Data pubblicazione e doi

THE ROOT PROJECT:

How to guarantee tomato cultivation in salty soils

AUTRICE

Gatti, N.[1], Bossi, S.[1], Mannino, G.[1], Campobenedetto, C.[2], Contartese, V.[2], Bertea, C.M.

[1]Dipartimento di Scienze della Vita e Biologia dei Sistemi, Via Quarello 15/A, 10135 Torino; [2]Greenhas Group S.p.A,12043 Canale (CN).

noemi.gatti@unito.it, giuseppe.mannino@unito.it, cinzia.berta@unito.it

SCUOLE

L'articolo è stato revisionato dagli studenti e dalle studentesse di seconda superiore dell'Istituto di istruzione superiore J. C. Maxwell di Nichelino (To) e dagli studenti e studentesse di terza superiore del Liceo Scientifico Carlo Cattaneo di Torino.

Today, one of the most discussed problems is the increase of global population. The need to meet the market demand for foods has directly influenced agricultural practices. Recently, food quality and environmental sustainability are also the focus of the public attention (Campobenedetto et al., 2021).

The challenge is the following: how to make agriculture environmentally friendly? Reducing pesticides, using water properly, and maintaining soil fertility are central topics in agronomic studies (Campobenedetto et al., 2021). Thus, agriculture will need to produce food for a growing world population using an ever-shrinking land area: climate changes, agricultural practices (i.e. excessive use of fertilizers) and water scarcity have resulted in increased salt concentration in the soil (Mannino et al., 2020). Soil salinity is not compatible with the current farming practices. The role of scientists is to understand how to make plants more resilient to salt stress using environmentally sustainable methods.

THE TOMATO PLANT

The tomato plant is one of the most important crops cultivated all over the world. Statistical studies have revealed that tomato is probably the second most consumed vegetable in the world after potato, with the great difference that tomato can be also consumed as a raw food or used for various preparations (sauces, soups, or juices). The interest around tomato can also be described from a commercial point of view: with a total agricultural value of 2.062 billion of dollars, the People's Republic of China is the first world producer followed by USA and India. Italy occupies the sixth position in the world ranking, but the first in Europe. Another reason why tomato is so important is related to varieties: tomato is the first vegetable classified for the number of species. Therefore, tomato is cultivated in a wide variety of habitats, also thanks to the fact that this plant can be cultivated in field as well as in greenhouses, in those areas in which the temperature is too cold (Foolad, 2007).

Tomato is also considered a protective food as it can play an important role in a balanced diet by contributing to the intake of vitamin C, vitamin A, lycopene, β-carotene and various minerals (Foolad, 2007).

Each plant has a scientific Latin name, due to the need to have the same classification system worldwide. This system involves the use of two names: one for the plant genus (the first, an adjective belonging to similar species) and another for the species (the second). This “two name system” is called binomial nomenclature. The scientific name of tomato plant is *Solanum lycopersicum L*. “*Solanum*” is the genus while *lycopersicum* is the species name: technically, it can be translated as “wolf peach” from the Greek *lykos* (“wolf”) and *persikòn* (“peach”). “L.” represents the initial of the scientist name who named the tomato: Linnaeus (Foolad, 2007).

The tomato natural habitats range from very dry to very wet environments in both lowlands and mountains. This great variability in habitats has contributed to a remarkable deal of variety in tomato morphology; common features include yellow flowers (with 5 petals), the ability to grow up to three meters in height, and a well-developed root system. Commonly, people refer to tomato as vegetable, but actually it is botanically classified as a fruit. Depending on the species, the color of the fruits varies from red to yellow to green. The shape of the fruits can be globular, round, oval or elongated (Foolad, 2007) (Fig. 1).

Traditionally, the tomato fruit is an important element of the Mediterranean diet, but the origin is found in the New World, specifically in Andean Mountain system. The tomato plant was imported to Europe thanks to the Spanish conquests in the American continent. The first evidence of the presence of tomatoes in Europe dates back to 1554 (Foolad, 2007).

