



## REVIEW: THE EFFECT OF SALT STRESS ON SOME PHYSIOLOGICAL AND BIOCHEMICAL COMPOSITION OF SOME CROPS

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### Abstract

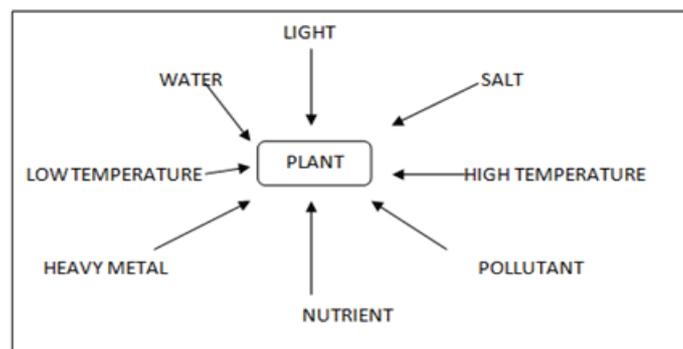
Abiotic stress like salt, drought, cold, high temperature and heavy metals largely influence crop productivity and development. Abiotic stress is a main threat for food safety as a result of the deterioration of environment caused by human activity and constant changes of climate. To handle successful abiotic stress problem different crops begin some physiological, molecular, and cellular modifications to adapt and respond to stresses. Good understanding of the crop responsiveness to stress conditions will help in both modern and traditional breeding techniques towards improving stress tolerance.

### Introduction

Abiotic stress like salt, drought, cold, high temperature and heavy metals largely influence crop productivity and development. Abiotic stress is a main threat for food safety as a result of the deterioration of environment caused by human activity and constant changes of climate. To handle successful abiotic stress problem different crops begin some physiological, molecular, and cellular modifications to adapt and respond to stresses. Good understanding of the crop responsiveness to stress conditions will help in both modern and traditional breeding techniques towards improving stress tolerance.

use of energy that the plant should use to take water from the soil solution and to improve its physiological adjustments. This leads to reduced yield and growth of plants. Salt stress decrease the relative leaf water content, water potential, leaf water relation parameters, osmotic potential, turgor potential, and ultimately inhibited plant growth and decreased the crops fresh weight (Jabeen and Ahmad, 2012). Water stress occur due to water deficit, caused by high soil salinity or drought. In case of high salinity, water exists in the soil but plants cannot uptake it, which is called physiological drought (Chaves *et al.*, 2003). When water is gradually lost from a completely saturated soil, firstly by draining freely under the effect of gravity, and the rate of loss progressively slows down till no more water drains away, the soil is called to be at field capacity. Further decrease in water by uptake by plant roots or by evaporation reduces the water content of the soil, till no more loss, a stage called the wilting point at which roots cannot absorb water important to meet their needs, and the plants wilt and die (Nagarajan and Nagarajan, 2010).

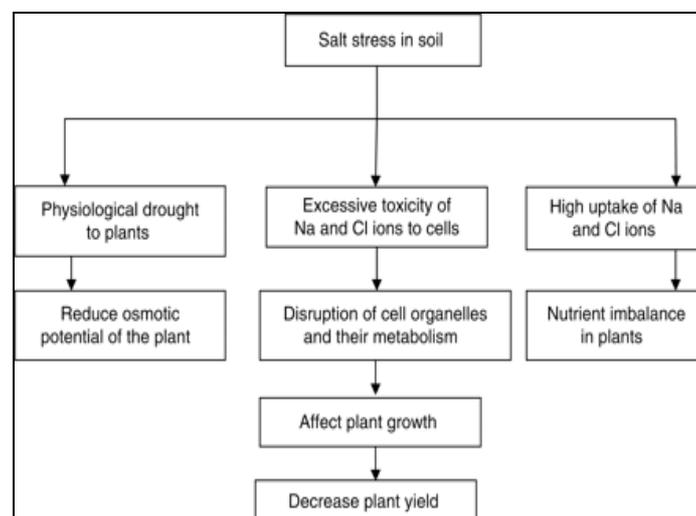
Understanding tolerance mechanism to salinity and drought is necessary to provide insights into the tolerance mechanism against these abiotic stresses at molecular, physiological, and biochemical levels. Improving tolerance of different crop plants could be happened by different ways such using bio and organic-stimulants that has been an important but largely unfulfilled aim of modern agricultural development.



