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An Outlook on the Chenopodium Quinoa Willd (Quinoa) Plant and the Role of the in Vitro Culture and Nanotechnology in Mitigation of Salinity Stress: A Review

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ABSTRACT

Climate change and scarcity of water resources in many countries of the Middle East have led to a drastic decline in the quantities of the main crops and exacerbated the problem of soil salinity. Therefore, it is imperative to find alternative crops that would enhance food security and can tolerate abiotic stresses such as soil salinization. Quinoa is a multi-purpose crop, grown mainly because of its historical, ecological, economic, and high nutritional value. The plant is highly adapted to different environments, and it can be grown in areas with marginal soils that are poor in rainwater. The emergence of this crop's importance and its distinctive properties began in the seventies of the last century. So, there is a need to increase this crop production under salinity conditions using new technology. The plant tissue culture can play an important role to give suitable conditions to study the plant responses to salinity stress via using different factors such as nanoparticles and others. Beside that, the other factors and environmental conditions can be easily controlled in vitro which makes the study easier. In this review, the description and response of the quinoa plant to salinity stress were summarized. Furthermore, the ability to use plant tissue culture and study the effect of adding nanoparticles (NPs) to the culture media to increase salt tolerance was the hot spot of this review. This was to find out the importance of nanoparticles and the in vitro plant tissue culture to increase the quinoa tolerance against salinity stress.

Keywords: Nanoparticles, Plant tissue culture, Physiological responses, Quinoa, Salinity stress.

INTRODUCTION

Salinity stress is one of the major constraints that threaten Middle East countries (Tnay, 2019). It had affected cereal crop productivity and caused soil deterioration, and pollution (Sabagh, et al., 2020). Quinoa can be considered as a new alternative crop for Mediterranean conditions affected by multiple abiotic stress (Bilalis, et al., 2019). This is because, quinoa has a

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high nutritional value (Bastidas, et al., 2016). Alternative crops such as sorghum, pearl millet, and quinoa are uniquely appropriate, as they perform well under conditions in which many other crops fail (Elouafi, et al., 2020). It is a good source of protein as this plant is almost the only one among the plants that contain the nine essential amino acids, and it is free of gluten (Elsohaimy, et al., 2015) (Vilcacundo and Hernandez-Ledesma, 2017). The protein content ranges from 12.5 to 16.7% (Pereira, et., 2019). Quinoa provides a large amount of energy as is wheat, rice, beans, and corn. The percentage of carbohydrates content ranges from 60 to 70% (Pereira, et., 2019). Quinoa contributes to controlling blood sugar levels because it is rich in fiber that reaches 12% of dry matter. As for the fatty acid, values range between 2.0 and 9.5%. It is a rich source of main fatty acids such as linoleic and α-linolenic (Filho, et al., 2017). Quinoa also has high quantities of minerals such as Ca, Fe, K, Mg, Mn, P, and Zn, besides the good quality of fatty acids (Pereira, et., 2019). As well as its seed contain vitamins such as B, C, and E and multiple antioxidants like flavonoids (Miranda, et al., 2012). Those flavonoids are useful in preventing degenerative diseases such as atherosclerosis, coronary heart disease, cancer, Alzheimer’s disease, and diabetes (Vilcacundo and Hernandez-Ledesma, 2017). The analysis results of seeds indicated their content of phytoecdysteroids (Kumpun, et al. 2011). Those phytoecdysteroids are the secondary metabolites of plants that have a protective role against nematodes and insects and have positive effects on the health of humans (Kumpun, et al. 2011). The seed coat accumulated saponins, which have insecticidal, fungicidal, antibiotic, and pharmacological characters (Vega-Galvez, et al. 2010). Thus, enhancing the plant’s defense against pathogens and pests.

Quinoa is a stress-tolerant plant, that can adapt to several environmental conditions. In recent years, quinoa has become an important model crop for understanding plants’ responses to water and salinity stress (Hinojosa, et al., 2018). Quinoa is a selective halophytic plant species. The most tolerant species can tolerate high salinity levels like that that exist in seawater (Adolf, et al., 2013). Indeed, the high salinity tolerance character of quinoa induced researchers' interest. New technologies can be used to maximize the benefits of salinity tolerance crops. Plant tissue culture can develop suitable conditions to study the stress of many plant species (Gulzar, et al., 2020). This is because all factors and environmental conditions can be easily controlled. The approach of nanoparticle (NPs) synthesis and their relative structure and size plays a crucial role in displaying the biological properties of plants (Rai, et al.,2018). Recently, researchers are attempting to exhibit the prospective effects of different kinds of nanoparticles in plants (Sturikova, et al., 2018). This includes increasing the plant tolerance against salt stress and other stresses. Furthermore, plant tissue culture could be a beneficial tool via using nanoparticles techniques to increase plant tolerance against stress. Therefore, studying the responses of in vitro grown quinoa to induced salinity stress, as well as the effect of the induced nanoparticles on the stressed plant can be considered an interesting field of research. The aim is to provide a complete review of the importance of quinoa’s production via tissue culture. Besides that, reviewing the ability of the nanoparticles for enhancement of salinity tolerance in other crops to be used in the future to increase the production of quinoa as an important food crop using tissue culture.

