 100% - GR, kallar grass and 50% - GR treatments respectively. Even though the 100% - GR was still more effective, kallar grass managed to remove more sodium than the 50% - GR by releasing  $H^+$  enabling the dissolution of native  $CaCO_3$  in the soil (Qadir *et al.*, 1996). *Sulla carnosa*, a halophyte, had a phytodesalinisation capacity of 0.3 t of  $Na^+$ /ha and managed to reduce the E<sub>c</sub> from 8.4 dS/m to 5.8 dS/m in the upper horizon (0-8cm) (Jlassi *et al.*, 2013). Shekhawat *et al.*, (2006) studied the effect of three halophytes, *Salsola baryosma*, *Haloxylon recurvum* and *Suaeda nudiflora* on the physical and chemical properties of a salinized soil. The different species managed to change EC, pH, exchangeable  $Na^+$ , exchangeable  $Ca^{2+}$  and ESP positively depending on the depth analysed for the reclamation of secondary salinization by

agricultural practises. Overall decreasing efficiency was reported as follows *H. recurvum* > *S. nudiflora* > *S. baryosma*. *H. recurvum* managed to decrease the ESP by up to 55%, and the EC by up to 14% over a period of time of one year (Shekhawat *et al.*, 2006).

In the High Valley of Cochabamba, many halophytes grow naturally such as *Salicornia pulvinata*, *Frankenia trianda*, *Distichlis humilis* or *Suaeda foliosa*. *Suaeda foliosa*, commonly known as q'awchi is widely used as an animal feed. It's tolerance to brutal climatic changes and salinity and its nutritional value comparable to other animal feeds such as alfalfa make it a particularly good specie for the reclamation of salt-affected soils (Ayala Flores *et al.*, 2009). Even though no studies could be found in the scientific literature on *Suaeda foliosa*, Zhao (1991) reported that for *Suaeda salsa*, a specie of the same genus as *Suaeda foliosa*, sodium content in the soil was reduce by 4.5% after a growing period of 120 days.

### 3. Objectives

The main objective of this paper is to evaluate remediation techniques specific to saline-sodic soil in order to contribute to the rehabilitation of salt-affected soils of the High Valley of Cochabamba. As explained in the first section of this report, salinity and sodicity have a negative impact on the physical and chemical properties of the soil, reducing therefore crop yield. We have seen in the scientific literature that three main strategies exist to reclaim salt-affected soils: leaching, chemical amendments and phytoremediation.

To achieve the main objective of this report, the following research question was elaborated:

“What is the potential of rehabilitation of saline-sodic soils of the High Valley of Cochabamba with gypsum, sulphur and *Suaeda foliosa*, respectively?”

In the attempt to answer the research question, three specific objectives were considered, the first two regarding the use of chemical amendments and the third one for the phytoremediation. Leaching will not be tested in this study since literature review showed that it is mainly used in the case of saline soils and not sodic ones. In addition, this technique necessitates a lot of water that is not necessarily available in the High Valley of Cochabamba.

Chemical amendments such as gypsum and sulphuric acid have proven to be effective in the reclamation of salt-affected soils. Their biggest advantages being their accessibility and low price for farmers. But the optimal dose to be applied to achieve a certain rehabilitation in terms of physical and chemical properties of the soil still lack accuracy for gypsum and sulphur. Indeed, the requirement depends on many factors and is directly influenced by the nature of the soil to be reclaimed.

Hence, the first objective of this report is to determine the dose to apply for an optimal reclamation of saline-sodic soils of the High Valley of Cochabamba in Bolivia with gypsum and sulphur. The following hypothesis can be stated to answer the first objective:

- H1: There is an optimal dose of gypsum/sulphur to reclaim the saline-sodic soils of the High Valley of Cochabamba.

The second objective can be formulated as to understand how the chemical amendments affect the chemical properties of the soil, in particular sulphur. Indeed, gypsum has already proven to be one of the most effective amendment. Whereas for sulphur, even though it becomes sulphuric acid when it reacts with the micro-organisms, not much information can be found as to how it affects the variables characterising salt-affected soils such as ESP, EC and pH. The following hypothesis can be then put in place to answer the second objective:

- H2: Sulphur has a similar effectiveness as gypsum in terms of soil chemical properties, to reclaim the saline-sodic soils of the High Valley of Cochabamba.

Literature review has also shown that phytoremediation can achieve positive results to reclaim salt-affect soils depending on the crop used. Halophyte have shown their capacity to accumulate sodium, but no specific studies were found for *Suaeda foliosa*, a native plant of the High Valley and commonly used as animal feed in the region. The third objective will be to evaluate the soil phytodesalinisation capacity of *Suaeda foliosa* for salt-affected soils of the High valley of Cochabamba. A sub-objective can be pointed out, to compare the potential of phytoremediation with the one of the chemical treatment. Hence, the following two hypotheses were made to answer to this third objective:

- H3: Phytoremediation with *Suaeda foliosa* is an effective solution to reclaim the salt-affected soils of the High Valley of Cochabamba.
- H4: The effectiveness of phytoremediation to reclaim the salt-affected soils of the High Valley of Cochabamba is comparable to the effectiveness of chemical amendments.

To test the different hypotheses elaborated to achieve the three objectives of this study, an experimentation was put in place using tools and methods described in the following section.

## 4. Material and Methods

### 4.1 Study site

The soils used in this experimentation were all previously sampled by Alexis Weber and Demis Andrade as part of the characterisation phase of the soils of the High Valley of Cochabamba. Two different locations were used to extract soil samples at a depth of 20 cm.

#### Saline-sodic soil 1

The first one being Santa Ana, part of the Municipality of Punata shown in figure 7 situated at 17° 32' 38.6" South and 65° 51' 41.9" West had a saline-sodic soil, based on the classification of the NRCS. The soil fraction in the first horizon (0-20cm), used for this study, is composed of 19.3% clay, 54.9% silt and 25.8% sand, the textural class is therefore silty loam.

#### Saline-sodic soil 2

The second site, Aramasi part of the Municipality of Cliza (Figure 7) located at 17° 35' 52.7" South and 65° 51' 34.9" West, was composed of another saline-sodic soil. The soil in the first horizon is composed of 33.4% clay, 63.8% silt and 2.8% sand, making it a silty clay loam (Weber, 2018).

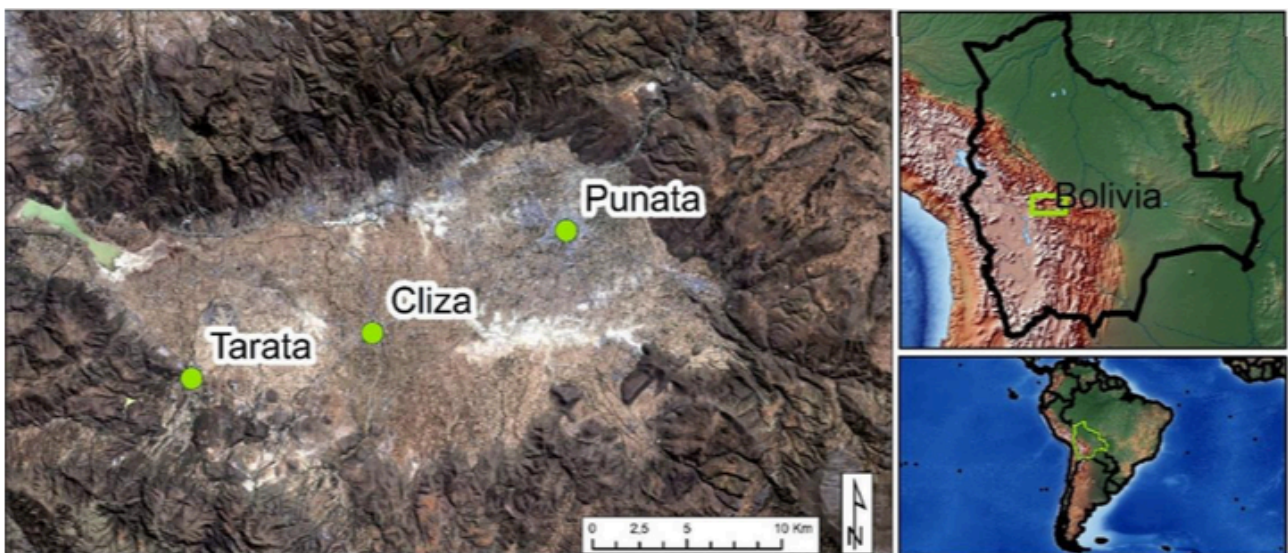


Figure 7: Location of the High Valley of Cochabamba and the soils used. (Weber, 2018)

Figure 8 shows the distribution of saline, saline-sodic, sodic or normal soil based on a simple classification only considering the ESP and EC predicted in the area studied by Weber (2018) in the high valley of Cochabamba.



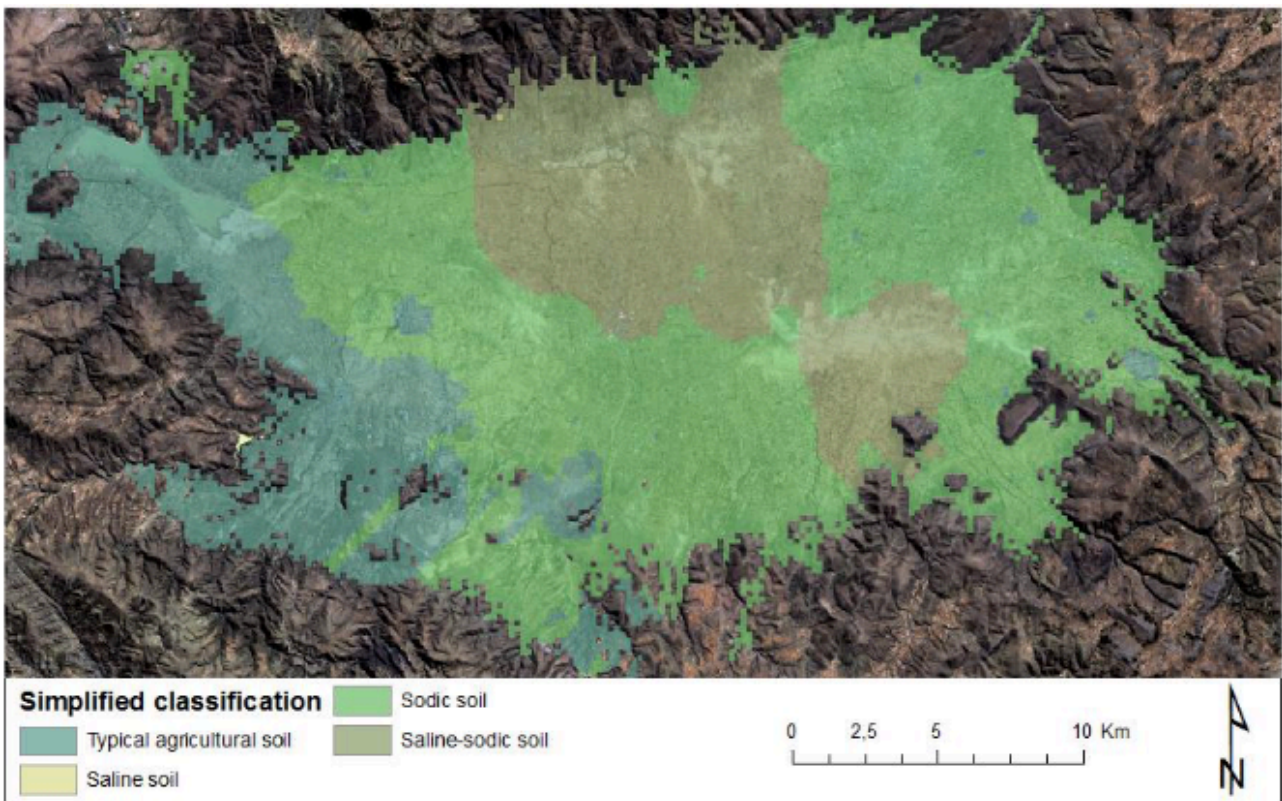


Figure 8: Simplified classification of the soils in the High Valley of Cochabamba based on ESP and EC. (Weber, 2018).

## 4.2 Treatments

The experiments were carried out under a plastic greenhouse, to control climate conditions, at the faculty of Agronomical and Animal Sciences of the Universidad Mayor de San Simon, Cochabamba, Bolivia.

### 4.2.1 Soil preparation

For all treatments, the soil was previously air dried and then sieved to 4 mm. The soil was thoroughly mixed in order to reduce heterogeneity. The bulk density was determined through the tapped density method chosen for its simplicity, which consists in mechanically tapping a known weight of soil contained in a graduated cylinder. Tapped density is then calculated by dividing the mass of the soil by the tapped volume of soil observed.

### 4.2.2 Chemical amendments

The different chemical treatments trials were carried out in perforated pots. The pots were perforated at their base in order to collect the leachates through a tube connected to a plastic container as shown in figure 9.