**Fig. 1:** Some types of abiotic stress factors that affect plants life.

Drought and Salinity are two main dangerous abiotic stresses that threat different crops growth and yield around the world (Guo *et al.*, 2014). One third of the world's population lives in water is scarce areas (FAO, 2003). Abiotic stress results from the development of economic sectors, population growth and the competition for water resources (Laraus, 2004). Water stress severity is expected to increase because of climate change, which resulted from increasing evaporation due to global warming (Sheffield *et al.*, 2012). Water stress influence more than 10 % of arable land; leading to desertification, while salinization is rapidly increasing dramatically declining average yields of different crops. Moreover, increasing salt concentration in the soil layers decreased the water potential of soil that badly affects plant tissue relative water content and plant water conductance (Munns, 2002). Excess salts accumulation in soils cause reduction in water potential values of soil solution that causes difficulty for plants to absorb the water from soil leading to "osmotic stress." High salts decrease plant development because these big amount of salts stimulates the

### Salinity:



**Fig. 2:** Effects of salt stress on plants (Evelin *et al.*, 2009)

Salinity is one of the main abiotic stresses in arid and semi-arid areas where more than 6% of the world surfaces are salt affected. NaCl is the greatest pervasive, superabundant and soluble salt in the world (FAO, 2008). In the arid and semiarid regions, lack of organic and inorganic nutrients and high rate of evapotranspiration causing increase in soil salinity and sodicity (Shannon *et al.*, 1994). The major cause of salty soil could be the use of low quality irrigation water. Where, the high quality water is always used for domestic purposes or in industry and polluted or salty water is used for lands cultivation (Bouwer, 2002). Salty soils can be divided according to the electrical conductivity of the soil saturation extract (EC) and by definition, in to soils with EC<sup>s</sup> of 4 dS/m or more are accounted as saline soils and soils with EC<sup>s</sup> greater than 15 dS/m which accounted as strongly saline soils (Omami, 2005).

The factors that responsible for soil salinization are many in number, such as human activities, climate, topography of lands, and salt constituents (Blumwald *et al.*, 2004). According to salt composition, numerous anions and cations are responsible for soil salinization but the major important ion precipitate are Cl<sup>-</sup> and Na<sup>+</sup> where Cl<sup>-</sup> causes high toxicity and nutrient imbalances in plants, while Na<sup>+</sup> particularly causes the soil dispersion (Hasegawa *et al.*, 2000). Increasing the concentrations of salt in soil layers alters plant natural process such as molecular, biochemical and physiological processes as well as crop yield that is reduced by soil or water salinization in terms of quantity and quality (Silvera *et al.*, 2001).

Intensity of salinity depends on the physics and chemistry of soils, the concentration of salt in irrigation water, plants type, irrigation schedules and plant growth stages (Vicente *et al.*, 2004). To cope with the problem of salinity, it is important to improve the salt tolerant genotypes. At low salinity levels the plant injury are caused by nutritional imbalances, osmotic stress, and ion toxicity (Wahome *et al.*, 2001). While, under moderate up to high salinity levels, the nutritional imbalances are caused by the

interferences of salt ions and their toxicity, which resulted from the accumulation of ions especially Na<sup>+</sup> and Cl<sup>-</sup> which are the major cause of salinity on biochemical and physiological composition in different crop plants (De-Pascale *et al.*, 2003). Efforts to produce salt tolerant genotypes need a good understanding of the effects of salinity on plants of different crops, responses of plants in terms of, biochemical, molecular and physiological activities to salinity and recognition of complex mechanisms of salt tolerance in plants.