Figure 1. Representation of tomato plant growth

FACTORS THAT NEGATIVELY IMPACT ON TOMATO PLANT GROWTH AND PRODUCTION

Tomato plants can be cultivated in greenhouses or open fields.

Plants grown in the open field are susceptible to many environmental stresses: high temperatures, high light intensity, drought, pathogen presence, CO_2 concentration and soil properties. These factors directly influence the health status, causing negative physiological changes in plants (Fig.2) (Zhou et al., 2022).

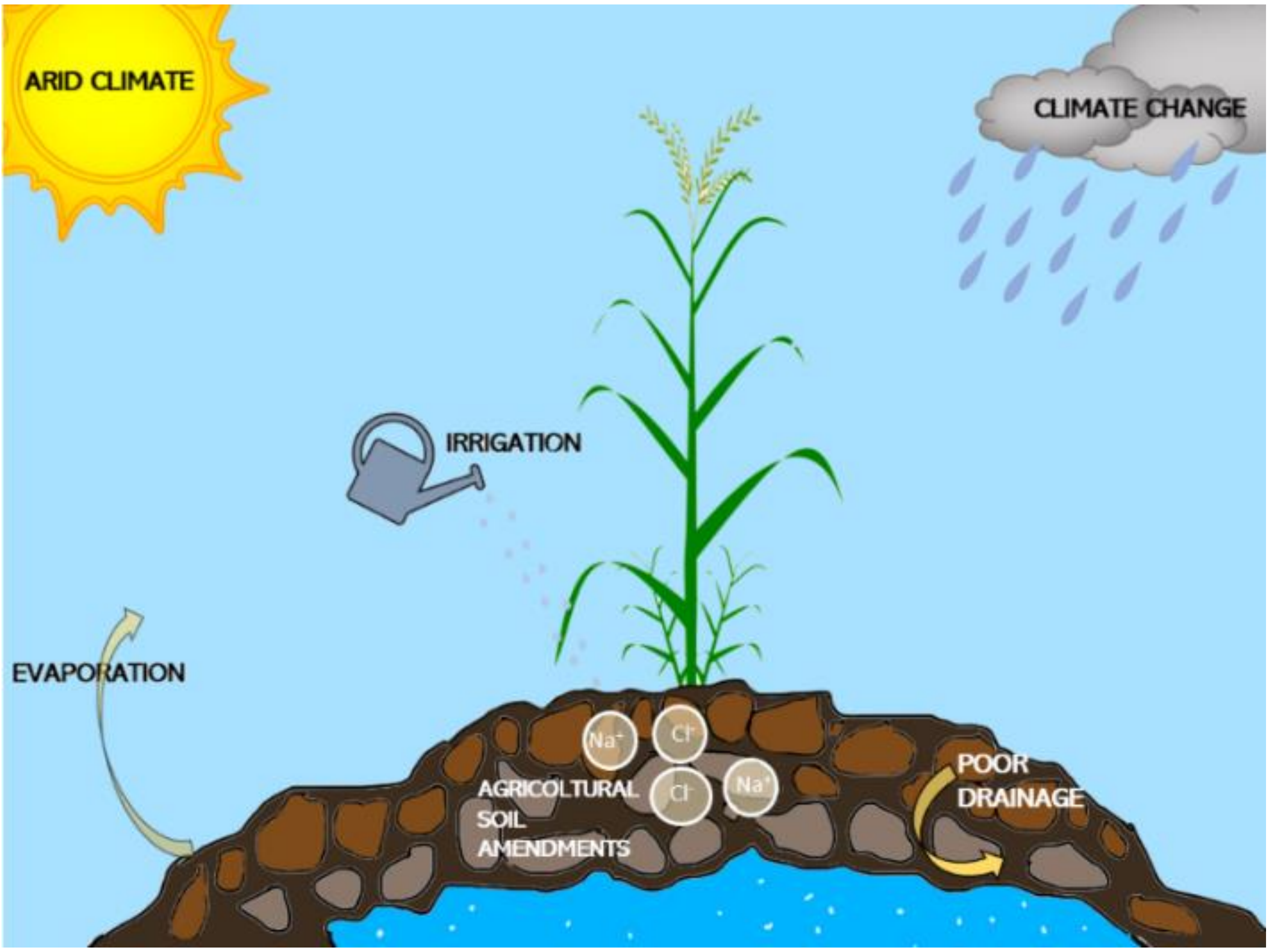

Figure 2. Schematic representation of phenomena that cause the increment of salt in soil

Focusing on soil properties, the increase of salty soils is a serious global problem for agriculture: salty soil is characterized by high level of salt. High concentration of salt in soil is a serious problem in agriculture: this phenomenon is especially linked to irrigated lands.

It has been estimated that more than 1 billion hectares worldwide are affected by salinity and the number is expected to grow (Munns, 2002) (Fig. 3).

Figure 3. Representation of countries with problems of salinization (Helmig, 2014)

When the health status of plant is negatively influenced by environmental factors, in plant physiology we talk about stress. Stresses can be defined also as an imbalance of resources. High amount of salt brings to salt stress, also called ionic stress (Munns, 2002; Saito & Matsukura, 2015).