Quinoa and abiotic stress

The eastern Mediterranean region is affected by important climatic changes, which caused significant shifts in climate from wet to dry (Roberts, et al., 2011). Plant growth and development are frequently affected by biotic and abiotic factors (Gull, et al., 2019) and (Ahmad, et al., 2021). Abiotic stresses are considered the major cause of crop losses (Mariani and Ferrante, 2017). They hurt plant growth, production, and yields (Ahmad, et al., 2021). The main abiotic stresses are drought, high salinity, heavy metals, and excess heat (Gull, et al., 2019). The use of suitable crops with improved tolerance to abiotic stresses could be one of the suitable options to cope with changing climatic conditions (Scheben, et al., 2016). The physiological and metabolic adaptations plants' responses to abiotic stress vary at the molecular, cellular, and organism levels. Understanding these mechanisms requires suitable stress models and stress markers. Therefore, in recent years quinoa has become an important model crop for understanding plants' responses to salinity stress (Hinojosa, et al., 2018).

Salinity stress

High salinity can consider as one of the main abiotic stresses. It can determine the yield and productivity of the crop, protein synthesis, and respiration. It can cause denaturalization of cell membrane, imbalance of nutrients, closure of stomata, and production of reactive oxygen species (Ali, et al., 2020). Quinoa as a facultative halophyte plant has salt tolerance greater than that of wheat, corn, and barley (Zeeshan, et al., 2020; Zahra, et al., 2020). Salinity tolerance among quinoa genotypes is widely variable. However, it does not correlate with the geographic distribution of quinoa (Hussain, et al., 2018). Additionally, a much higher salinity tolerance level was found in a wild relative of quinoa (Chenopodium hircinum) than quinoa cultivars (Schmockel, et al., 2017). Generally, quinoa can be considered among halophytic plants that can tolerate high levels of salinity (Adolf, et al., 2013). It can tolerate salt concentrations ranging from 150 mM NaCl up to 750 mM NaCl which is equivalent to electrical conductivity 15 dS m−1 - 75 dS m−1 (Orsini, et al., 2011); that is greater than seawater salinity 45 dS m−1(Adolf, et al., 2013). While, when soil solution exceeds electrical conductivity ~4 dS m−1 (40 mM NaCl), the yields of glycophytic crops, such as rice, wheat, and corn start to drop (Sun, et al., 2017; Gunes, et al., 2007). Nevertheless, quinoa salt tolerance is widely varied among its genotypes (Schmockel, et al. 2017). In the last 20 years responses to salinity stress have been studied in quinoa extensively (Adolf, et al., 2013).

To maintain internal plant water status within physiologically acceptable limits, there was a wide range of morphological, physiological, and even biochemical responses that can adjust water demand and uptake in response to the environment and soil water content. Hence, understanding the physiological mechanisms associated with plant responses to salinity stress is essential for determining the critical concentration that the plant can handle. Therefore, in recent years quinoa has become an important model crop for understanding plants' responses to salinity stress (Hinojosa, et al., 2018). Salinity affects plant growth by causing morphological changes in many growth parameters (Safdar, et al.,2019). The shoots appear through a decrease in the length of the stem and a reduction in the number of leaves, as well as the reduction of the lateral branches and the diameter of the plant members (Hussain, et al., 2017). The state of water in plant leaves is very sensitive to salinity, through which the reactions of the plant to stress are indicated or determined (Safdar, et al.,2019). The high concentrations of salinity led to a decrease in the relative water content of quinoa seedlings (Koyro and Eisa, 2008).