### 1. Morphological effects of salt stress on crop plants:

Many researchers recorded reduction in plants growth under salt stress conditions, but the level of this reduction depended on environmental conditions, level of salt, stages of growth and type of plants. Salinity causes reduction in germination rate, germination index, seedling length, germination percentage, root /shoot length ratio and seed vigor (Khodadad, 2011). Salinity inhibits rapidly stems and leaves growth, whereas roots elongation may increase. Ion toxicity is the main cause of growth reduction under saline conditions (Chinnusamy *et al.*, 2005). Many researchers recorded that plant growth reduced under saline irrigation condition. The first effect of salinity on plants is the reduction in its growth parameters caused by the fall in osmotic potential which reduced the uptake of nutrients and water by stressed roots (Jose *et al.*, 2017). Root and shoot growth reductions are clearer and cause severe, senescence, necrosis, and chlorosis, of old and young leaves (Munns, 2002). Salinity has also been found to change the morphology of root system and decrease the plant total root length (Álvarez *et al.*, 2014). A general reduction in fresh and dry weights has been recorded in most plant tissues exposed to salinity, and it is especially noticeable in the shoot system. Different researchers have revealed the reduction in fresh and dry weights to the decrease in the number of leaves or in leaf abscissions. Another typical response to salt stress is a reduction in total leaf area (Jose *et al.*, 2017).

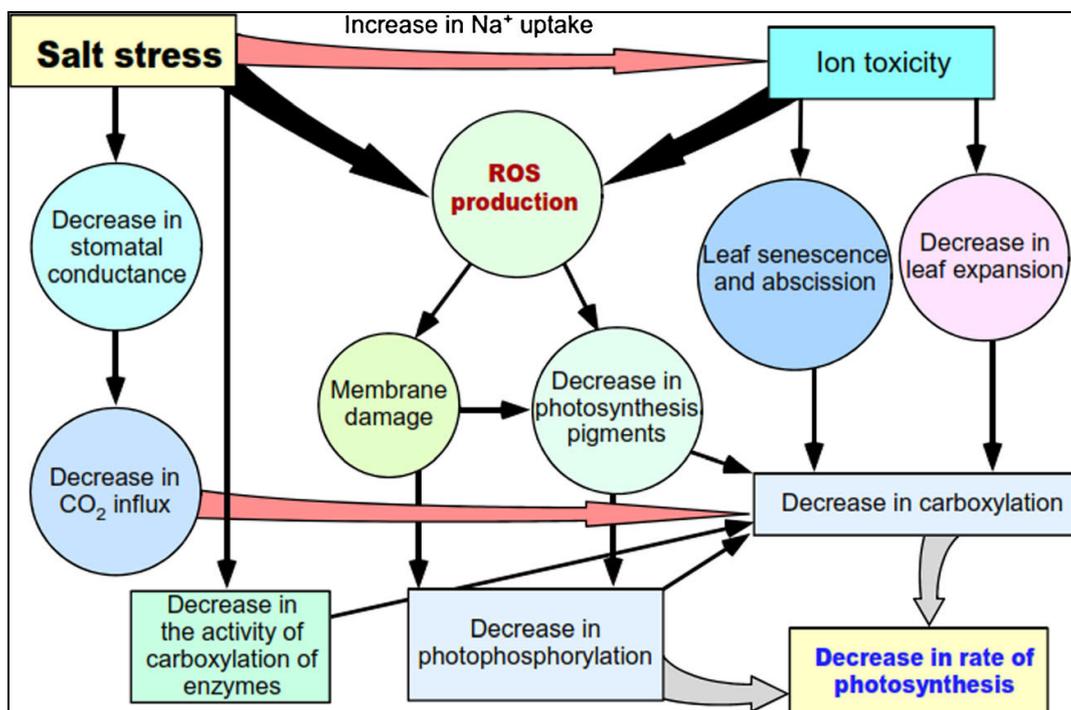


Fig. 3: Influence of drought stress on morphological, physiological and biochemical characters (Farooq *et al.*, 2015).