The molecule of salt is constituted by the union of two ions, Na^+ (sodium) and Cl^- (chlorine). A high percentage of these ions in the soil reduces the ability of plants to absorb water with their roots. In addition, Na^+ tends to be accumulated in the leaves, causing leaf aging (visible to the naked eyes because leaf tissue starts to turn brown). This phenomenon leads to the cessation of vital leaf activities such as photosynthesis. Photosynthesis is the process by which inorganic substances (water, minerals and CO_2) are converted into organic substances (sugars) with the aim to provide the plant nutritional needs. High percentage of salt in cells damage also proteins (Munns, 2002; Saito & Matsukura, 2015).

Ionic stress can also cause the production of certain molecules called ROS (Reactive Oxygen Species) such as superoxide radical ($^1O_2^-$), hydrogen peroxide (H_2O_2) or free radical (OH^-). The reduction of water absorption in roots can causes dehydration in plant tissues.

The plant prevents the risk of water loss by closing the stomata. This process represents a defense mechanism. However, the stomata closure stops the absorption of CO_2, fundamental for the photosynthetic process. During photosynthesis, the energy is transported through a movement of electrons to the different cellular structures. At the end of the process, the energy is used to transform the carbon of CO_2 into sugars. If carbon is not present, sugar cannot be produced, but the energy of the electrons must still be dissipated. So, the electrons are transported to molecular oxygen, leading to ROS production (Mehler reaction) (Fig. 4). ROS are toxic and represent a symptom of plant stress. ROS can damage DNA, proteins and cell membranes (Ahanger et al., 2017).