Figure 9: Chemical amendment pots connected to plastic container to collect leachates.

Only the sodic soil from Santa Ana was used for this experiment. Two different chemical amendments were used; gypsum and sulphur. As previously explained, the choice of these two amendments was made because they are the most common amendments and therefore easily obtainable in the High Valley of Cochabamba. Also, they are relatively cheap compared to other amendments. The purity of both amendments was evaluated. The gypsum requirement was estimated in order to reduce ESP down to 15%. This requirement was calculated based on the gypsum requirement (GR) formula previously mentioned:

$$GR = (0,0086 * F * BD * D * CEC * (ESP_i - ESP_f))$$

CEC was measured with a modified Metson method used at the faculty in Bolivia which consists in removing the ions adsorbed on the exchange complex by addition of 20ml of sodium acetate at pH7 to 1g of soil. The sodium acetate is then quantified by desorption with addition of 95% ethanol and potassium chloride. In order to have an estimation of the SAR, magnesium, calcium and sodium had to be estimated. Total calcium and magnesium were calculated with the complexometric titration method also known as the Versenate method which was the only method available at the soil lab of the faculty in Bolivia. As for the sodium it was estimated with the *Laquatwin Na-11* portable device. Initial ESP was then estimated with the following equation showing the relationship between SAR and ESP (Richards *et al.*, 1954):

$$ESP = \frac{100 * (-0.0126 + 0.01475 * SAR)}{1 + (-0.0126 + 0.01475 * SAR)}$$

The sulphur requirement (SR) was determined by the following formula (Richards *et al.*, 1954):

$$SR = \frac{GR}{5,38}$$

Two different doses were applied for each amendment, one at 100% and one at 50% of the corresponding requirements. As the GR formula isn't a perfect evaluation of the requirement, two doses have been chosen to see if an effective reclamation could be done by only applying half of what is supposedly needed. Five replicates were done for each treatment and two controls, soil without any amendments, were put in place. The soil was thoroughly mixed with the amendments powder in a plastic bag before being put into the pots. Each pot contained therefore:

- a height of 2 cm of gravels having a diameter between 4 mm and 6.3 mm, which corresponded approximately to 0.4 kg of gravel,
- 2 kg of dry soil mixed with either 100% - GR, 50% - GR, 100% - SR or 50% - SR,
- A total of 450 ml of water applied layer by layer.

The soil was packed in the pots layer by layer and was tapped to reach a bulk density of 1.4 g.cm<sup>3</sup>. Four leaching solutions were collected every two weeks from the 7<sup>th</sup> of April until the 30<sup>th</sup> of May. The quantity of water applied were calculated according to the protocol used by Ahmad *et al.*, (2015). 450 ml determined by the pore volume formula, were first applied to reach 75% of soil saturation:

$$Pore\ Volume = \theta_v * \pi * r^2 * h$$

with  $\theta_v$  being the volumetric water content,  $\pi$  is a constant equal to 3.14, r and h are the radius and the height of the pots expressed in cm.

660 ml (100% saturation) were then applied in between each leachate collect. The control which did not contain any amendments also went through the leaching process.

Figure 10 shows the configuration of all the treatments once put in place in the greenhouse of the faculty.



Figure 10: Pot experiment layout in the greenhouse at the faculty of Agronomical and Animal Sciences of the Universidad Mayor de San Simon, Cochabamba, Bolivia.

#### 4.2.3 Phytoremediation

The plant used for the phytoremediation treatment was *Suaeda foliosa*, commonly named Q'awchi, as it is a native halophyte used for animal feed as previously mentioned in the state of art. These plants were grown in the greenhouse of the Research Centre for Halophytes, Toledo, Oruro, Bolivia.

Three different phytoremediation treatments were carried out in non-perforated pots, organised in a completely randomized design as followed:

Treatment A: sodic soil 1 + *Suaeda foliosa* of 3-4 months of age,

Treatment B: sodic soil 2 + *Suaeda foliosa* of 3-4 months of age,

Treatment C: sodic soil 1 + *Suaeda foliosa* of 1-2 months of age.

All treatments were composed of 5 replicates and 1 control. Each pot, aside from the controls which did not have any plants, contained:

- a height of 1.5 cm of gravels having a diameter between 4 mm and 6.3 mm, which corresponded approximately to 0.3 kg of gravel,
- 2 kg of dry soil,

- Three *Suaeda foliosa* plants,
- A total of 300 ml of tap water applied every 4 cm of soil layer.

The soil was put into the pots layer by layer and each layer was humidified to assure field capacity to avoid the death of the plants. Once the experimentation put into place, the pots were irrigated every 5 to 10 days with 100 ml of tap water depending on the dryness of the soil by simple observation.

## 4.3 Analyses

### 4.3.1 Chemical amendments

#### A. Leachate analysis

All the leachates were analysed by atomic absorption spectroscopy (spectrAA Varian 220) in Gembloux throughout the month of June. The spectrometer uses the absorption of optical radiation of the different elements measured to determine the concentration in each sample of leachate.

The electrical conductivity and the pH of the leachate samples were measured with the Oakton PCD650 device. SAR was calculated with the following formula previously mentioned:

$$SAR = \frac{[Na^+]}{\sqrt{\frac{[Ca^{2+}] + [Mg^{2+}]}{2}}}$$

Where concentrations are expressed in mmol/L.

The total cumulated concentration of the cations after the final leachate was calculated by multiplying the concentrations in each leachate by the volume of leachate collected. The quantities were then summed up.

#### B. Soil analysis

Soil were sampled with a core of about  $\frac{3}{4}$  of the height of the pots. All the soil samples were sent to the laboratory of soil analysis of the Liège province. The analyses and methods used are summarised in table 4.

Table 4: Methods used by the external lab for the soil sample analysis.

Element analysed	Method used
pH	Modified method of the ISO 10390 standard, KCl 1N
Electrical Conductivity	1:5 soil to water extract
Cation Exchange Capacity	Modified method of the ISO 23470 standard
Calcium, magnesium, potassium, sodium	Metson method at pH7 (Ammonium acetate + ethanol + sodium chloride)
Calcium, magnesium, potassium	Lakanen-Erviö method pH4.65 (Ammonium acetate + Ethylenediaminetetraacetic acid)

A correlation between the concentrations of cations measured with the Lakanen-Erviö method and measured with the Metson method.

ESP was calculated to only take into account the most important cations with the following formula:

$$ESP = \frac{Na^{+}}{Ca^{2+} + Mg^{2+} + K^{+} + Na^{+}}$$

Since the electrical conductivity was measured with the 1:5 soil to water ratio, the EC<sub>e</sub> threshold fixed by the USDA (1954) was divided by a factor given by Sonmez *et al.*, (2008) in order to compare it to the results. This factor depends on the texture of the soil analysed, in our case a loamy soil, giving therefore the factor value of 7.62.

### C. Mass balance

A mass balance (MB) for calcium in the soil of a pot for the gypsum treatment was estimated by the following equation:

$$MB [mg] = \text{Initial Calcium quantity per pot} + \text{Calcium quantity in gypsum per pot} - \text{Calcium leached per pot}$$

## 4.3.2 Phytoremediation

### A. Shoot analysis

In order to analyse the salts present in the shoots of the plants, a mineralisation of the dry matter had to be done to solubilise the mineral elements (Van Ranst *et al.*, 1999). The destruction of the organic matter was done by adding 30 ml of a 50/50 acid mix of nitric acid at 65% and perchloric acid at 70% to each sample of dry matter in a beaker. The samples were then left to rest for 16 hours. The next morning, the samples were agitated in slow motion and heated until the residues were

completely dehydrated. Five millilitres of hydrochloric acid were then added and left to rest for a few minutes. The solution was then transferred in 25 ml flasks and diluted with distilled water. Afterwards, the samples had to be filtered with 602 H<sup>1/2</sup> filters and transferred in 50 ml containers, ready to be analysed by atomic absorption spectroscopy. Magnesium, potassium, sodium and calcium were measured using different dilutions when necessary, depending on the sample analysed (See appendices). The values obtained were then reported to the collected dry matter weight.

#### *D. Soil analysis*

The same soil sampling method and analyses as for the chemical amendment experimentation were carried out.

#### 4.3.3 Water analysis

The electrical conductivity, pH and sodium content in the irrigation water were also measured.

#### 4.3.4 Statistical analysis

Statistical analyses were realised with R studio version 1.0153. One-way ANOVA were applied for the parameters analysed, exchangeable cations, pH, CEC, EC, ESP. Duncan's multiple range tests for the soil before and after treatment was realised for the phytoremediation and chemical treatments. Dunnett's tests were also done to compare the different treatments with their control.

## 5. Results and discussion

Before moving on to the results of the experimentation a small parenthesis should be done: the correlation between the concentrations of calcium, magnesium and potassium measured with the Lakanen-Erviö method and measured with the Metson method was highly significant. For calcium  $r = 0.91$  and  $p < 0.001$ , for magnesium  $r = 0.83$  and  $p < 0.001$  and for potassium  $r = 0.94$  and  $p < 0.001$ . Since the concentration of sodium was analysed only with the Metson method and a good correlation was found, all the statistical analyses have been done with the values obtained with the Metson method.

### 5.1 Properties of the soils before treatment and irrigation water

Table 5 shows the initial properties of the two saline-sodic soils used.

*Table 5: Properties of the soils before treatments.*

Soil	pH	EC [ $\mu\text{S}/\text{cm}$ ]	CEC [ $\text{cmol}_\text{c}/\text{kg}$ ]	Ca [ $\text{cmol}_\text{c}/\text{kg}$ ]	Mg [ $\text{cmol}_\text{c}/\text{kg}$ ]	K [ $\text{cmol}_\text{c}/\text{kg}$ ]	Na [ $\text{cmol}_\text{c}/\text{kg}$ ]	ESP [%]
Saline-sodic soil 1	9.61	2982.1	5.08	5.01	0.53	0.17	13.11	69.66
Saline-sodic soil 2	9.86	2611.1	8.16	8.90	0.33	0.73	17.96	64.33
Q'awchi original soil	7.25	745.1	6.88	9.32	1.40	1.52	0.92	6.96

The phytoremediation which was carried out in the saline-sodic soil 2 did not come through, since all the plants died progressively in a month. On the other hand, in the saline-sodic soil 1, the plants survived but with a certain lack of wellness. This is a quite peculiar since the ESP and EC values of the saline-sodic soil 1 which are of 69.66% and 2982.13  $\mu\text{S}/\text{cm}$ , respectively are higher than the ones of the saline-sodic soil 2 which are of 64.33% and 2611.13  $\mu\text{S}/\text{cm}$ . On the other hand, the concentrations of the exchangeable cations are higher in the saline-sodic soil 2 than in the sodic soil 1 except for the magnesium. The higher sodium content in the sodic soil 2, which was evaluated at 17.96  $\text{cmol}_\text{c}/\text{kg}$  compared to the one of the saline-sodic soil 1 of 13.11  $\text{cmol}_\text{c}/\text{kg}$ , must've affected the plants. Another reason for the plants to have not survived in the saline-sodic soil 2 could be the texture of the soil, which was clayey. The drainage could have been reduced and there might have been a lack of oxygen for the transplanted plants (Drew, 1997).

Electrical conductivity, pH and sodium content of the irrigation water are shown in table 6. As electrical conductivity and sodium content values of the water are well below the electrical conductivity and sodium content values of the leachates it can safely be considered that they didn't influence the results of the leachates. Ayers and Westcot (1985) reported that an EC over 750  $\mu\text{S}/\text{cm}$  will start inducing salinity problems for crops.

Table 6: Electrical conductivity, pH and sodium content of the irrigation water.

EC [ $\mu\text{S}/\text{cm}$ ]	pH	Na [mg/L]
230	8.1	25

## 5.2 Chemical amendments

In this section, the results of the leachate and soil analysis of the chemical amendments treatments will be presented and discussed.

### Properties of the Amendments

The purity of the gypsum and sulphur were evaluated at 91.74% and 97.5% respectively. For the gypsum the content of calcium was of 18.5%. The gypsum and sulphur requirements calculated for the different treatments are indicated in table 7.

Table 7: Corresponding doses of gypsum and sulphur for 100% - GR, 50% - GR, 100% - SR and 50% - SR treatments and calcium content of gypsum in the pots.

	100% - GR	50% - GR	100% - SR	50% - SR
Weight per pot [g/pot]	23.05	11.53	4.28	2.14

### 5.2.1 Leachate analysis

Figure 11, 12, 13 and 14 show the evolution of the concentration of sodium, magnesium, potassium and calcium, respectively, leached for the different treatments and the control. The means and standard errors for the concentration of the four cations in each leachate collected can be found in the appendices. There is no data for the 1<sup>st</sup> leachate of the control as there wasn't enough leachate collected to do the necessary measures.

From the figures below, it appears that apart from calcium, cations seem to have a similar tendency and rate of leaching between treatments. The sodium, magnesium and potassium curves have a sharp and sustained decrease up to the 2<sup>nd</sup> leachate with a slightly less sharp one until the 4<sup>th</sup>



leachate for all the treatments. The 100% - SR treatment appears to be the one that leaches the most cations.