The decrease in leaves number as a result of salinity could be contributed to its direct effect on cell division which resulted from fall in nucleic acid synthesis and/or stimulation of its break down. The decrease in leaf number because of salt stress could also be due to the increase in leaf abscission caused by hormonal imbalance that caused by the increase in ABA and decrease in IAA levels in stressed plants if compared to control leaf (Wu *et al.*, 2005). The reduction in the leaf area might be considered as a resistance mechanism that minimizes the loss of water through transpiration (Ruiz-Sánchez *et al.*, 2000). Increasing salt concentration in irrigation water was limiting leaf area, caused plant growth reduction, and changing the relation between root and the aerial parts. Salinity stress makes different crop plants showed drier root mass than shoot, causing increase in root to shoot ratio (Fernández-García *et al.*, 2014). The changes formed in leaf anatomy are also an important method to study the effect of abiotic stress, including salt stress on different crops (Acosta-Motos *et al.*, 2015). A field experiment was carried out by Longstreth and Nobel (1979), to study the effect of different salinity levels on three plants with different responses to salinity (*Atriplex patula*, salt-tolerant plant; *Gossypium hirsutum*, moderately salt-tolerant plant; and *Phaseolus vulgaris*, salt-sensitive plant). To found the changes that happened in plant leaf anatomy, these scientist measured different leaf parameters, like leaf succulence, epidermal, mesophyll and leaf thickness; the surface area of mesophylls per unit leaf surface area; the diameter of spongy cells; the length and diameter of palisade cells and the ratio of mesophyll cell surface area to leaf surface area. The salt-tolerant specie which were irrigated with different saline solutions (0.05, 0.1, 0.2, 0.3 and 0.4 M), revealed more leaf thickness resulted from the increase in mesophyll and epidermal thickness. Finally, great increases were obtained in the leaf succulence records. While opposite effects were obtained in the other two species which were less tolerant to salt stress. In addition, Romero-Aranda *et al.* (1998) recorded anatomical disturbances caused by chloride salts such as NaCl, KCl and  $\text{CaCl}_2$  in both tolerant (*Cleopatra mandarin*) and sensitive (*Carrizo citrange*) citrus varieties. They noticed changes in leaf anatomical characters in both varieties, such as the increase in the lower area/volume ratio of mesophyll cells and leaf thickness. Salt stress also reduced the intercellular air spaces and increased the succulence of leaves and the surface/volume ratio of tissue and cells density. These results indicated that irrigated citrus crop with saline water caused increase in leaf thickness combined with several metabolic changes such as low  $\text{Mg}^{+2}$  content, low chloride overloading, chlorophyll damage and stomatal closure, which may contribute decline in photosynthesis. Navarro *et al.* (2007) noticed also anatomical changes under saline conditions in *Arbutus unedo* leaves by optical microscopy in cross sections. A comparison between saline-irrigated and control plants revealed that no significant difference in the cell size of the upper palisade layer. While, significant increases appeared in the cell size of the lower palisade layer which in parallel with the levels of NaCl salinity (0 mM, 52 mM and 105 mM NaCl). These authors also observed a great reduction in the intercellular air spaces