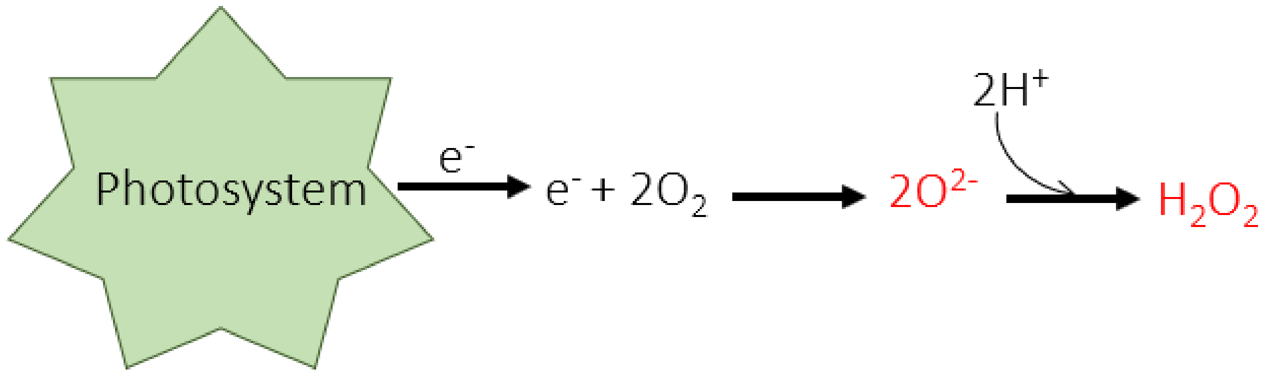

Figure 4.- Schematic representation of Mehler Reaction. e^-= electron; $2O_2$= molecular oxygen; $2O^{2-}$= superoxide radical (ROS); H_2O_2= hydrogen peroxide (ROS).

The reduction of absorbed water, the accumulation of ions in leaves, the reduction of photosynthesis activity, and the production of ROS inflict a poor health status to the plant: vegetative growth stops, and fruit quality drops (Munns, 2002; Saito & Matsukura, 2015).

Focusing on tomato plant, soil salinity causes suppression of seed germination, vegetative growth, leaf and root expansion, and the number of fruits per single plant and their quality are also negatively influenced (Fig. 5) (Munns, 2002; Saito & Matsukura, 2015).

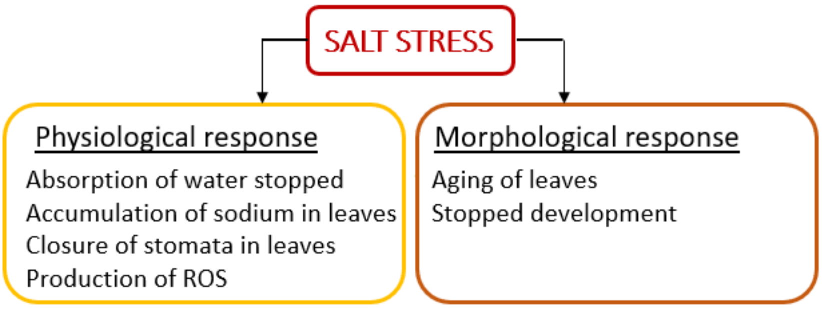

Figure 5. Schematic summary of the adverse effects of salt stress on plants.

PLANT BIOSTIMULANTS: A POSSIBLE SOLUTION TO IMPROVE SALT STRESS TOLERANCE

Soil salinity compromises soil fertility, that is the ability to support the nutritional needs of plants. It has been estimated that about 40% of the world's agricultural areas are subjected to fertility loss. Nowadays, having agronomic techniques with low environmental impact, able not only to enhance plant defense against soil conditions but also to ensure a high demand for food with high nutritional values, is becoming essential. Science is being called upon to address this need (Campobenedetto et al., 2021; Castiglione et al., 2021).

Plant biostimulants are a new type of products that can be found on the market, ideal for meeting the guidelines of sustainable agriculture. They are defined by the EU as "products that stimulate plant nutrition processes regardless of the nutrient content of the product with the sole purpose of improving one or more of the following plant or plant rhizosphere characteristics: nutrient use efficiency, tolerance to abiotic stress, quality traits, availability of confined nutrients in soil, or rhizosphere". It is possible to find this definition in the European Regulation (EU) 2019/1009, where we can find also rules about biostimulant formulation (Regulation of the European Parliament and of the Council Laying down Rules on the Making Available on the Market of EU Fertilising Products and Amending Regulations (EC) No 1069/2009 and (EC) No 1107/2009 and Repealing Regulation (EC) No 2003/2003, 2019). Biostimulants can be composed of a wide range of substances and/or microorganisms that can support plant growth at every stage (from germination to fruit harvest): they are able to increase root uptake capacity and improve the efficiency of nutrient distribution in plant tissues, enhance plant defense and tolerance mechanisms, and even crop quality (Mannino et al., 2020). In case of stress, plants possess biochemical systems of defense, normally called stress resistance systems. Biostimulants are able to improve plant performance, even in a hostile environment for the plant: this means that they can also improve the resistance system of plants. Biostimulants belong to various categories depending on the substances they are composed of: algal extracts, protein hydrolysates, humic substances, or microorganisms (Campobenedetto et al., 2021; Castiglione et al., 2021).