Five quinoa (Chenopodium quinoa Willd) genotypes (ICBA-Q1, ICBA-Q2, ICBA-Q3, ICBA-Q4, and ICBA-Q5) were evaluated for salinity tolerance under four artificially induced salinity (5, 10, 15, 20 dS m-1) levels (Qureshi and Daba, 2020). They had shown significant differences on multiple growth parameters with the increasing salinity levels. There was a significant reduction at the highest salinity level on all the measured parameters such as; plant height, germination percentage (GP), biomass, grain yield and shoot and root dry matter (Qureshi and Daba, 2020). As well as on proline content which is synthesized under stress conditions as a defense mechanism against induced stress. Proline is one of the most important osmotic regulators during salt stress

(Shafi, et al., 2019). It is an amino acid that accumulates in plants when exposed to stress and plays the role of an effective osmotic protector (Shafi, et al., 2019; Sharma, et al., 2019). An excess of proline in the cell leads to the exclusion of sodium and chlorine ions in the cell subjected to salt stress (Torabi., 2014). It works on the stability of cell membranes by binding with phospholipids, and it is considered a hydroxyl radical scavenger (Azooz, and Ahmad., 2016). Ruiz et al., 2016, stated that the quinoa plant has various mechanisms to tolerate salinity stress among those were an accumulation of phenolic compounds and proline as compatible solutes (Talebnejad and Sepaskhah, 2016).

Most quinoa genotypes had not been affected at salinity levels ranging from 100 to 200 mM NaCl. While, the onset of germination can be delayed at salinity levels between 150 to 250 mM NaCl (Stoleru, et al., 2019). The process of quinoa germination had been affected under salinity conditions due to changes in sugar metabolism and invertase activity. however, at the seedling stage sugar levels can be increased or decreased in roots grown under salinity levels below 200 to 400 mM NaCl according to quinoa genotypes (Hinojosa, et al., 2018).

Quinoa tissues can accumulate K+ to maintain efficient osmotic adjustment of leaf cells under 320 mM NaCl level. Under 400 mM NaCl level Na+ concentrations increased 10 times in quinoa leaves followed by a two-fold increase in quinoa leaf (Adolf, et al., 2012). Shabala, et al. (2011) stated that barely - salt-tolerant genotypes had high concentrations of K+ under NaCl level 320 mM, to get an efficient osmotic adjustment that was required to keep leaf cell growth and expansion. Therefore, the ability of quinoa to elevate K+ uptake under salinity conditions can be considered as a remarkable ability under salt stress levels. Keeping of high K+/Na+ ratio by increased leaf K+ can be considered an important indicator for salinity tolerance in quinoa (Shabala, et al., 2012) and (Almeida, et al., 2017).

Quinoa plants can reduce the desirable effect of ionic toxicity resulting from high Na+ and Cl− accumulation under 400 mM NaCl level by K+ retention. This K+ retention minimize Na+

and Cl− toxicity and restricted their accumulation (Shabala and Pottosin, 2014).

Tissue culture role in the propagation of Quinoa

Plant tissue culture had been used for different plant species to increase the mass production of plants under controlled environments (Kim, et al., 2017). The role of plant tissue culture in different studies such as salinity stress is very important (Thorpe, 2007). Different studies have used the quinoa plants for in vitro propagation to study different factors such as breeding, propagation for food use, and many others. A protocol of complete micropropagation for quinoa coastal cultivars has been developed with high success rates and this can be considered as a starting point that enhances the breeding efforts for basic knowledge and production of quinoa (Regalado, et al., 2020). Sufficiently high success rates for a complete regeneration procedure for quinoa, and the effect of plant growth regulators such as 6-Benzylaminopurine (BAP) and α-Naphthalene Acetic Acid (NAA) at different concentrations as well as in combination and used various amounts of sugar (sucrose) in vitro could be the starting point for the development of other biotechnological tools (Al Gethami and El Sayed., 2020). Eisa et al., (2005) reported the first procedure of somatic embryogenesis in Quinoa using hypocotyl explants of quinoa seedlings on a modified Murashige and Skoog (MS) medium supplemented with 0.45 µM 2,4-D which could contribute to further advances in this area. The first study of an appropriate in vitro regeneration procedure for quinoa by immediate organogenesis through cotyledonary node explants was reported by Hesami, et al. (2018). This can enhance quinoa breeding programs and can also be exploited for active biomolecules of this valuable plant (Hesami et al., 2018).

Plant in vitro organogenesis where was utilized to establish a regeneration system by indirect organogenesis in quinoa, that can be further used for genetic transformation (Hesami and Daneshvar., 2016). However, till now there are few attempts to utilize tissue culture to study the salinity stress under in vitro conditions for quinoa. Furthermore; using some new technologies such as nanoparticles can help in the process of plant tolerance versus salt stress. Till now there are some attempts to use nanoparticles with plants to increase growth. It would be a great idea if we have added a nanoparticle technique to quinoa in vitro shoots and show their effect on production and salinity stress.