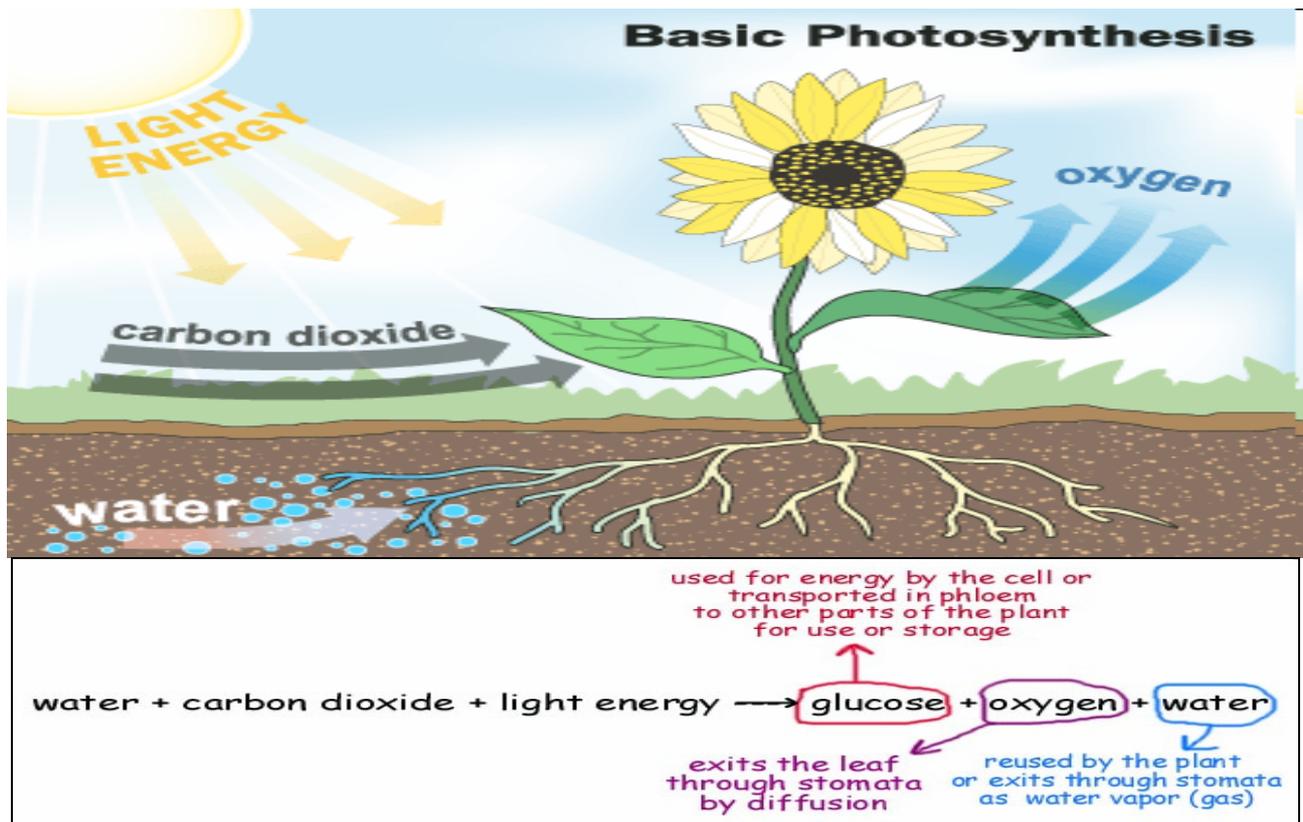
in the spongy mesophyll tissue in stressed leaves compared to untreated leaves, and this reduce the conductance of  $\text{CO}_2$ . Fernández-García *et al.* (2017) studied the leaf mass/ leaf area ratio in henna plants treated with high salinity irrigation level. Stressed henna plants react to high and low salinity by improves leaf mass to leaf area. They also observed that high salinity level caused increase in leaf thickness in salt-stressed henna plants which may maximize the photosynthesis potential. Furthermore, Kelij (2013) reported decrease in the numbers of vascular bundles under salt stress conditions in stem of *Aeluropus littoralis*. In addition, Naz *et al.* (2013) showed reduction in the metaxylem area of five ecotypes of *Aeluropus lagopoides* by increase in salt concentration at different ecotypes. Behrouz *et al.* (2017) recorded also in some halophytes, sharp reduction in xylem vessel diameter after salinity treatments which reduced up to 800 mM, also they noted reduction in phloem diameter under NaCl stress. They added that the most anatomical and morphological characters like stem diameter, stem length, metaxylem diameter, vascular bundles, and phloem diameter were reduced under salt stress conditions.