Frequently, in order to guarantee plant development in agriculture, pesticides are used to protect plants from pests and fertilizers are used to provide nutrients. Fertilizers and pesticides are also called agrochemicals. Nowadays, the use of agrochemicals is often discussed and the possible damages caused by these products to the environment is documented. Biostimulants are formulated to reduce the use of agrochemicals: they can help to meet the demand for high-quality food while protecting the environment. Biostimulants are often formulated from waste materials such as agro-industrial by-products, also respecting the philosophy of the circular economy (Campobenedetto et al., 2021; Castiglione et al., 2021; Mannino et al., 2020).

THE AIM OF THE ROOT (RESILIENCE TO SALINITY IN TOMATO) PROJECT

As previously mentioned, salinity and soil fertility loss, especially in coastal and irrigated lands, are urgent issues. In the near future, many areas of the planet will not be able to be cultivated without changing the agricultural practices.

In this context, to be resilient to salt stress, plants have to develop a good root system. The primary role of roots is anchoring plants to the soil and to recruit nutrients and water. They are the plant organs directly in contact with the soil: they are the first part of the plant suffering from ionic stress. In recent years, scientists have discovered the importance of root architecture under stress conditions, but we still have a limited knowledge about this process.

The goal of the ROOT project is to understand how to increase the resilience of plant root system under salinity stress. The project focuses on tomato due to its importance in

European agriculture and Mediterranean diet. To achieve this goal, ROOT has several key aspects:

- identify genes involved in root development

- find specific traits of a variety more tolerant to salt stress

- understand how biostimulants can help tomatoes to be more resilient to salt stress.

The ROOT project will contribute to develop future cultivation systems for tomato also in areas affected by salinization.

(https://www.suscrop.eu/projects-first-call/root).

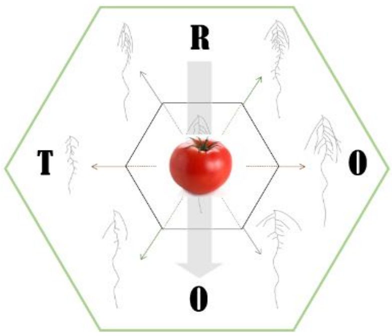

Figure 6. Logo of the project "Resilience to salinity in tomato – ROOT" (ERA-NET Cofund SusCrop 2018)

SOME RESULTS

A biostimulant based on tannin substances derived from waste material was tested on tomato plants. Condensed tannins are known for their antioxidant properties. In particular, Gallic Acid is a molecule highly present in a tannin mixture and can influence root growth. The application of this biostimulant at root level (via soil fertigation) has been shown to improve the development of root system (increased number of lateral roots) in tomato. In case of salt stress, the application of this biostimulant is able to restore the natural morphology of roots (Campobenedetto et al., 2021).

Recently, tomato fruits have also been tested. In presence of salinity stress, fruits tend to be smaller and with a higher concentration of polyphenols (antioxidant substances produced by tissues in response to stress). With the application of the biostimulant, not only the polyphenols were reduced, but also the amount of lycopene and β-carotene was higher. Lycopene is a pigment responsible for the red color of tomato and it is known for its antioxidant properties (able to prevent cancer). β-carotene is the main precursor of vitamin A, which is essential for the visual ability of our eyes. Therefore, with the biostimulant treatment it is possible to improve not only the plant health, but also the fruit quality while respecting the environment.