Role of nanotechnology to mitigate salinity

Using nanomaterials in all life aspects now is a hot topic. Different nanomaterials have been used in plants for different purposes. The uptake, translocation, and accumulation of nanomaterial by the plant are determined by plant species (Singh, et al., 2015). There are different methods by which the metal NPs can be passed up by plants. They can be taken up either as the metal NPs that oxidized to metal ions or NPs themselves (Rico, et al., 2011). Recently, several studies indicated that the mediated effect of nanoparticles on plant growth and development is dependent on their type, size, and concentrations (Das and Das 2019). Ashkavan, et al. (2015) stated that nanoparticles can maintain critical physiological and biochemical aspects. Using nanoparticles has been reported to increase plant tolerance against salinity. For example; the application of nanoparticles with different concentrations on bread wheat (Triticum aestivum L.) that were subjected to drought stress resulted in increased growth, yield, and starch content (Jaberzadeh, et al., 2014). While application of nanoparticles with basil (Ocimum basilicum) which were subjected to salinity stress resulted in a significant increase in growth parameters and chlorophyll and proline content (Kalteh, et al., 2018). The application of titanium oxide (nano TiO2) with different concentrations of 0.01, 0.02, and 0.03% in drought-stressed Wheat (Triticum aestivum L.) increased the growth, yield, gluten, and starch content of wheat (Jaberzadeh, et al., 2014).

Recently, many reports have cleared the role of NPs in plant tissue culture, for example; adding silver nanoparticles to the medium in which Tecomella undulating stem explants were used the number of shoots, percentage of shoot induction, and formation of callus was enhanced (Aghdaei, et al., 2012). Sarmast and Salehi 2016, stated that the percentage of shoot production number of shoots, and shoot length were increased when culturing Tabernaemontana undulata nodal explants on MS medium that had been contained Ag NPs. The addition NPs to plant tissue culture medium has a significant effect on the formation of roots, the proliferation of callus, somatic embryogenesis, and multiplication of shoots by changing the activity of antioxidant enzymes (Sadidi, et al., 2020).

Regarding quinoa the only published application on the effect of nanoparticle quinoa was done by Al Gethami and El Sayed., (2020). Al Gethami and El Sayed., (2020) concluded that the ZnO Nanoparticles could be a positive effect on the micropropagation of quinoa plants through increasing germination rate, number of shoots, and leaves. Based on their experimental results the concentrations of ZnONPs in range (2-10 mg/l) may a positive effect on micropropagation of the Quinoa plant especially on rooting formation stage, but the concentrations of ZnO-NPs that higher than 10 mg/l hurt the growth and development of Quinoa plant. Figure 1 summarized a general description of in vitro plants, responses under induced salinity, stress, and the effect of NPs to mitigate the induced stress injuries. Nanoparticles could be used in plant tissue culture to increase salt stress tolerance. This could be concluded from other studies on other plant species. The summary of the effect of the nanoparticles on in vitro culture plants could be seen in the scheme in Figure 1.