Similar changes in morphological characters under salt stress conditions were obtained by Akıncı and Lösel (2010) who mentioned that the stress conditions caused great decrease in stem height, leaf area index, leaf number, fresh and dry weights of cotton (*Gossypium hirsutum*) plants and some *Cucurbitaceae* species. They added that the stress conditions not only influence on morphological characters but also changes bio-mass ratio. Similar results observed on *Salvia officinalis* (Ben Taarit *et al.*, 2009); *Thymus capitatus* (Ezz El-Din *et al.*, 2009 and Khalil, 2016); *Nigella sativa* (Hussain *et al.*, 2009); and basil (Said-Al Ahl and Mahmoud, 2010).

## 2. Physiological effects of salt stress on crop plants:

### 2.1 Effect of salinity on photosynthesis:

Photosynthesis process is the most important process in which green plants make their own food, they convert solar energy into chemical energy and produced organic compounds and oxygen by  $\text{CO}_2$  fixation. Photosynthesis process is badly affected by salt stress in different ways, like the inhibition of  $\text{CO}_2$  fixation and concentration due to stomatal closure, the reduction or destruction of photosynthetic pigments including carotenoids, chlorophyll a and chlorophyll b (Qados, 2011), and adversely affect the photosynthetic processes, and electron carrier (Sudhir *et al.*, 2005). The inhibition in photosynthesis process due to salinity resulted from reduction in chlorophyll concentration and content. Total photosynthesis rate declined due to decrease in leaf characters such as reduction in leaf expansion and development, as well as increase in leaf abscission, increase the exposure to salinity caused, membrane damage, complete stomatal closure, ion toxicity, which become the main factors responsible for photosynthetic diminish. Generally, the total carotenoid and chlorophyll concentrations in plant leaves are reduced under salt stress condition where the chlorosis begin in the oldest leaves during the salinity stress (Farooq *et al.*, 2015).



Iyengar and Reddy (1996) reported decreases in photosynthetic rate under saline conditions caused by number of factors including:

1. Reduced water availability and high osmotic potential in plants cells caused decrease in the  $\text{CO}_2$  conductance and cell membrane dehydration as well as reduction in photosynthetic electron transport due to reduction in intercellular spaces.
2. Toxicity of sodium chloride ions, the  $\text{Cl}^-$  ion prevent  $\text{NO}_3^-$  and N uptake by the roots caused inhibition in photosynthetic rate, and consequently reduced the absorption of  $\text{NO}_3^-$  and N which related to the osmotic stress that may explain the bad effect of salt stress on photosynthesis process (Fisarakis *et al.*, 2001).
3. The complete close of stomata due to abscisic acid accumulation which causes decrease in  $\text{CO}_2$  supply so that the availability of  $\text{CO}_2$  for photosynthesis process restricted.
4. Changes in enzyme activities.
5. Advancing senescence of leaves induced by salinity stress.

The reduction in leaves number due to salt stress, could be due to its direct effect on cell division that caused by decrease in nucleic acid formation and/or stimulation of its break down (Ashraf and O'leary, 1996). The reduction in number of leaves as a response to stress conditions could also be contributed to stimulation of leaf abscission caused by hormonal imbalance that resulted from decreased IAA levels and increased ABA level in stressed as compared with control leaves (Wu *et al.*, 2005). Moreover, the total leaves area /plant badly reduced by the increase in severity and duration of stress conditions, especially in the sensitive varieties. furthermore, Chaitante *et al.* (2000) recorded that salt stress greatly reduced leaf area expansion and root length but these two processes are not equally influenced, root

growth is reduced to a less extent than leaf growth, and photosynthate partitioning is changed due to increment in root/shoot ratio (Akıncı and Lösel, 2009).