GLOSSARY

β-carotene	Antioxidant molecule. Precursor of vitamin A. Carotenoid (40 atoms of carbon)
Crop	A cultivated plant grown on a large scale for commercial purposes
Drought	Event of prolonged shortages in the water supply
Environmentally sustainable	Approach to preserve natural resources and protect global ecosystems
Ionic stress	Salt stress: bad state of plant health in presence of high salt concentration.
Ions	Atoms with a net electric charge caused to the loss or gain of one or more electrons
Humic substances	Organic substances main components of the humus.
Lycopene	Molecule responsible for the red color of tomato, antioxidant compound. Carotenoid (40 atoms of carbon)
Linnaeus	Swedish scientist
Microorganisms	Microscopic organism, such as bacteria or fungi
Morphology	A particular form or structure
Pests	Insects or other animals that attack crops
Rhizosphere	Portion of soil around the root; the chemistry of this soil portion is directly influenced by the growth, respiration, nutrient exchange and root exudates
Root system	Network of all the roots (primary and secondary) in a plant
Soil fertility	Capacity of a soil to provide essential nutrients to crops
Stomata	The complex of two cells on the leaves whose role is to create an opening fundamental for gas exchanges (entry of CO_2 and

	exit of O_2) and plant transpiration (loss of water vapor in the atmosphere). In case of stress, stomata are closed to prevent water loss.
Tannin substances	Group of bitter and astringent compounds, they belong to the family of polyphenols. Antioxidant compounds.

REFERENCES

Ahanger, M. A., Tomar, N. S., Tittal, M., Argal, S., & Agarwal, R. (2017). Plant growth under water/salt stress: ROS production; antioxidants and significance of added potassium under such conditions. *Physiology and Molecular Biology of Plants*, *23*(4), 731-744.

Campobenedetto, C., Mannino, G., Beekwilder, J., Contartese, V., Karlova, R., & Bertea, C. M. (2021). The application of a biostimulant based on tannins affects root architecture and improves tolerance to salinity in tomato plants. *Scientific Reports*, *11*(1), 1-15.

Castiglione, A. M., Mannino, G., Contartese, V., Bertea, C. M., & Ertani, A. (2021). Microbial biostimulants as response to modern agriculture needs: Composition, role and application of these innovative products. *Plants*, *10*(8), 1533.

Foolad, M. R. (2007). Genome Mapping and Molecular Breeding of Tomato. *International Journal of Plant Genomics*, 2007, 1-52.

Helmig, I. R. (2014) Kinetic approach for modeling salt precipitation in porous-media.

Mannino, G., Campobenedetto, C., Vigliante, I., Contartese, V., Gentile, C., & Bertea, C. M. (2020). The application of a plant biostimulant based on seaweed and yeast extract improved tomato fruit development and quality. *Biomolecules*, *10*(12), 1662.

Munns, R. (2002). Comparative physiology of salt and water. *Plant, Cell & Environment*, 25 (2), 239-250.

Regulation of the european parliament and of the council laying down rules on the making available on the market of EU fertilising products and amending Regulations (EC) No 1069/2009 and (EC) No 1107/2009 and repealing Regulation (EC) No 2003/2003, 2019 Official Journal of the European Union 114 (2019).

Saito, T., & Matsukura, C. (2015). Effect of Salt Stress on the Growth and Fruit Quality of Tomato Plants. In Abiotic Stress Biology in Horticultural Plants. *Springer Japan* (pp. 3-16).

Zhou, X., Joshi, S., Patil, S., Khare T., Kumar, V. (2022). Reactive oxygen, nitrogen, carbonyl and sulfur species and their roles in plant abiotic stress responses and tolerance. *J Plant Growth Regul* 41, 119–142

ROOT Project: https://www.suscrop.eu/projects-first-call/root