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IMPACT OF SALT AND METALLIC STRESS ON THE SODIUM AND POTASSIUM UPTAKE BY THE *VICIA FABA* L. PLANTS

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ABSTRACT

Environmental stresses, in particular salinity and heavy metal pollution, constitute a real problem menacing our ecosystems with detrimental effects on crop production and biodiversity. The present work aims to study the combined effect of salt stress by different concentrations of sodium chloride "NaCl" (0, 100, 200 meq) and metallic stress by different concentrations of lead "$Pb(NO_3)_2$" (0, 1000, 3000, 5000 ppm) on the sodium and potassium contents variations of the aerial and underground parts of *Vicia faba* L. plants.
The results obtained show that increased salinity causes an important accumulation of sodium in *Vicia faba* L. plants; with preferential sequestration in the roots. However, potassium compartmentalizes in the plant leaves studied with decreasing levels in response to high concentrations of sodium chloride and lead in the culture medium.

Keywords: *Vicia faba* L., sodium chloride, lead, sodium, potassium.

Introduction

Lead is one of the most widespread contaminants in the environment (Jarup, 2003; Hernández-Ochoa, 2005), toxic even at low concentrations for many living organisms (CITEPA, 2009). In fact, after arsenic, lead is considered the most dangerous pollutant (ATSDR, 2017; ATSDR, 2020). Their accumulation in the environment can affect the humans and animals health (Wang et *al*., 2003). In plants, it disrupts membrane and parietal structures, modifies water status, disrupts the absorption and/or translocation of essential mineral elements (calcium, manganese, zinc, iron, etc.) or even reduces photosynthesis (Seregin and Ivaniov, 2001; Sharma and Dubey, 2005).

To this contaminant is added the salinity factor, an ecological constraint for soils all over the world, particularly in the Mediterranean basin (Khan and Panda, 2008). It is a major constraint that affects the plants growth and development especially in arid and semi-arid regions (Bouassaba and Chougui, 2018). It is often associated with drought and leads to a reduction in cultivable land (Marcum, 2006).

Salinity is one of the major factors responsible for soil deterioration, making it unsuitable for agriculture. Due to their excessive salts concentration, saline soils constitute an unfavorable environment for the growth of most legumes (Benidire *et al*., 2017). It disrupts the plants mineral nutrition, by limiting the uptake of certain essential elements such as potassium and calcium (Haleem and Mohammed, 2007; Maksimović *et al*., 2010; Perveen *et al*., 2012).

This work consists in evaluating the combination impact of two abiotic stresses, lead and sodium chloride, on the mineral nutrition of broad bean plants (*Vicia faba* L.).

Material and Methods

Plant material and growth conditions

The plant species chosen for the realization of this work is the broad bean (*Vicia faba* L.), a plant of the fabaceae family selected because of its rapid growth and its important biomass.

The substrate used is a mixture of sand and peat (2V/V). The sand was taken from the sea side, sieved to remove all debris, washed with salt spirit then with faucet water several times and rinsed with distilled water and dried in the open air.

The broad bean plants cultivation is carried out in a controlled greenhouse where sowing is done in pots with a volume of 1000 ml and a diameter of 12.5 cm filled with previously prepared substrate (mixture of sand and peat). Plants hydromineral nutrition is ensured by watering three times a week with distilled water, replaced one time out of three by a nutrient solution of the Hogland type (Hoagland and Arnon, 1938).

The two-month-old *V. faba* L. plants are irrigated with a solution of lead nitrate ($Pb(No_3)_2$) at concentrations of 1000 ppm, 3000 ppm and 5000 ppm added to NaCl at 100 meq.l^{-1} and 200 meq.l^{-1} and in the presence or absence of a chelator (Na EDTA) added in an equimolar quantity to the lead.

Analyzes performed

- Extraction and dosage of the aerial and underground parts sodium content.
- Extraction and dosage of the aerial and underground parts potassium content.

The data obtained are subjected to a variance analysis with two factors in fixed randomization of classification, carried out by the software STATBOX 6.40. For the means comparison, the Newman Keuls test at the 5% threshold was used.

Results

Sodium content

Na^+ content of the *Vicia faba* L. aerial parts

The figure 1 reveals a higher level of sodium accumulated in the *V. faba* L. aerial parts in the presence of 200 meq.l^{-1} of NaCl with an average of 121.02 ppm followed by an average of 103.20 ppm in the presence of 100 meq.l^{-1} of NaCl.

The variance analysis of the Na^+ contents accumulated in the aerial parts reveals a clear "NaCl" effect (P = 0.00) on the expression of this parameter, unlike the "lead" effect which remains very far from conditioning the variations obtained (P = 0.87).