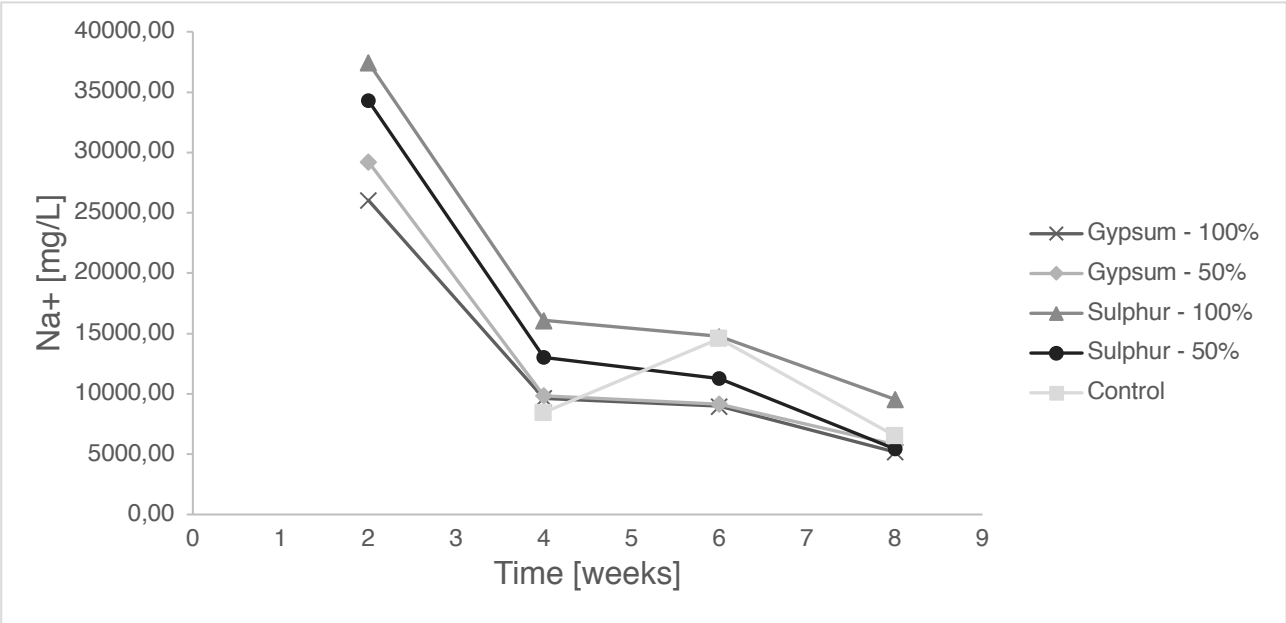


Figure 11: Evolution of the sodium concentration in the leachates collected during the 8 weeks for treatment 100% - GR, 50% - GR, 100% - SR, 50% - SR and the control.

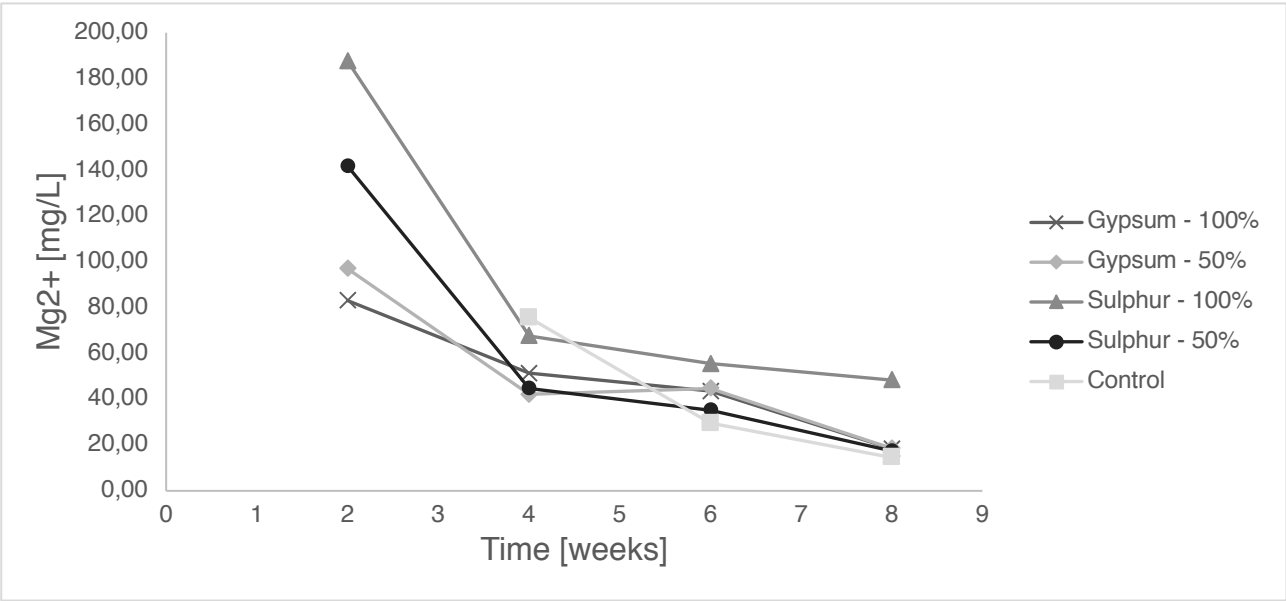


Figure 12: Evolution of the magnesium concentration in the leachates collected during the 8 weeks for treatment 100% - GR, 50% - GR, 100% - SR, 50% - SR and the control.

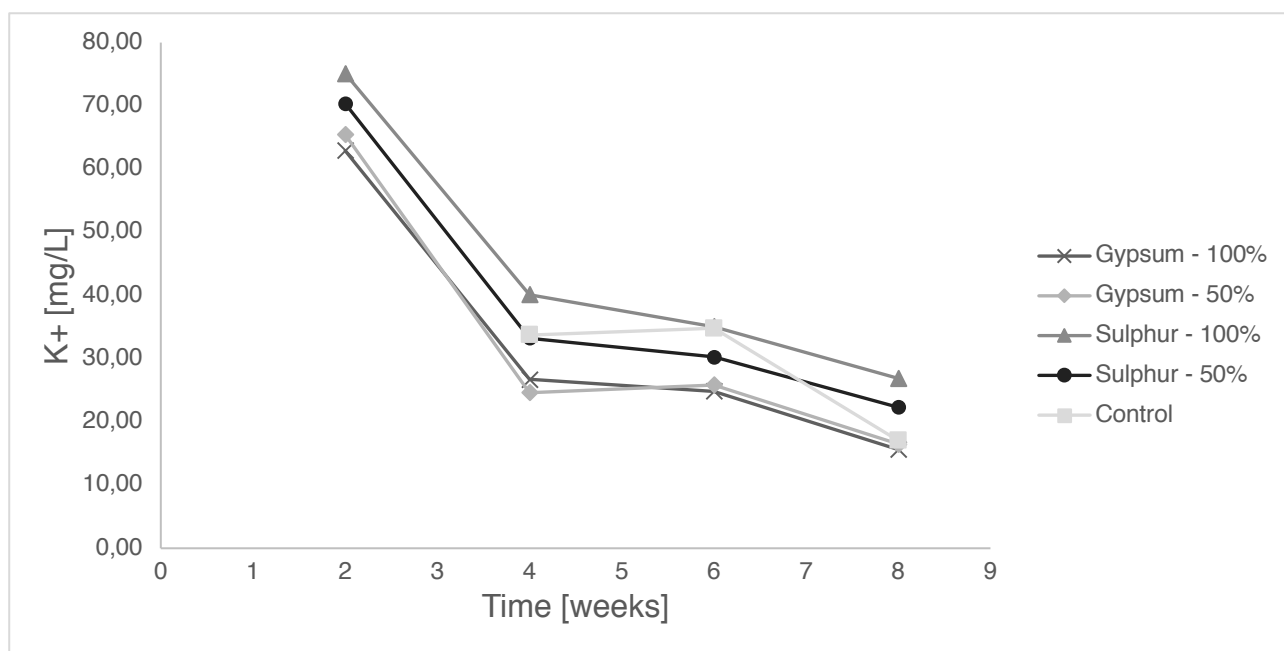


Figure 13: Evolution of the potassium concentration in the leachates collected during the 8 weeks for treatment 100% - GR, 50% - GR, 100% - SR, 50% - SR and the control.

The calcium curves are quite different from the curves of the other cations. After the second leachate there is a separation between the gypsum treatments and the sulphur treatments. The sulphur treatments tend to keep the same decreasing concentration tendency, but the gypsum treatments have peaks. The 100% - GR suddenly peaks at the second leachate, probably due to the source of calcium the gypsum brings. For the 50% - GR treatment the peak appears at the 3<sup>rd</sup> leachate. These peaks could possibly mean that some of the calcium were leached before they had a chance to replace the exchangeable sodium.

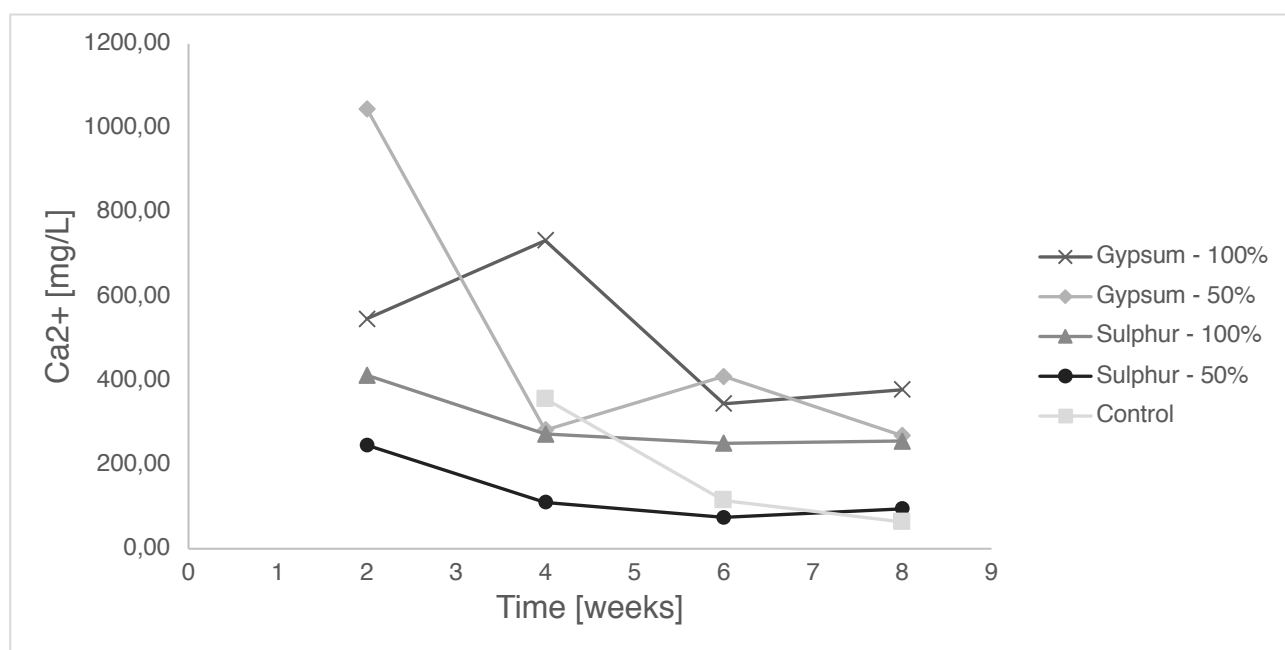


Figure 14: Evolution of the calcium concentration in the leachates collected during the 8 weeks for treatment 100% - GR, 50% - GR, 100% - SR, 50% - SR and the control.

Table 8 shows the means and standard errors of the total cumulated concentration of the different cations leached for each treatment and the result of Duncan's test ( $p < 0.05$ ). The concentrations of cations in the first leachates were not taken into account due to missing data for the volume of leachate collected for some of the pots.

The 100% - GR treatment was the only significantly different from other treatments and the control for calcium. As for the other cations the 100% level for both amendments were the ones which leached the most total quantity and were always significantly different from the control. Results are quite similar whether it is the sulphur treatment or the gypsum treatment for the same level doses (100% or 50%).

This demonstrates that most of the treatments did improve slightly the saline conditions of the soil. The results of these cumulative quantities also potentially show that sulphur is as efficient as gypsum. The significant difference for calcium with the 100% - GR treatment is probably due to the fact that gypsum brings a direct source of calcium whereas for sulphur the source is indirect. Indeed, as previously explained, when sulphur is applied it dissolves the calcite already present in the soil that will then act as a source of calcium to replace the sodium. As previously said, the first leachate isn't taken into the calculation here, if it were, higher quantities of cations leached would've been found and maybe more significant differences.