The two important factors causing stomatal closure are always the reduction in water potential of leaf tissues and cells and the increase in the content of gaseous carbon within the leaves (Schulze, 1986). Since, stomata limitation is drastically affected when a plant is under stress condition than when it is not, accordingly photosynthesis always reduces in parallel with, or greater than, stomatal conductance (Cornic *et al.*, 1989). Massaci *et al.* (2008) indicated on cotton (*Gossypium hirsutum*) that photosynthesis process was hardly affected by stress conditions. In the coconut palm, the stress caused photosynthetic inhibition, which is due to the reduction in  $\text{CO}_2$  permeability from the surrounded atmosphere to the leaf intercellular spaces due to stomatal closure (Gomes and prado, 2007). In addition, salinity could induce reversible and significant alterations in the physiological stress indicators which related to plant water relations, leaf gas exchange and growth, but they were unable to induce any changes in the photochemical activity of young *J. curcas* plants (Silva *et al.*, 2010b), although salt stress induced significant reduction in leaf chlorophyll content (Silva *et al.*, 2010). This decline in the photosynthetic pigment accumulation as a response to salt stress was due to the ultra-structural damage of plastid tissue especially the membranes protein of thylakoids that in turn causes untying of photo system 2 that captures photons, so its efficiency decreased, thus causing ATP and NADPH production, decreases in electron transfer, and eventually  $\text{CO}_2$  fixation processes (Abdalla and El-Khoshiban, 2007) on different wheat varieties. Similar results also obtained by Topbjerg *et al.* (2015) on different wheat varieties. Najar *et al.* (2019) also reported on *Medicago truncatula* reduction in photosynthesis rate by the inhibition of  $\text{CO}_2$  assimilation and conductance caused by PSII damage.

## 2.2. Effect of Salinity on photosynthetic pigments:

Several researchers reported decreases in total carotenoid and chlorophyll contents of plant leaves under salt stress, where the chlorosis begin in the oldest leaves at salt stress conditions (Agastian *et al.*, 2000). There is strong evidence that salt stress reduced chlorophylls, carotenoids and photosynthetic enzymes, (Stepien and Klobus, 2006). Kennedy and De Filippis (1999) recorded declined in the concentration of protochlorophyll, chlorophylls, and carotenoids of *Grevillea licifolia* plant. They also indicated that the reduction in protochlorophyll was more than Chla and carotenoids are more decreased, where anthocyanin pigments are significantly increased in this case under salt stress. Also, Amuthavalli and Sivasankaramoorthy (2012) demonstrated on pigeon pea that salt treatments caused diminish in the content of all photosynthetic pigments and to a certain extent changed their ratios. They returned the decrease in their content to plant mineral deficiency. They also added that carotenoid decreased to a lesser extent than Chl. Moreover, Meloni *et al.* (2003) revealed also that ion accumulation in leaves of stressed plants adversely affected Chlorophyll content. In addition, Garsia-Sanchez *et al.* (2002) explained that the decrease of Chlorophyll concentration in the plants grown under saline conditions could be due to both the inhibition in synthesis of that pigment and to the increase in their degradation. They also added that, Chla was better protected or less sensitive to salt stress compared to Chlb. Moreover, Khavari-Nejad and Mostofi (1998) stated in an experiment on salinity effects on leaves of different tomato cultivars that the concentration of carotene and total chlorophyll were reduced under NaCl stress. Similar results obtained by Alamgir and Ali (1999) who stated under salinity conditions, that the leaf pigments in nine varieties of rice plant were decreased. Furthermore, Lu and Vonshak (1999) reported that the ratio of carotenoid/chlorophyll in the *Cyanobacterium (Spirulina platensis)* did not changed, but there was a significant reduction in the phycocyanin/ chlorophyll ratio. Moreover, Rahneshan *et al.* (2018) found that the photosynthetic pigments, including chla, chlb, and total chl, significantly decreased by high and moderate salinity levels in sensitive plants, while they remained unchanged in tolerant plants. The reduction of in the content of these pigments under salt stress conditions is caused by the fast break down or slow synthesis of these pigments in plant cells. Several researches have been stated the role of carotenoids pigments as an effective antioxidant that stabilize and protect the photochemical processes of photosynthesis under salt stress condition. Carotenoids play an important role in preventing the chlorophyll-photosensitized formation of  $^1O_2$  by intercepting chlorophyll triplet states and protection of photosynthesis. Accumulation of carotenoids in different crops for osmotic regulation in stressed leaves had been recorded by Khan *et al.* (2001) on wheat, Gunes *et al.* (2008) on chickpea cultivars, and Khalil and Abdel-Salam (2015) on *Origanum vulgare*. On the other hand, Wang and Nil (2000) showed that salt stress induced increases in chlorophyll accumulation in *Amaranthus* plant.