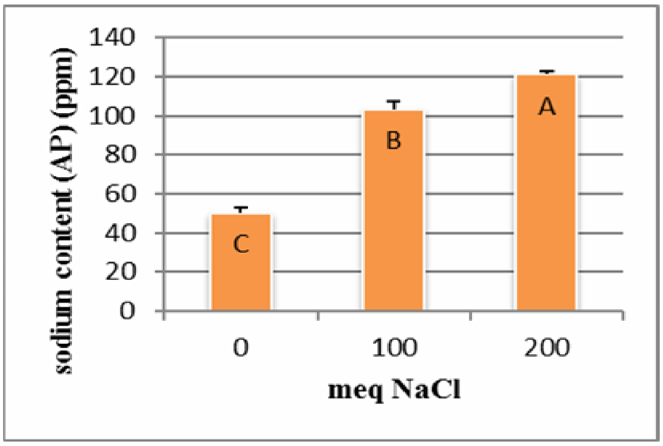

Fig. 1 : Combined effect of Pb and NaCl on the *Vicia faba* L. aerial parts (AP) Na^+ content. The results are presented as the mean ± σ and are grouped according to the NaCl factor using the Newman-Keuls test at 0.05.

Na^+ contents of the *Vicia faba* L. underground parts

The *V. faba* L. plants have a sodium accumulating capacity at the root level which increases proportionally with increasing NaCl supplied concentration (figure 2).

The variance analysis of this parameter indicates that its externalization is closely dependent on the "NaCl" factor (P = 0.00) while the "Lead" effect proves to be less significant (P = 0.04). By correlating the two study factors effect (NaCl, Pb) we find that the plants of Vicia faba L. react in the same way to the presence of lead, whatever its concentration. Indeed, this factor remains without significant effects on the expression of this parameter.

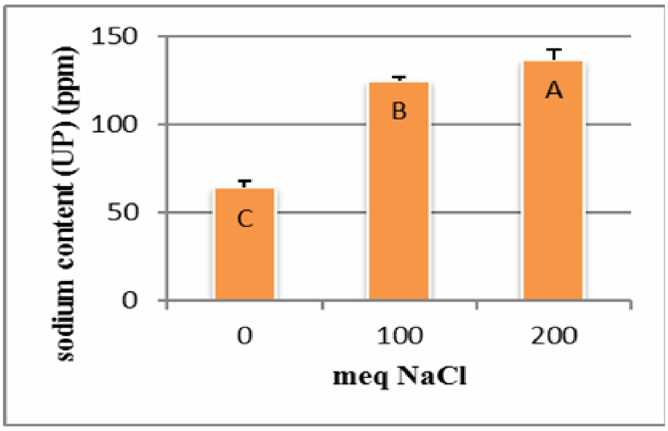

Fig. 2 : Combined effect of Pb and NaCl on the *Vicia faba* L. underground parts (UP) Na^+ content. The results are presented as the mean ± σ and are grouped according to the NaCl factor using the Newman-Keuls test at 0.05.

Potassium content

K^+ content of *Vicia faba* L. aerial parts

In the light of figure 3, it is established that the stems and leaves K^+ content are visibly high in all the unstressed control plants. While the addition of the saline solution to NaCl causes a decrease in the leaves and stems k^+ contents of the plant under study.

The variations analytical study of this parameter indicates that its elaboration is strongly governed by the factor "NaCl" (P = 0.00).

The study of the "Lead" factor makes it possible to discern two homogeneous groups A and B (figure 4), indicating that Lead acts on the expression of this parameter mainly by its presence or absence and the concentration has a less significant effect.

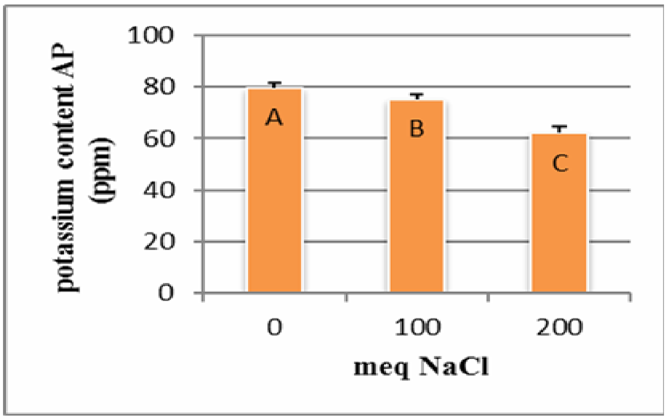

Fig. 3 : Combined effect of Pb and NaCl on the *Vicia faba* L. aerial parts (AP) K^+ content. The results are presented as the mean ± σ and are grouped according to the NaCl factor using the Newman-Keuls test at 0.05.