Table 8: Means (first row in cell) and standard errors (second row in cell) of the total cumulated quantity of cations leached for the different treatments. Duncan's test at  $p < 0.05$ , values with different letters differed significantly.

Treatment	Ca [mg]	Mg [mg]	K [mg]	Na [mg]
100% - GR	277.75	23.31	13.42	4798.0
	$\pm 111.26$ a	$\pm 4.66$ a	$\pm 2.66$ a	$\pm 730.6$ a
50% - GR	104.56	12.96	7.81	2892.3
	$\pm 41.61$ b	$\pm 6.80$ bc	$\pm 3.48$ b	$\pm 1343.9$ c
100% - SR	78.71	17.26	10.69	4395.6
	$\pm 18.59$ b	$\pm 5.38$ ab	$\pm 0.82$ ab	$\pm 556.3$ ab
50% - SR	22.34	9.67	8.19	3051.1
	$\pm 5.99$ b	$\pm 3.26$ bc	$\pm 2.31$ b	$\pm 1073.1$ bc
Control	23.55	6.07	7.28	3046.8
	$\pm 0.57$ b	$\pm 0.42$ c	$\pm 1.48$ b	$\pm 619.2$ bc

Figure 15 shows the evolution of the SAR of each leachate collected over time.

Final SAR values were of 368.13, 482.98, 774.25, 724.77 and 1043.54 for the 100% - GR, 50% - GR, 100% - SR, 50% - SR treatments and control respectively. All the final values for the

different treatments are lower than the one of the control. A separation can be observed between the sulphur treatments and the gypsum treatments, with higher values of SAR for the sulphur ones. The lower SAR for the gypsum treatments compared to the sulphur treatment can be explained by the significant difference found for calcium concentration between the two types of amendments (table 6). Indeed, a much higher calcium content can be found in the soil which were treated with gypsum, bringing an extra source of calcium, therefore reducing the SAR.

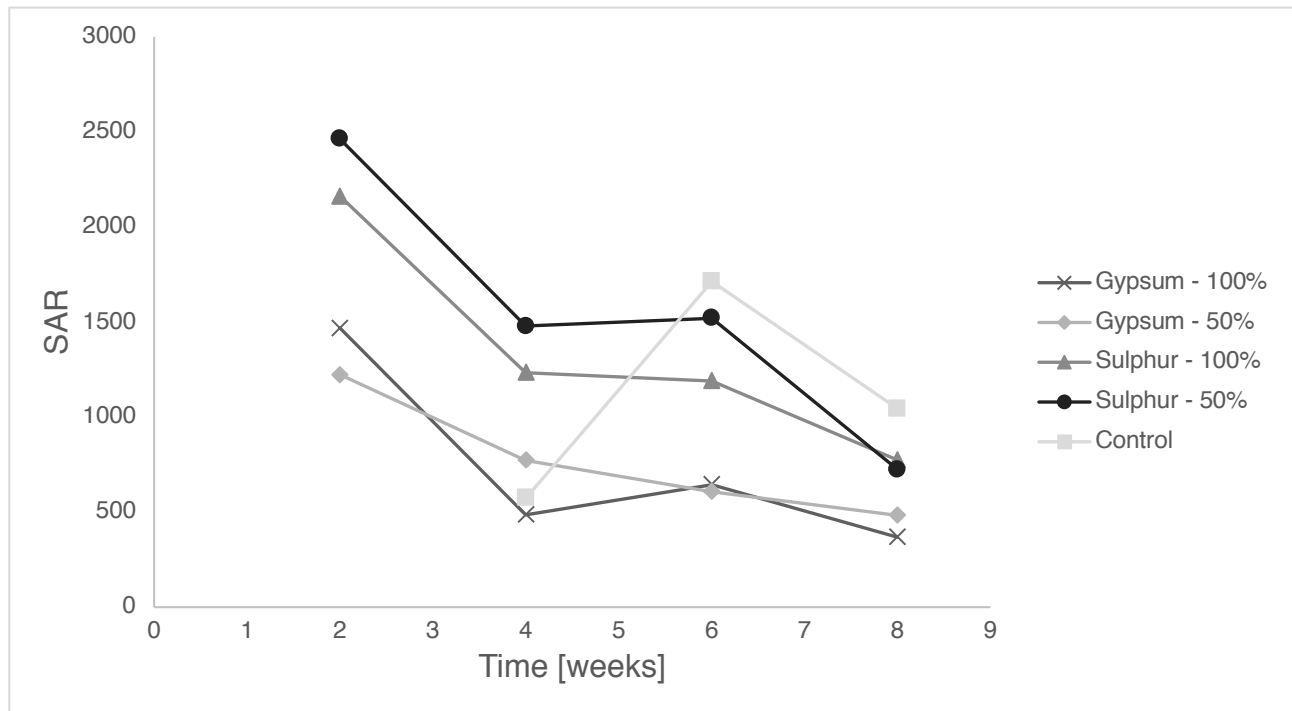


Figure 15: Evolution of the SAR of the leachates over time.

Figure 16 and 17 show the evolution of pH and electrical conductivity of the four collected leachates for each treatment.

Final pH values of the leachates for treatment 100% - GR, 50% - GR, 100% - SR, 50% - SR and the control were of 8.27, 8.30, 8.24, 8.74 and 9.11, respectively. The overall tendencies for all treatments is a slight decrease of pH over time, except for the 100% - SR treatment which has a sharp decrease from the 3<sup>rd</sup> to the 4<sup>th</sup> leachate, this might be due to the oxidisation of sulphur to form  $H_2SO_4$  which is what is to be expected but the fact that this drastic decrease did not occur for the 50% - SR is peculiar. An analyse of  $SO_4^-$  would've been interesting to see how the sulphur had reacted to confirm that assumption.

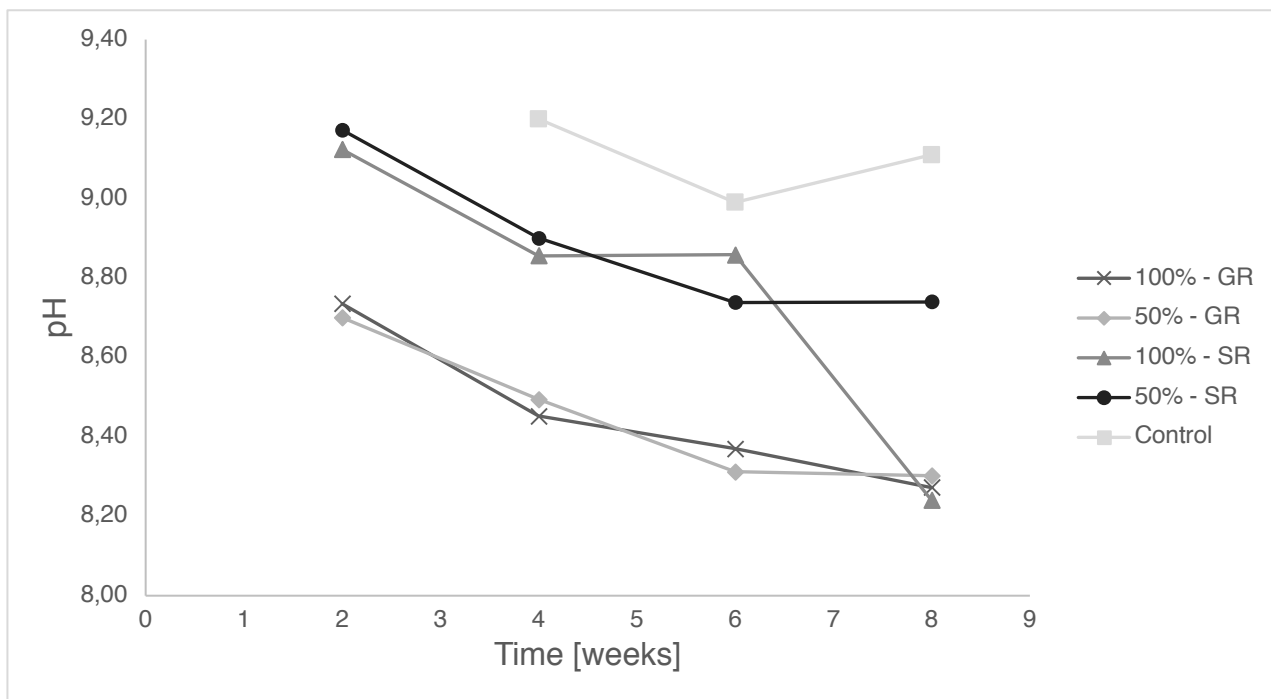


Figure 16: Evolution of the pH of the leachates collected over time for the 100% - GR, 50% - GR, 100% - SR and 50% - SR treatments

Final EC values for treatment 100% - GR, 50% - GR, 100% - SR, 50% - SR and the control were of 12.45, 16.26, 21.42, 8.24 and 11.5 \*10<sup>3</sup>  $\mu$ S/cm, respectively. For all treatments, EC drops drastically between the first and second leachates, after that the decrease between each leachate is softer. It is of course no surprise that the EC tendency is similar to the one of the cations concentrations since EC is correlated to the salt content.

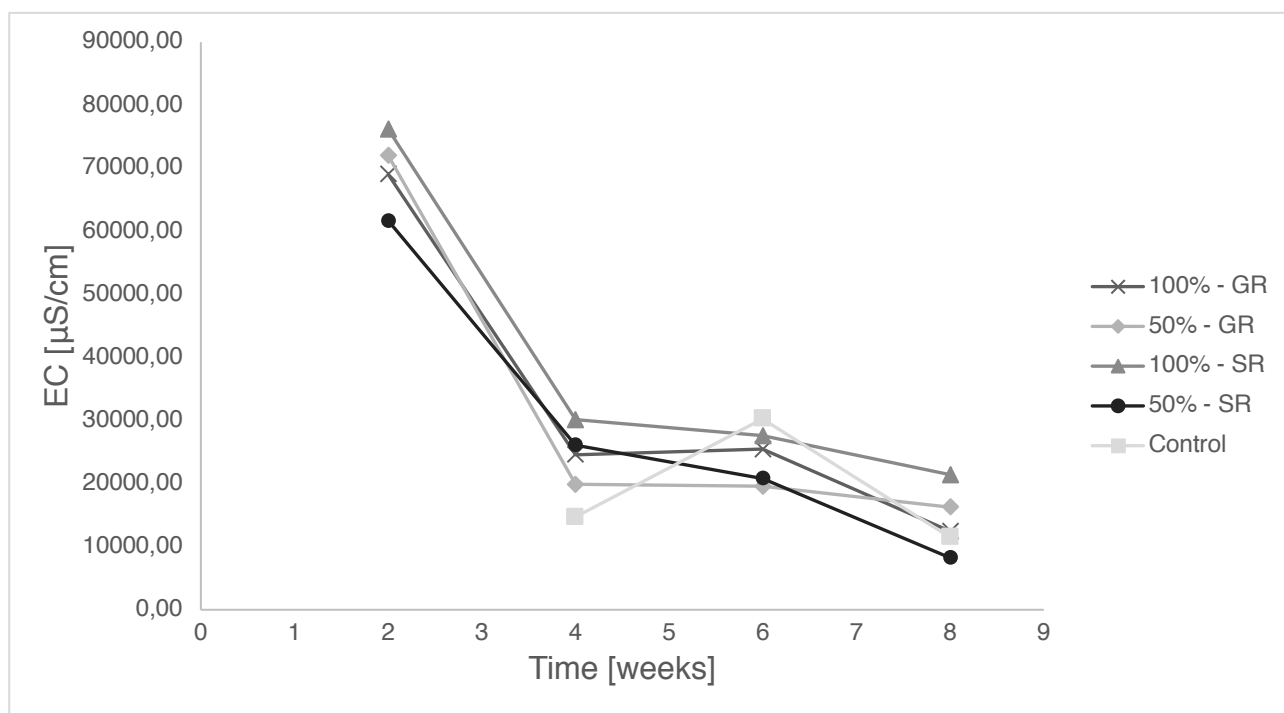


Figure 17: Evolution of the electrical conductivity of the leachates collected over time for the 100% - GR, 50% - GR, 100% - SR and 50% - SR treatments.

Treatments therefore did improve the saline and sodic conditions of the soil since part of the cations were leached. Let's recall the two first hypotheses put in place:

- H1: There is an optimal dose of gypsum/sulphur to reclaim the saline-sodic soils of the High Valley of Cochabamba.
- H2: Sulphur has a similar effectiveness as gypsum in terms of soil chemical properties, to reclaim the saline-sodic soils of the High Valley of Cochabamba.

As we have seen, significant differences were found between the two different doses for both amendment. This would mean that applying 100% of the GR/SR or 50% would impact significantly the final results. Other doses could be considered between 50% and 100% of the requirements to see if the 100% dose is still necessary. In this case, the first hypotheses can't yet be confirmed. The sulphur treatments compared to the gypsum treatments did not differ significantly either, in terms of total cumulated cations leached and the EC of the 4<sup>th</sup> leachate. This would validate the hypothesis stating that sulphur is as efficient as gypsum. However, leaching of cations isn't the only parameter to take into account for the reclamation of salt-affected soils.