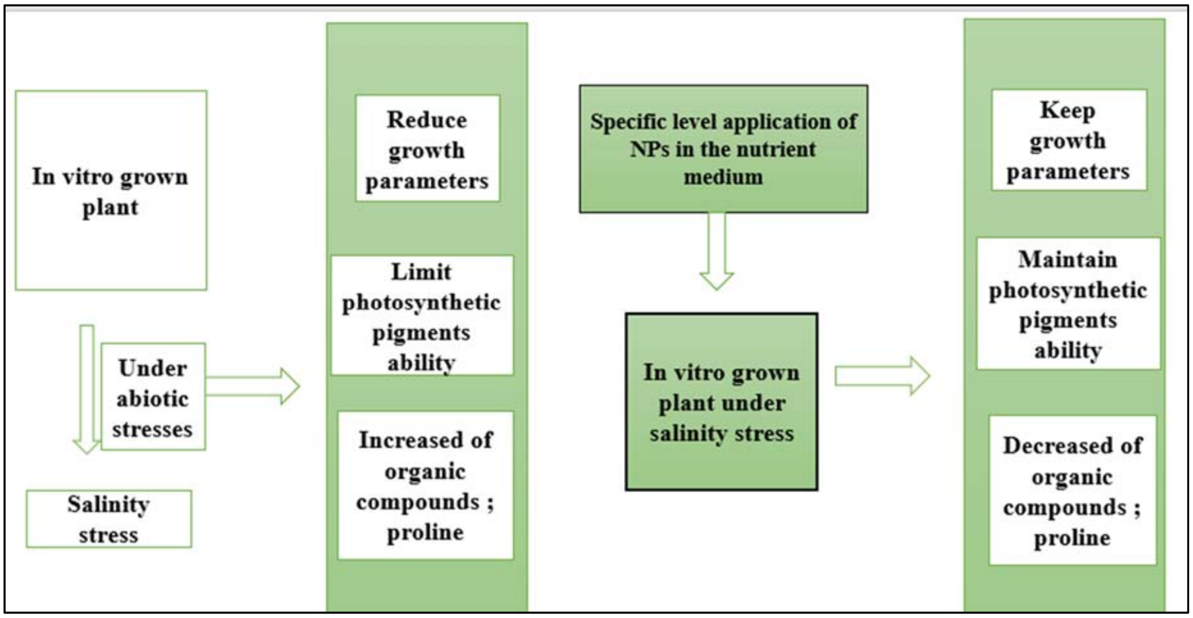

Figure. 1 General description of *in vitro* plant responses under induced salinity, stress, and the effect of NPs to mitigate the induced stress injuries.

5. Conclusions

Quinoa has been accepted as a great and adaptative crop as it can grow in an area different from where it originated. This was revealed the adaptation potential of quinoa to different environmental conditions. On other hand, plant tissue culture can develop suitable conditions to study the stress of many plant species. This is because all factors and environmental conditions can be easily controlled. As well as nanoparticle synthesis and their relative structure and size play a crucial role in plant growth. Therefore, in vitro imposed salinity stress and nanoparticles can be used to analyze quinoa responses to salinity. Recently, researchers have been attempting to exhibit the prospective effects of different kinds of nanoparticles in plants. The current review provides insight into the effect of NP and its application in plant tissue culture under controlled conditions. The previously mentioned studies on the effect of NPs on in vitro grown plants can confirm the remarkable linking between two approaches; namely, plant tissue culture and nanotechnology. Taking into consideration that the specific application of NPs can prodigiously aid in ameliorating the negative impact of induced salinity stress on in vitro grown plants.

This study will pave the way to conduct different studies on the quinoa plants to enhance the utilization of different NPs on in vitro grown quinoa. This topic will serve as a basis for guiding research on the quinoa plant using new techniques to provide its production under stress conditions.

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نظرة عامة على نبات الكينوا **Chenopodium quinoa Willd** و دور الاكثار الدقيق وتقنية النانو في التخفيف من إجهاد الملوحة: مراجعة

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ملخص

أدى تغير المناخ وندرة الموارد المائية في العديد من دول الشرق الأوسط إلى انخفاض حاد في كميات المحاصيل الزراعية الرئيسية وتفاقم مشكلة ملوحة التربة. لذلك، من الضروري إيجاد محاصيل بديلة من شأنها أن تعزز الأمن الغذائي وتتحمل الضغوط غير الحيوية مثل تملح التربة. الكينوا محصول متعدد الأغراض، يُزرع بشكل أساسي بسبب قيمته التاريخية والبيئية والاقتصادية والغذائية العالية. يتكيف النبات بشكل كبير مع البيئات المختلفة، ويمكن زراعته في مناطق ذات تربة هامشية فقيرة في مياه الأمطار . بدأ ظهور أهمية هذا المحصول وخصائصه المميزة في سبعينيات القرن الماضي. لذلك، هناك حاجة لزيادة إنتاج هذا المحصول في ظل ظروف الملوحة باستخدام التكنولوجيا الجديدة. يمكن أن تلعب زراعة الأنسجة النباتية دورًا مهمًا في توفير الظروف المناسبة لدراسة استجابات النبات لإجهاد الملوحة من خلال استخدام عوامل مختلفة مثل الجسيمات النانوية وغيرها. إلى جانب ذلك، يمكن التحكم في العوامل والظروف البيئية الأخرى بسهولة في المختبر مما يجعل الدراسة أسهل. في هذه المراجعة، تم تلخيص وصف نبات الكينوا واستجابته لإجهاد الملوحة. علاوة على ذلك، كانت القدرة على استخدام الانسجة النباتية ودراسة تأثير إضافة الجسيمات النانوية (NPs) إلى وسط الاستزراع لزيادة تحمل الملح هي النقطة الساخنة في هذه المراجعة. كان هذا من أجل معرفة أهمية الجسيمات النانوية الانسجة النباتية لزيادة تحمل الكينوا ضد إجهاد الملوحة.

الكلمات المفتاحية: الجسيمات النانوية، زراعة الأنسجة النباتية، الاستجابات الفسيولوجية، الكينوا، إجهاد الملوحة.