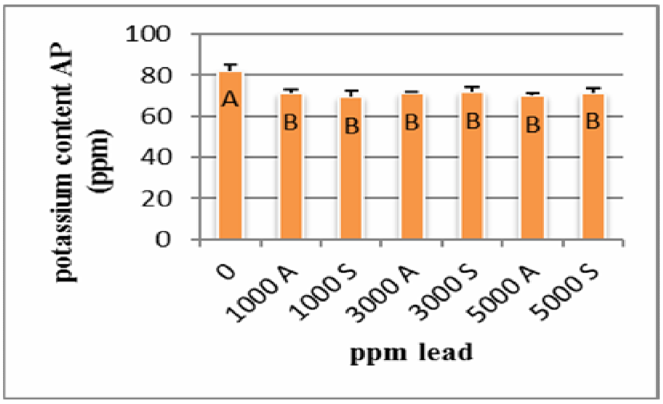

Fig. 4 : Combined effect of Pb and NaCl on the *Vicia faba* L. aerial parts (AP) K^+ content. The results are presented as the mean ± σ and are grouped according to the lead factor using the Newman-Keuls test at 0.05.

K^+ contents of *Vicia faba* L. underground parts

The results obtained for potassium in the roots of broad bean plants show that these values decrease significantly with increasing soil salinity (figure 5).

When the plants are stressed with lead, it should be noted that the k^+ level is lower in the roots of the treated plants compared to the control plants. This decrease is greater when the lead concentrations provided in the culture medium are higher (figure 6).

The variations analytical study of this parameter highlights a very highly significant effect of the "NaCl" factor (P = 0.00) and even of the "Lead" factor (P = 0.00).

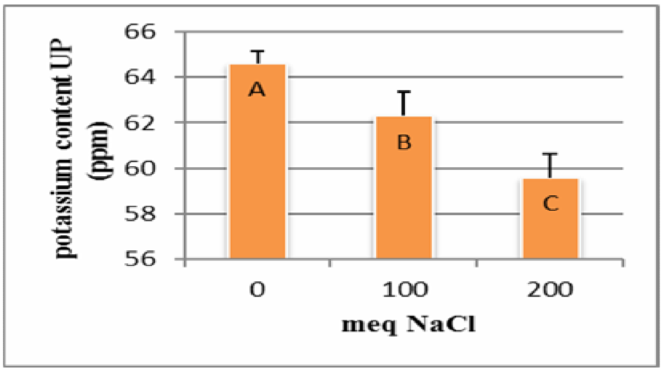

Fig. 5 : Combined effect of Pb and NaCl on the *Vicia faba* L. underground parts (UP) K^+ content. The results are presented as the mean ± σ and are grouped according to the NaCl factor using the Newman-Keuls test at 0.05.

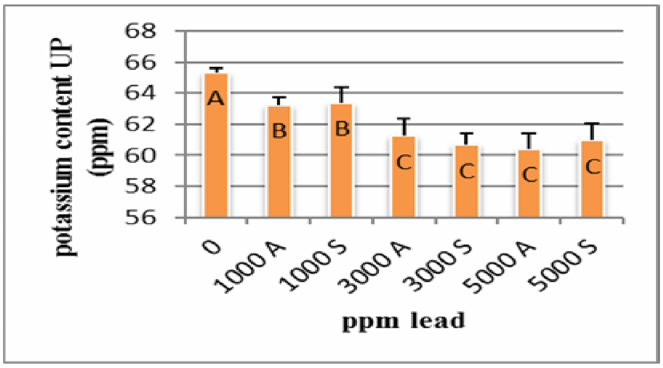

Fig. 6 : Combined effect of Pb and NaCl on the *Vicia faba* L. underground parts (UP) K^+ content. The results are presented as the mean ± σ and are grouped according to the lead factor using the Newman-Keuls test at 0.05.

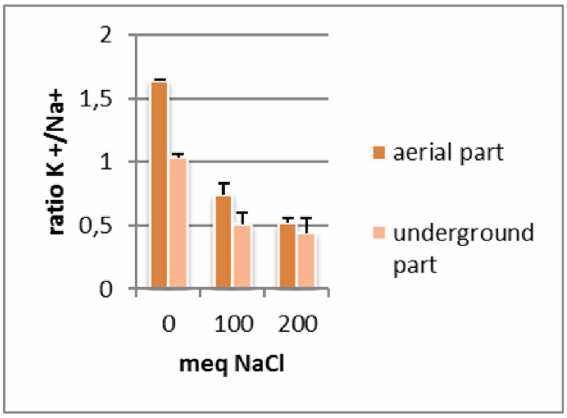

Fig. 7 : Effect of NaCl on the *Vicia faba* L. underground parts and aerial parts K^+/Na^+ ratio.