### 5.2.2 Soil analysis

Table 9 presents the results of Dunnett's test performed to evaluate the efficiency of the chemical amendments treatments.

When comparing the soil properties after the chemical treatments with the control only the pH of all treatments was significantly different at  $p < 0.0001$ . Calcium content differed significantly at  $p < 0.0001$  and final ESP differed significantly at  $p < 0.001$  only for the 100% - GR treatment. Calcium content for 50% - GR has a high standard error, this is due to the fact that one of the pots had a calcium content of 2.79 cmol<sub>c</sub>/kg when the four other pots had a calcium content ranging from 8 to 11 cmol<sub>c</sub>/kg. Should this value had been excluded, a significant difference between the calcium concentration of the 50% - GR and the control might have been observed. Potassium content also differed significantly at  $p < 0.01$  for the 100% - SR treatment.

Table 9: Comparison between soil properties after the chemical amendments treatments and the control with Dunnett's test. Means (first row) and standard errors (second row) of the chemical amendments treatments and the control. Significance codes: “\*\*\*” :  $p < 0.000$ , “\*\*” :  $p < 0.001$ , “\*” :  $p < 0.01$ , “.” :  $p < 0.05$ .

Treatment	pH	EC [ $\mu\text{S}/\text{cm}$ ]	CEC [ $\text{cmol}/\text{kg}$ ]	Ca [ $\text{cmol}/\text{kg}$ ]	Mg [ $\text{cmol}/\text{kg}$ ]	K [cmol/kg]	Na [ $\text{cmol}/\text{kg}$ ]	ESP [%]
100% - GR	8.56	1254.7	6.06	14.24	0.73	0.28	5.80	27.11
	$\pm 0.09$ ***	$\pm 69.2$	$\pm 0.96$	$\pm 0.42$ ***	$\pm 0.16$	$\pm 0.06$	$\pm 1.97$	$\pm 7.45$ **
50% - GR	8.77	1172.7	5.37	8.68	0.73	0.30	6.89	43.43
	$\pm 0.03$ ***	$\pm 195.4$	$\pm 0.93$	$\pm 3.43$	$\pm 0.09$	$\pm 0.07$	$\pm 0.76$	$\pm 13.26$
100% - SR	8.51	1367.1	4.09	5.96	0.72	0.37	7.96	52.98
	$\pm 0.02$ ***	$\pm 211.9$	$\pm 0.82$	$\pm 0.17$	$\pm 0.12$	$\pm 0.05$ *	$\pm 0.65$	$\pm 2.12$
50% - SR	8.75	1264.7	4.72	5.77	0.70	0.23	9.29	58.02
	$\pm 0.03$ ***	$\pm 278.6$	$\pm 1.34$	$\pm 0.17$	$\pm 0.10$	$\pm 0.13$	$\pm 0.79$	$\pm 2.18$
Control	9.18	1249.1	5.72	6.06	0.66	0.17	7.76	52.66
	$\pm 0.01$	$\pm 28.3$	$\pm 0.96$	$\pm 0.07$	$\pm 0.012$	$\pm 0.03$	$\pm 1.75$	$\pm 6.42$

The higher content in calcium for the 100% - GR can be explained by the fact that gypsum brings an external source of calcium. Also, it was found that the 100% - GR treatment was the only one to decrease the ESP significantly. This could help to validate the first hypothesis concerning the optimal dose, but further analyses should be done. It is also important to recall here that the control also went through a leaching process. Even though no amendments were applied, part of the cations might have been leached explaining the lack of difference observed between the treatments and the control. Furthermore, when we take a look at the scientific literature, similar trials were carried out on a longer period of time and giving more significant results (Qadir *et al.*, 1996, Ahmad *et al.*, 2015). Should the treatments had gotten more time to take effect, particularly for the sulphur treatments, more significant differences between the treatments and the control might have been observed.

### Mass balance

The variables used for the mass balance and the result of the mass balance for a pot are shown in table 10. The final calcium quantity in the soil measured by the laboratory are also given to compare the result.

Table 10: Values of variables used to calculate the mass balance and the result of the mass balance.

Calcium quantity	100% - GR	50% - GR
Initial [mg/pot]	2004	2004
Gypsum [mg/pot]	4040	2020
Leached [mg/pot]	325	205
Mass balance [mg/pot]	5719	3819
Final [mg/pot]	5696	3472

Final quantity and mass balance for the 100% - GR are almost the same but there is a difference of 347 mg for the 50% - GR. This might be due to the higher standard error of the final concentration of the calcium in the soil final for the 50% - GR (table 9).

## 5.3 Phytoremediation

In this section, the results of the shoot and soil analysis of the phytoremediation treatments will be presented and discussed.

### 5.3.1 Shoot Analysis

The means and standard errors of dry matter weight are presented in table 11.



Table 11: Means and standard errors of the dry matter weight of the phytoremediation treatments and the control.

Treatment	Dry matter [g/pot]
3-4 months plants	0.3082 ± 0.0334
1-2 months plants	0.1225 ± 0.0260
Control	0.3080

Figure 18 corresponds to the differences between the concentration of the cations in the shoots of the experimental Q'awchi plants grown in the saline-sodic soil 1 and the ones in the shoots of a Q'awchi grown in the original soil used at the Research Centre for Halophytes in Bolivia. A negative value means that the concentration in the shoots has decreased following the cultivation in the saline-sodic soil 1.

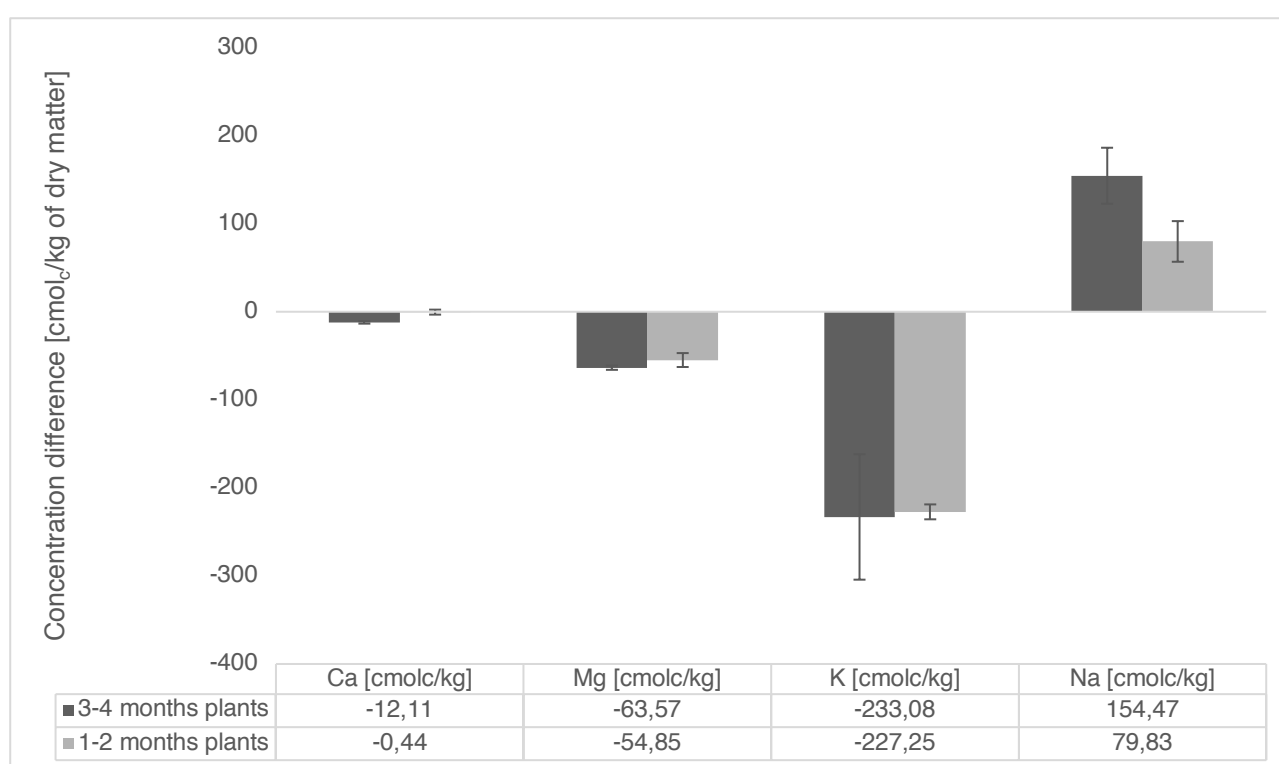


Figure 18: Difference between final and initial concentration of cations in the shoots of *Suaeda foliosa*.

The only cation accumulation in the shoots that can be seen is the sodium and was evaluated at 154.47 cmol<sub>c</sub>/kg of dry matter and 79.83 cmol<sub>c</sub>/kg for the plants of 3-4 months and 1-2 months respectively. High negative values of potassium for both treatments of -233.08 and -227.25 cmol<sub>c</sub>/kg can be observed.

The higher accumulation of sodium in the 3-4 months can be explained by the fact that these plants were older, their roots had therefore developed more compared to the 1-2 months plants. Nevertheless, for the two different age plants part of the roots were damaged during the transfer from their original soil to the saline-sodic one, this might have affected the absorption capacity of the plants. A high negative value for potassium can be explained by the behaviour of saline-sodic soils

where excessive sodium interferes with potassium uptake (Grattan & Grieve, 1999). Moreover, the concentrations in calcium, magnesium, potassium and sodium of the Q'awchi original soil mentioned in section 5.1, table 5 were of 9.33 cmol<sub>c</sub>/kg, 1.40 cmol<sub>c</sub>/kg, 1.52 cmol<sub>c</sub>/kg and 0.92 cmol<sub>c</sub>/kg. Whereas in the saline-sodic soil 1 they were of 5.01 cmol<sub>c</sub>/kg, 0.53 cmol<sub>c</sub>/kg, 0.17 cmol<sub>c</sub>/kg and 13.11 cmol<sub>c</sub>/kg. Concentrations of calcium, magnesium and potassium were therefore higher in the original soil and the concentration of sodium was lower. This could explain the tendencies represented in figure 18. So, there is in fact an accumulation of sodium in the shoots, but a longer trial should be pursued to evaluate more precisely the phytodesalination capacity of *Suaeda foliosa*. Roots should've been analysed too as this is how the plant absorbs the ions, higher concentrations of sodium could've been found.

### 5.3.2 Soil Analysis

Soil analyses for the phytoremediation treatments with the control are presented in table 12.

No significant differences between the treatments and the control were found for either of the variables except for the electrical conductivity with a p-value < 0.01 for the plants of 1-2 months and with a p-value < 0.001 for the 3-4 months plants.

The EC differed from the control but was in fact higher for the phytoremediation treatments. This is probably due to the way the soil sampling was done, as two simple cores from the top of the pot down to  $\frac{3}{4}$  were taken. In some studies reported in the literature, sampling was divided into two: one in the upper part (0 – 10 cm) and another one in a lower part (10-20 cm) of the pots. The two samples were compared, and it was shown that SAR, Na<sup>+</sup> and EC decreased in the higher horizon. This can be explained by the fact that the roots were mainly in the higher depths of the pots (Rhabi *et al.*, 2010, Jlassi *et al.*, 2013). A separation of the two depths would've been interesting.

Table 12: Comparison between soil properties of the phytoremediation treatments with the control with Dunnett's test. In each cell, first row corresponds to the mean and second row to the standard error. Significance codes: "\*\*\*\*" :  $p < 0.000$ , "\*\*\*" :  $p < 0.001$ , "\*\*" :  $p < 0.01$ , "." :  $p < 0.05$ .

Treatment	pH	EC [μS/cm]	CEC [cmol <sub>c</sub> /kg]	Ca [cmol <sub>c</sub> / kg]	Mg [cmol <sub>c</sub> / kg]	K [cmol <sub>c</sub> / kg]	Na [cmol <sub>c</sub> / kg]	ESP [%]
3-4 months plants	9.71 ±0.08	3599.1 ±421.1 **	8.05 ±2.15	5.45 ±0.15	0.59 ±0.02	0.24 ±0.01	20.14 ±2.75	76.03 ±2.65
1-2 months plants	9.64 ±0.13	2982.3 ±459.6 *	7.68 ±1.83	5.59 ±0.13	0.59 ±0.02	0.25 ±0.00	19.88 ±3.20	75.28 ±2.92
Control	9.69 ±0.00	1637.1 ±0.0	8.72 ±0.00	5.42 ±0.00	0.59 ±0.00	0.24 ±0.00	18.56 ±0.00	74.80 ±0.00

The third hypothesis, which stated that remediation with *Suaeda foliosa* would be effective to reclaim salt-affected soils in the high valley of Cochabamba therefore lacks results in order to be confirmed. Once more, phytoremediation trials are usually carried on a longer period (Zhao, 1991, Rabhi *et al.*, 2009). A two-month trial clearly wasn't long enough for *Suaeda foliosa* to prove its phytodesalination capacity.