Discussion

The variations in the mineral balance recorded following the exposure of plants to an abiotic stress (saline, metal, etc.) suggest an adaptation kind developed by plants to confront the severe environment conditions. These variations depend on several factors such as the plant growth stage, the growing conditions, the stress type and its intensity.

Our results show that salinity induces a significant increase in the accumulated sodium rate in the aerial and underground parts of *V. faba* L. plants. This increase is positively correlated with the sodium chloride concentrations provided in the plants growing medium. These results are consistent with those of Chadli and Belkhodja (2007) and Benidire *et al.* (2017) who observe that the Na^+ content increases significantly in *V. faba* L. plants with increasing salinity.

By comparing the two parts of the plant studied, we observe a higher accumulation of Na^+ in the roots of the *V. faba* L. compared to the aerial parts, by limiting its transfer in large quantities in the aerial parts while thus creating a form of adaptation of plants in a salty environment (Snoussi, 2015). The same results are reported by Snoussi (2015) on broad bean plants.

The limitation of the accumulation of sodium in the aerial parts of the plant is due either to a blockage of the passage of this element at the root level or to a system of recirculation of sodium from the leaves to the roots of *V. faba* L. Moreover, there is a positive correlation between salt exclusion and salinity tolerance in several species (Lee *et al.*, 2003; Munns and James, 2003; Zhu *et al.*, 2004).

On the other hand, most glycophytes accumulate Na^+ and exclude K^+ when the availability of the two ions is similar or even when Na^+ is higher (Flowers and Lauchli, 1983) Our results confirm those reported by these authors; indeed, we found that the presence of high concentrations of salts in the *V. faba* L. plants culture medium decreases the absorption of potassium by the roots and thus its translocation to the plant aerial parts.

Potassium accumulates more in control plants, while a remarkable regression in the levels of this cation is recorded in stressed plants. It is important to note the extreme poverty in this element when the salt and/or metal stress becomes increasingly important. Similar results were found by Benidire et *al.* (2017). High salt levels are known to restrict plant production due to nutritional imbalance due to loss of nutrient uptake and/or transport to the shoot leading to ionic deficiencies (Munns, 2002; Heidari and Jamshid, 2010).

Indeed, the regression of k^+ contents can be explained by the difficulty of assimilation of this ion by the roots and its transfer to the aerial part following the high osmotic pressure and the ionic imbalance of the food environment (Snoussi, 2015). This could be explained by the competition between Na^+ and K^+ at the absorption sites (Cuin *et al.*, 2009; Panda and Khan, 2009).

The K^+ migrates remarkably towards the plant aerial parts. Its contents are higher in the plants aerial parts than in the underground parts.

The K^+/Na^+ selectivity ratio was higher in the leaves than in the roots (figure 7) but it decreased in the two parts studied following the increase in salt stress. Plants generally maintain a high cytosolic K^+/Na^+ ratio in the normal physiological state (Keisham *et al.*, 2018) this ratio will depend on the combined action of the different transport systems located at the membranes plasma level and vacuolar and involving the more or less selective pathways of K^+ and Na^+ ions (Maathuis and Amtmann, 1999).

The alteration of the K^+/Na^+ ratio is due to the increase in the influx of Na^+. Under salinity conditions, Na^+ influx is facilitated by pathways that typically function for K^+ influx, as the ionic radii of Na^+ and K^+ in their hydrated forms are similar, making discrimination between the two ions difficult (Keisham *et al.*, 2018). This suggests that the plants which successfully grow in saline medium are those which maintain a higher K^+/Na^+ ratio in their cytoplasm than in the rhizosphere (EL-iklil *et al.*, 2002).

The results indicate that the presence of lead in the *V. faba* L. culture medium does not have a significant effect on the variations in Na^+ levels in the tissues of these plants. Whereas the accumulation of K^+ is slowed down in response to metallic stress. The regression is more pronounced in the root parts than in the aerial parts. Our results corroborate those of Bouziani et *al*. (Bouziani et *al*., 2018) which show that *V. faba* L. plants decrease their root K^+ levels in response to the presence of increasing concentrations of lead in their culture media. Furthermore, the presence of lead is responsible for the inhibition and blocking of nutrients at root penetration sites (Godbold and Kettner, 1991).

Conclusion

Under salinity conditions, Na^+ accumulates more in the roots of *V. faba* L. plants. The sequestration of this ion in the root parts presents a form of resistance developed by the plant to attenuate toxicity in the aerial parts, which resulted in a preferential accumulation of K^+ in the aerial parts of the *V. faba* L. compared to the roots.

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