## 5.4 Overall before/after treatments

Although when comparing the treatments with the control might seem as the treatments didn't have much effect, more differences can be observed when comparing the soil before and after treatments.

Table 13 presents the result of Duncan's test for the properties of the soil before and after treatments and the histograms given out by R studio can be found in the appendices.

Unfortunately, none of the treatments achieved to reduce the EC, pH and ESP down to the threshold values put in place by the USDA salinity classification for the soils to be considered normal. Nevertheless, some significant differences can be found between the soil before and after treatments particularly for the chemical treatments.

Table 13: Comparison between soil properties before and after treatments with the Duncan's test at  $p$ -value  $< 0.05$ . Means (first row) and standard errors (second row) of the soil before and after the different treatments. Values with different letters differ significantly.

Treatment	pH	EC [ $\mu$ S/cm]	CEC [cmol/kg]	Ca [cmol/kg]	Mg [cmol/kg]	K [cmol/kg]	Na [cmol/kg]	ESP [%]
3-4 months	9.71	3599.1	8.05	5.45	0.59	0.24	20.14	76.03
plants	$\pm 0.08$ a	$\pm 421.1$ a	$\pm 2.15$ a	$\pm 0.15$ c	$\pm 0.02$ ab	$\pm 0.01$ bc	$\pm 2.75$ a	$\pm 2.65$ a
1-2 months	9.64	2982.3	7.68	5.59	0.59	0.25	19.88	75.28
plants	$\pm 0.13$ a	$\pm 459.6$ b	$\pm 1.83$ a	$\pm 0.13$ c	$\pm 0.02$ ab	$\pm 0.00$ bc	$\pm 3.20$ a	$\pm 2.92$ a
100% - GR	8.56	1254.7	6.06	14.24	0.73	0.28	5.80	27.11
	$\pm 0.09$ c	$\pm 69.2$ c	$\pm 0.96$ ab	$\pm 0.42$ a	$\pm 0.16$ a	$\pm 0.06$ ab	$\pm 1.97$ d	$\pm 7.45$ d
50% - GR	8.77	1172.7	5.37	8.68	0.73	0.30	6.89	43.43
	$\pm 0.03$ b	$\pm 195.4$ c	$\pm 0.93$ b	$\pm 3.43$ b	$\pm 0.09$ a	$\pm 0.07$ ab	$\pm 0.76$ cd	$\pm 13.26$ c
100% - SR	8.51	1367.1	4.09	5.96	0.72	0.37	7.96	52.98
	$\pm 0.02$ c	$\pm 211.9$ c	$\pm 0.82$ b	$\pm 0.17$ c	$\pm 0.12$ a	$\pm 0.05$ a	$\pm 0.65$ cd	$\pm 2.12$ b
50% - SR	8.75	1264.7	4.72	5.77	0.70	0.23	9.29	58.02
	$\pm 0.03$ b	$\pm 278.6$ c	$\pm 1.34$ b	$\pm 0.17$ c	$\pm 0.10$ a	$\pm 0.13$ bc	$\pm 0.79$ c	$\pm 2.18$ b
Saline-Sodic	9.61	2982.1	5.08	5.01	0.53	0.17	13.11	69.66
Soil	$\pm 0.01$ a	$\pm 264.5$ b	$\pm 1.07$ b	$\pm 0.00$ c	$\pm 0.02$ b	$\pm 0.04$ c	$\pm 0.46$ b	$\pm 0.51$ a

Figures 19, 20 and 21 show the histograms of pH, EC and ESP, respectively, for all the treatments including the soil before treatment for a more visual representation.

#### A. pH

The pH has been found to be reduced significantly only for the chemical treatments compared to the soil before treatment. Soil pH was originally at 9.61 and was reduced down to 8.56, 8.77, 8.51 and 8.75 for the 100% - GR, 50% - GR, 100% - SR and 50% - SR respectively.

The two higher doses of chemical amendments have been found to be the most effective for a pH reduction. This may have been due to the higher leaching of the cations, particularly sodium and (bi)carbonates which are strong bases and increase the pH when they precipitate, forming  $\text{Na}_2\text{CO}_3$  or  $\text{NaHCO}_3$ . Furthermore, in the case of gypsum, the source of calcium brought could have replaced the exchangeable sodium, enabling it to precipitate and form the neutral salt  $\text{Na}_2\text{SO}_4$  (Legros, 2007, Abdel-Fattah, 2012). It has been found that in the case of sodic calcareous soils, pH and ESP are proportional (Gupta *et al.*, 1981, Abrol *et al.*, 1980). Although the calcareous characteristic of the soil wasn't measured here, we can see that when a pH reduction occurred an ESP reduction was also observed.

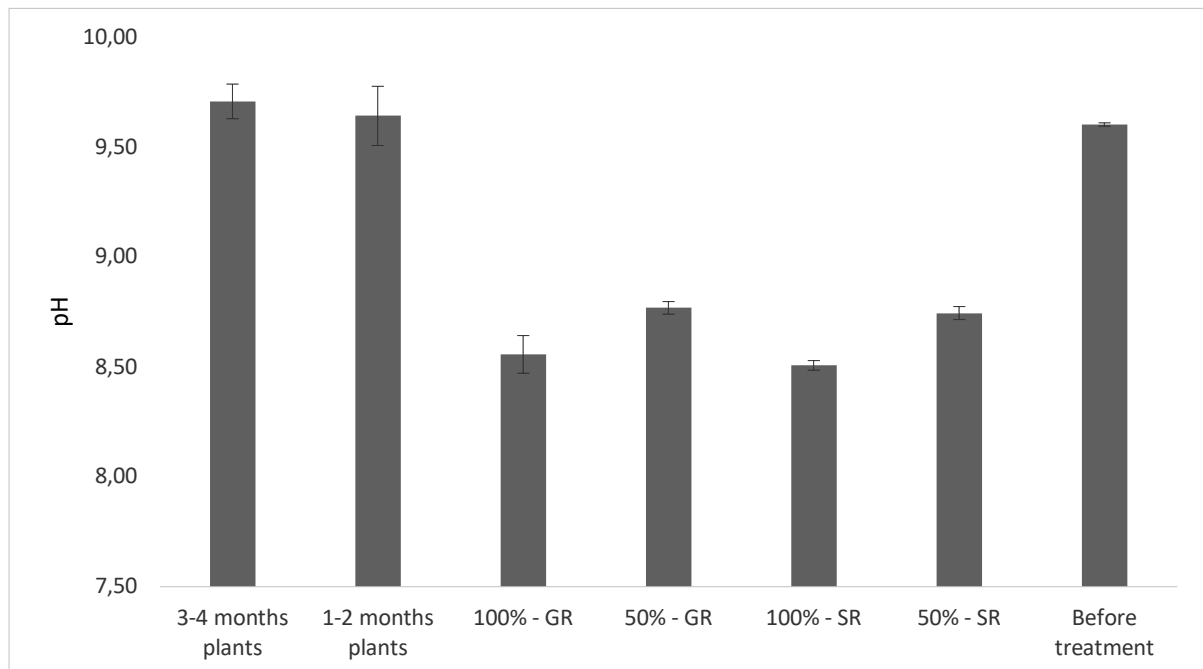


Figure 19: Histogram with error bars of soil pH before and after treatments.

## B. EC

For all the chemical treatments a significant difference was found when comparing with the soil before treatment. Soil for treatments at 100% - GR, 50% - GR, 100% - SR and 50% - SR had EC values of 1254.73, 1172.73, 1367.13, 1264.73  $\mu\text{S}/\text{cm}$  respectively (Table 12). EC was reduced by more than 50% from its initial value for all the chemical treatments. When divided by 7.62 the factor previously mentioned for the conversion of  $\text{EC}_e$  into  $\text{EC}_{1:5}$ , the threshold value of  $\text{EC}_e$  fixed by the USDA is equal to 525  $\mu\text{S}/\text{cm}$ . So even though EC were considerably reduced, no treatment managed to reduce the EC for them to be considered non-saline.

On the other hand, phytoremediation did not have similar positive results. On the contrary, the 3 – 4 months plants treatment increased EC significantly compared to the soil before treatment, reaching a value of 3599.13  $\mu\text{S}/\text{cm}$ , corresponding to an increase of 20%. As for the 1-2 months plants treatment the final EC value did not change significantly. As previously explained in section 5.3.2., this might be due to the soil sampling technique.

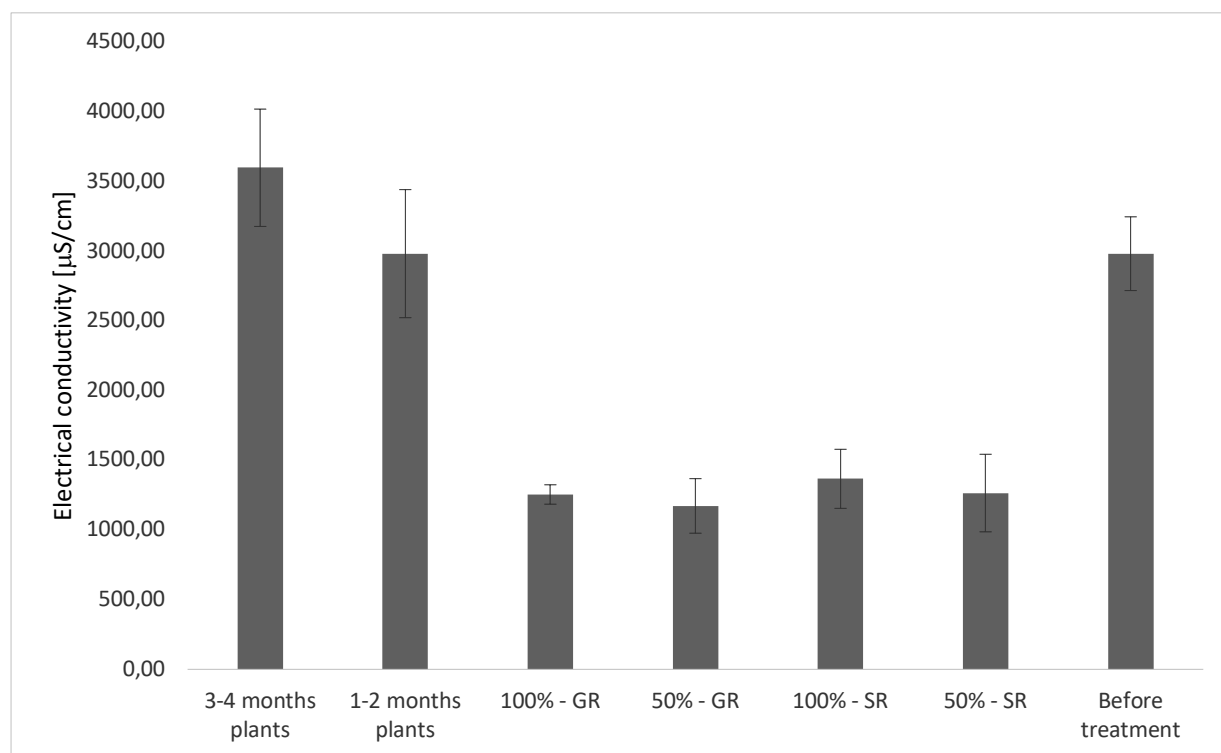


Figure 20: Histogram with error bars of the electrical conductivity [ $\mu\text{S}/\text{cm}$ ] before and after treatments.

## C. ESP

Again here, ESP decreased only for the chemical treatments and differences are observed between the sulphur treatments and the gypsum treatments. Final values for the 100% - GR, 50% - GR, 100% - SR and 50% - SR treatments were of 27.11%, 43.43%, 52.98%,

58.02% respectively. The 100% - GR treatment managed to reduce the most the ESP, but still not down to 15%. This is contradictory since the gypsum requirement were calculated as to decrease ESP down to 15%. When looking at the values used for the calculation of the gypsum requirement, they seemed to have been over-evaluated compared to the ones given by the external lab. CEC was evaluated at 20 cmol<sub>c</sub>/kg instead of 5.08 cmol<sub>c</sub>/kg and ESP was estimated at 75% instead of 70%. But the higher values used would've given a higher gypsum requirement than needed. This suggests that in the case of our soils, higher gypsum requirement should've been given in order to reach the ESP value of 15%. What is also interesting to observe here is the fact that as previously studied, the chemical treatments did not differ significantly from their control but did when comparing the soil before/after treatment, except for the 100% - GR treatment. The question then arises whether it was really necessary to put amendments, or could the leaching process be sufficient with the soils treated here.

As for the phytoremediation treatments no ESP reduction were observed. Even if not significantly, they actually increased ESP up to 76 and 75% compared to the initial soil which had a value of 70%. This can be explained by the increased content in sodium. Indeed, a significant increase in sodium concentration can be observed for both phytoremediation treatments. Sodium present on the exchange complex might have passed into the solution but were not absorbed by the plant roots. Furthermore, there was a concentration of 25 mg/L of sodium in the irrigation water. This might have increased the sodium concentration for the phytoremediation treatment since they were not leached like the chemical amendment treatments were.

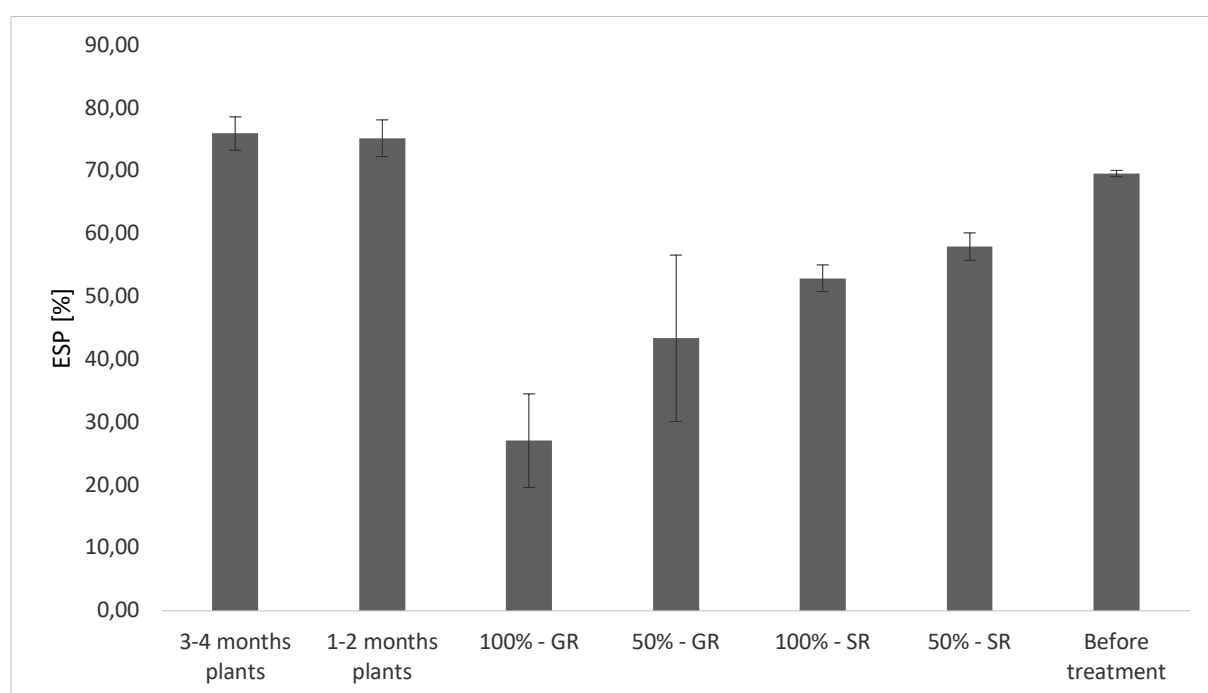


Figure 21: Histogram with error bars of the ESP [%] before and after treatments.

#### D. Concentration of cations

Figure 22 shows the difference between the concentration of calcium, magnesium, potassium and sodium before and after treatments in the soil. A positive value means that the initial concentration was higher than the one after treatment.

Magnesium and potassium had almost the same initial and final values with a difference of only under 1 cmol<sub>e</sub>/kg. The only cation for which a positive value can be found is the sodium for all the chemical treatments. In order of decreasing concentration, 100% - GR > 50% - GR > 100% - SR > 50% - SR with values of 7.32, 6.23, 5.15 and 3.83 cmol<sub>e</sub>/kg respectively. On the other hand, final concentration of calcium exceeded initial concentration by 9.23 cmol<sub>e</sub>/kg for the 100% - GR treatment and by 3.67 cmol<sub>e</sub>/kg for the 50% - GR treatment. This can be explained by the fact that gypsum brings an external source of calcium when the sulphur treatment does not. Increased concentrations of sodium in the soil were also found after the phytoremediation treatments. This was not the expected result as the plants are supposed to accumulate the sodium. After treatment, for the plants of 3-4 months final concentration was 7.02 cmol<sub>e</sub>/kg higher and for the plants of 1-2 months 6.77 cmol<sub>e</sub>/kg higher. Again, this might be due to the soil sampling technique, as explained in section 5.3.2..

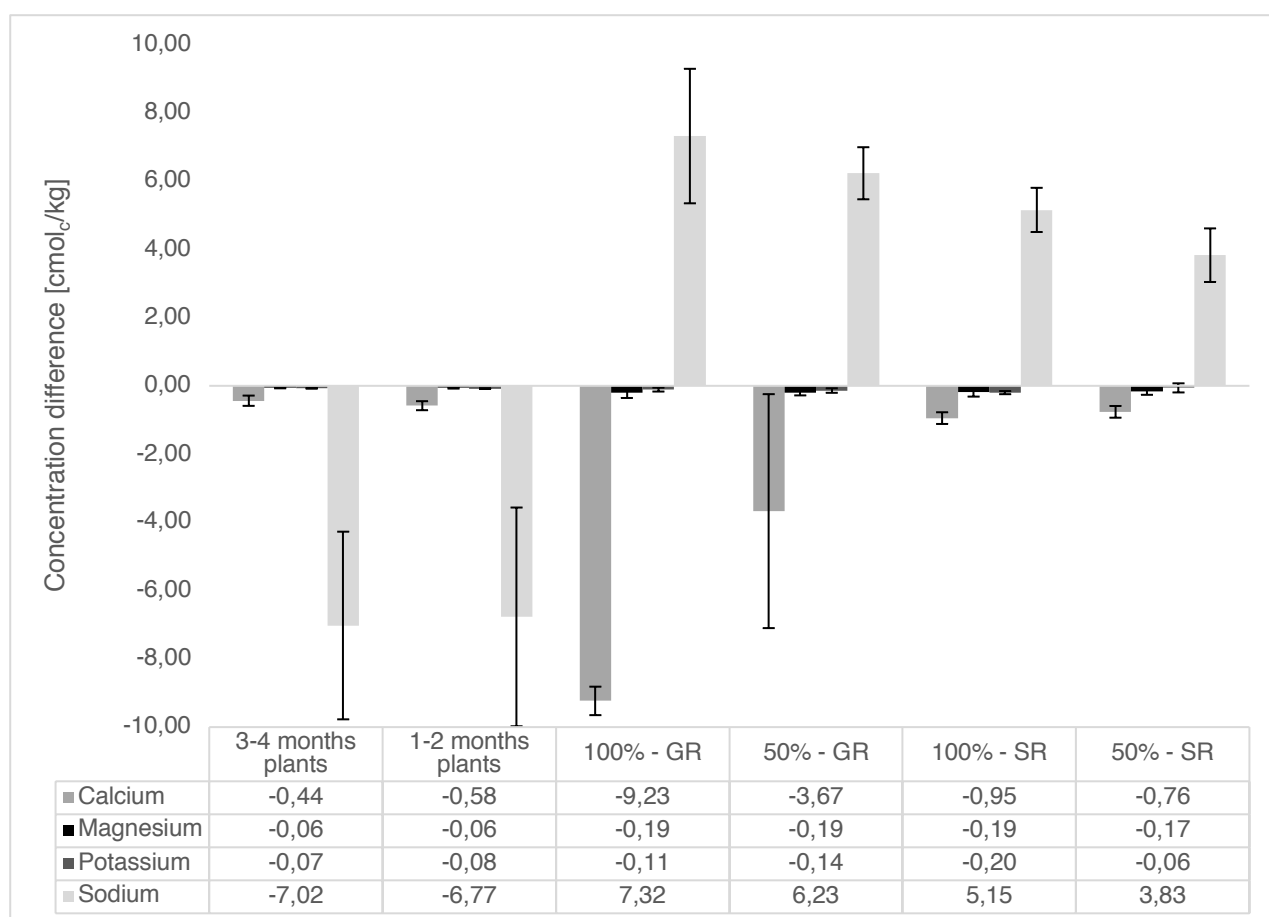


Figure 22: Difference between concentration in cmol<sub>e</sub>/kg of soil of the Calcium, Magnesium, Potassium and Sodium in the soil before and after treatments.



Table 14 shows on first hand, the correlation between the accumulated cations in the shoots and the difference between cations in the soil before/after treatments. On a second hand it shows the correlation between the cumulated cations leached and the difference between cations remaining in the soil before/after treatments. Positive correlation can't be considered good since a negative correlation would be the desired result. As we can see, none of the correlations are significant except for the calcium for the chemical amendments treatments. This is likely due to the calcium content of gypsum which influenced the amount of calcium leached.

*Table 14: Correlation coefficient (r) and p-values of cations in the shoots vs cations removed from soil for the phytoremediation treatments and cations in the leachates vs cations removed from soil for the chemical amendments treatments.*

Treatment		Phytoremediation				Chemical amendments			
Cation correlation		Ca soil vs Ca shoots	Mg soil vs Mg shoots	K soil vs K shoots	Na soil vs Na shoots	Ca soil vs Ca leach	Mg soil vs Mg leach	K soil vs K leach	Na soil vs Na leach
r		0.51	-0.032	0.59	-0.02	-0.82	0.011	-0.21	0.19
p		0.13	0.93	0.075	0.96	9e-06	0.96	0.36	0.43

## 5. Conclusions and recommendations

To conclude let's recall our three main objectives:

- The determination of an optimal dose for the reclamation of saline-sodic soils of the High Valley of Cochabamba in Bolivia with gypsum and sulphur respectively.
- The understanding of how the chemical amendments affect the chemical properties of the soil, in particular sulphur to establish if it's as effective as gypsum in the reclamation of salt-affected soils.
- The evaluation of the phytodesalination capacity of *Suaeda foliosa*.

To answer the two first objectives, a brief review of the results should be done. It has been found that all the chemical treatments did improve the saline-sodic conditions of the soil. Electrical conductivity (EC), Exchangeable Sodium Percentage (ESP) and pH were effectively reduced for all the chemical treatments when compared with the soil before treatments. On the other hand, less significant differences were found when comparing the chemical treatments between them. Indeed, the EC reduction didn't differ between the treatments. The same pH reduction was found for the same levels of requirement of gypsum and sulphur respectively. However, the highest decrease of ESP was found for the 100% - GR followed by the 50% - GR and the two sulphur treatments. This suggests that there would be a difference between the doses applied for the gypsum. On the other hand, the two sulphur treatments did not differ significantly which could suggest that a 100% or 50% of SR would have the same effect. Furthermore, when comparing the chemical treatments with the control which went through a simple leaching process, no significant differences were found apart for a pH reduction. The chemical amendments might not have had enough time to react particularly the sulphur which needs to go through an oxidising phase before it can actually have a significant effect on the reclamation of salt-affected soils. In order to correctly answer the two first objectives, a longer-term experimentation should be carried out and an intermediate level of the requirement, particularly for the gypsum, should be tried out.

For the third objective, results were less concluding. Even though there was an accumulation of sodium in the shoots as expected, the difference between the ESP, EC, pH and sodium content of the soil before and after treatment weren't as satisfactory. Indeed, all of these variables increased in the soil after the two months phytoremediation treatments. A long-term experiment of at least 6 months should be carried out to evaluate correctly the phytodesalination capacity of *Suaeda foliosa*. Furthermore, there should be a separation between the depths at which the roots are and the rest of the soil to evaluate more accurately the absorption of the cations, particularly sodium.

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# Appendices

## **Appendix A: Leachate and shoot analysis protocol**

Different dilutions of the leachates, in 25 ml flasks, had to be done in order to be able to measure the samples' concentration in the scales available depending on the element analysed. Depending on the scale used different standard solutions had to be made to calibrate the device. The results of the different samples obtained were multiplied by their corresponding factor of dilution.

### Phytoremediation

- Potassium

A scale of 15 to 500 ppm was used and standard solutions for the calibration curve were prepared using a stock solution of K at 1000 ppm. Factors of dilution were of 0 or 25 depending on the sample measured.

- Magnesium

A scale of 0.02 to 5 ppm was used. Standard solutions were prepared with a stock solution of Mg at 100 ppm and Strontium Chloride Hexahydrate ( $\text{SrCl}_2 \cdot 6\text{H}_2\text{O}$ ) at 10000 ppm. Factors of dilution were of 100 or 250 depending on the sample measured.

- Calcium

A scale of 0.01 to 3 ppm was used with a factor of dilution of 250. Standard solutions were prepared using stock solution of Ca at 100 ppm and Potassium chloride (KCl) at 10000 ppm.

- Sodium

A scale of 0.01 to 2 ppm was used for the measures of the sodium with a factor of dilution of 1250. A stock solution of Na at 1000 ppm and Potassium chloride at 10 000 ppm was used to prepare the standard solution.

### Chemical amendments treatments

- Potassium

Two different scales were used for this element, the first for concentration between 1 and 6 ppm and the second one for concentration between 15 and 500 ppm. Standard solutions for the calibration curve were prepared using a stock solution of K at 100ppm for the 1-6 ppm scale and of K at 1000 ppm for the 15-500 ppm scale. Factors of dilution were of 0, 5 or 10 depending on the sample measured.

- Magnesium

A scale of 0.02 to 5 ppm was used. Standard solutions were prepared with a stock solution of Mg at 100 ppm and Strontium Chloride Hexahydrate ( $\text{SrCl}_2 \cdot 6\text{H}_2\text{O}$ ) at 10000 ppm. Factors of dilution were of 100 or 500 depending on the sample measured.

- Calcium

A scale of 0.01 to 3 ppm was used. Standard solutions were prepared using stock solution of Ca at 100 ppm and Potassium chloride (KCl) at 10000 ppm. Factors of dilution were of 250 or 500.

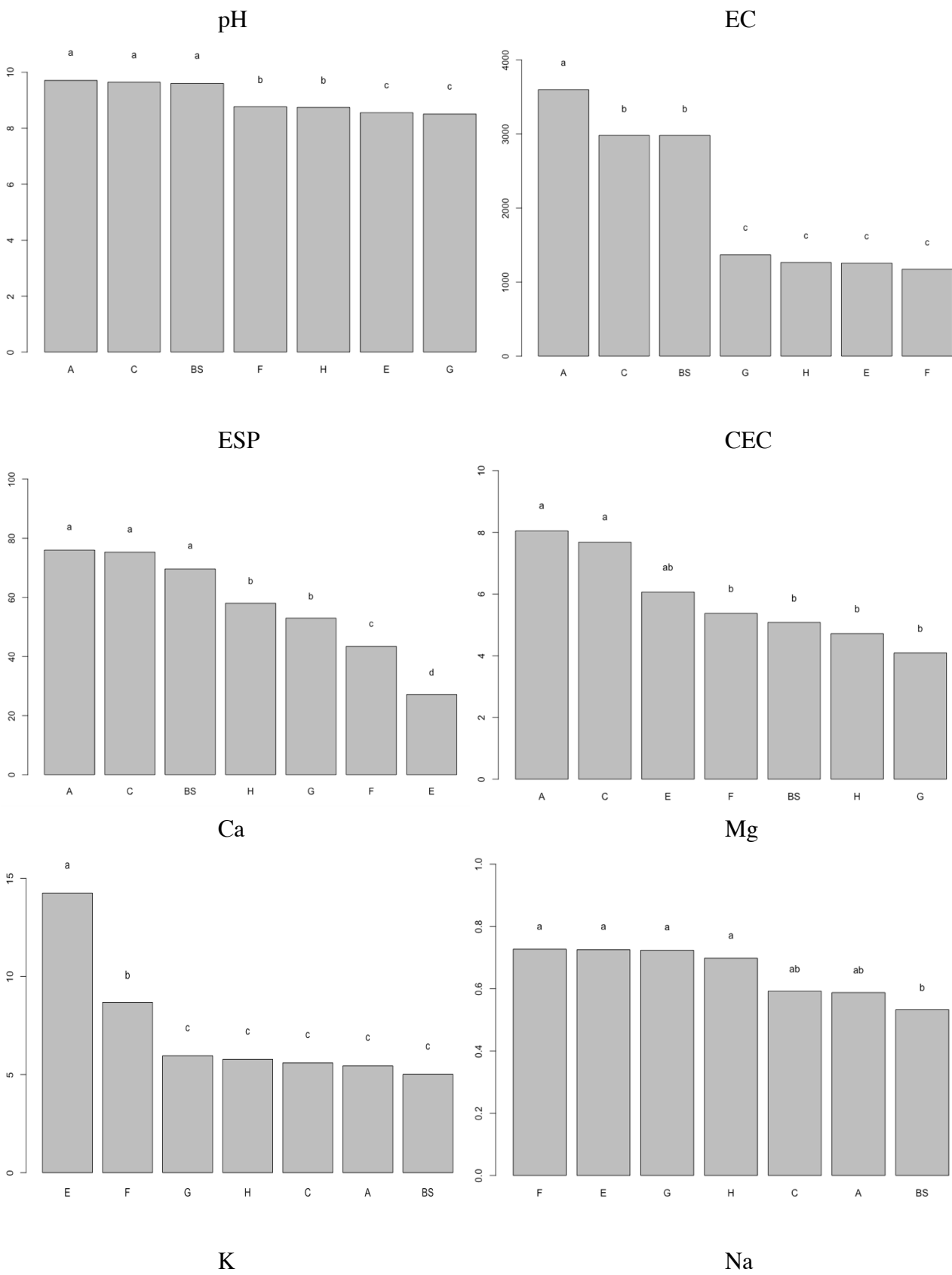
- Sodium

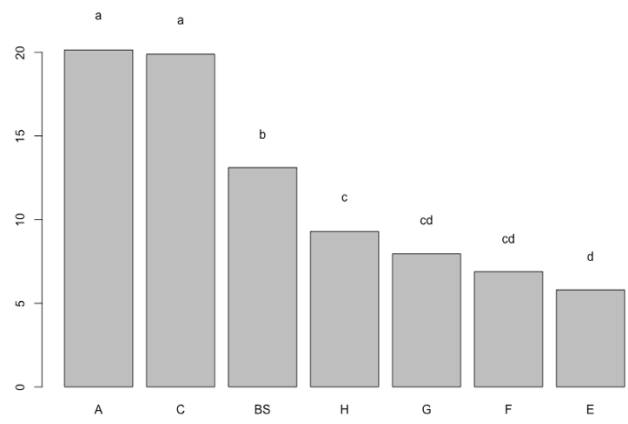
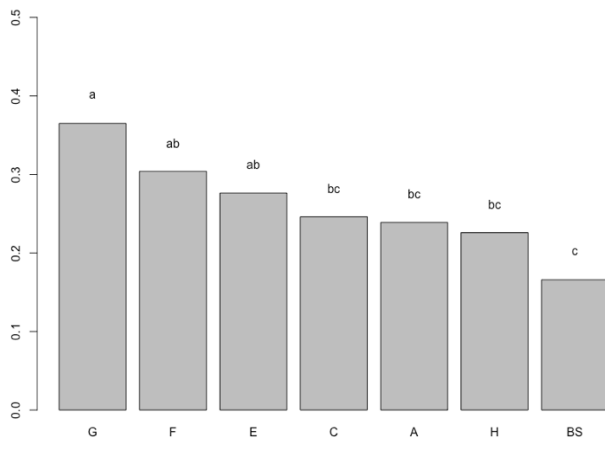
A scale of 2 to 400 ppm was used for the measures of the sodium. A stock solution of Na at 1000 ppm and Potassium chloride at 10 000 ppm was used to prepare the standard solution. Dilutions of either 1250 or 500 were done depending on the sample.

**Appendix B: Concentration of sodium, calcium, magnesium and potassium in the collected leachates**

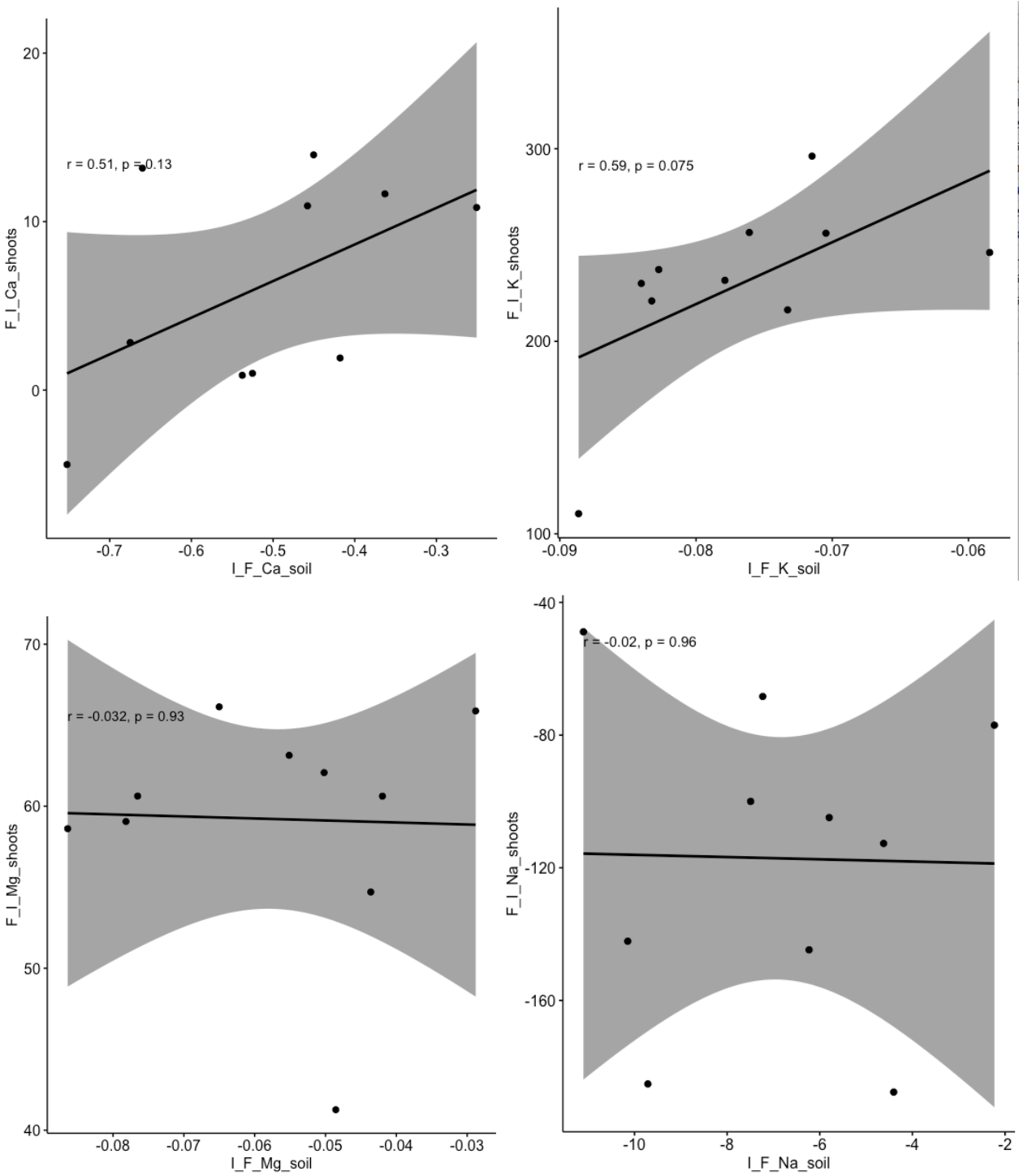
Treatment	Leachate 1	Leachate 2	Leachate 3	Leachate 4
<b>Sodium [mg/L]</b>				
100% - GR	26045.5±1189.2	9638.9±2869.5	8977.4±2762.8	5183.9±1527.9
50% - GR	29242.6±3478.7	9847.0±4668.3	9153.9±1264.3	5789.1±1363.7
100% - SR	37456.2±16247.8	16082.7±1152.6	14738.9±1422.7	9548.7±5272.2
50% - SR	34304.4±15117.8	13024.6±3823.3	11270.2±3114.3	5437.7±2686.2
Control	0.0	8436.5	14565.8±484.63	6529.0
<b>Calcium [mg/L]</b>				
100% - GR	546.1±144.4	732.4±421.4	344.5±78.4	378.2±126.4
50% - GR	1045.6±302.9	282.3±137.6	409.4±168.8	268.8±90.0
100% - SR	412.4±189.0	272.3±116.0	250.6±148.3	255.9±248.8
50% - SR	246.1±103.2	110.5±47.6	74.7±17.5	95.2±39.9
Control	0.0	356.5	115.0±24.4	63.8
<b>Magnesium [mg/L]</b>				
100% - GR	83.1±11.6	51.3±13.5	43.6±10.6	18.4±4.2
50% - GR	96.9±21.1	42.1±24.9	44.7±6.1	18.5±5.3
100% - SR	187.7±16.1	67.6±20.7	55.6±26.1	48.4±26.7
50% - SR	141.8±76.0	44.8±16.36	35.2±10.4	17.4±4.2
Control	0.0	75.7	29.5±4.9	14.5
<b>Potassium [mg/L]</b>				
100% - GR	62.9±4.5	26.7±6.5	24.9±3.8	15.6±2.5
50% - GR	65.4±2.9	24.6±11.5	25.8±3.7	16.5±3.6
100% - SR	75.1±12.4	40.1±5.6	35.1±1.6	26.9±8.7
50% - SR	70.4±22.3	33.3±7.7	30.3±4.7	22.3±6.2
Control	0.0	33.8±	34.8±1.2	17.0

**Appendix C: Graphical results of Duncan’s test in R studio**





**Appendix D: Phytoremediation correlation (Table 13)**



**Appendix E: Chemical amendments correlation (Table 13)**