## 2.3. Effect of Salinity on osmotic adjustment mechanisms and water relations:

Salinity stress diminished the leaf water relations including leaf turgidity percent, turgor potential, water potential, water relation parameters, osmotic potential, as

well as plant growth, and plant fresh weight. Under salinity stress, water potential of the soil solution become reduced that inhibited seed germination caused by sodium toxicity, which make water absorption is difficult by seeds. Moreover, the relative water content declined due to salt application, the reduction in relative water content, and a loss of cell turgor reduced water availability for cell elongation (Jabeen and Ahmad 2012). There are two important things:

- i) In high salinity levels, plants accumulate great amounts of  $Cl^-$  and  $Na^+$  ions in their leaves than normal situation that caused decrease in leaf osmotic and water potentials and induced more reduction in their water potentials.
- ii) The reduction in hydraulic conductance of the root caused decrease in the water quantity that flow from the root system to the leaves, resulting in water deficiency in the leaf tissues.

Osmotic potential resulting from the net accumulation of different types of solutes in response to water stress resulting from salinity or drought by keeping cell turgor, also osmotic adjustment necessary to allow growth to continue under low water potential condition (Gomes *et al.*, 2006). Turner and Jones (1980) had defined osmotic adjustment as “the lowering water deficits or salinity”, osmotic adjustment always depends on photosynthesis process to get the compatible solute when cell dehydration becomes dangerous, and photosynthesis is delay causing in a less amount of solute supply for osmotic adjustment mechanism. With continued water stress as a result of salinity, osmotic adjustment delays, but cannot completely stopped. Silva *et al.* (2010) explain osmotic stress as a physiological event often related to excessive water deficit that can decrease plant growth parameters. They added that the osmotic adjustment is a cellular adaptive process related to stress-tolerant plants, that making them able to complete growing under drought or salt stress, which is usually defined as a decrease in the cell sap osmotic potential resulting from a net increase in intracellular solutes rather than from a loss of cell water (Kusaka *et al.*, 2005). This mechanism had been considered as an important physiological adaptation characteristic associated with drought or salt tolerance plants, and it has drawn much attention during the last years (Hussine *et al.*, 2009).

Several physiological works indicated that at stress conditions, non-structural carbohydrates including sucrose, alcohols, sugar, and hexoses increased in amount in different plant tissues. However, seeds of different plants accumulated large amounts of total soluble sugar and proline for osmotic regulation as a response to salinity, which improved their water relations and the enzyme activities which regulate the process of germination. Plants also accumulate organic solutes like glycine betaine, sucrose, glutamic acid and proline (Hussain *et al.*, 2016). They also added that the accumulation of these and other organic ions increase osmotic regulation, resulting in an increase in osmotic pressure and decrease in water potential resulting in inward diffusion of water from the surrounding media which lead to maintenance of cell turgor and cell expansion.

Increasing salt concentration in root area decreases the water potential of leaf and consequently, the uptake of nutrients and water by plants root are hard or impossible (Romero-Aranda *et al.*, 2001). Lu *et al.* (2002) in a study on halophyte plant recorded that evaporation rates and leaf water potential reduced by increasing salt concentration. Similar

results were recorded in *Brassica* genus (Sohan *et al.*, 1999). According to Song *et al.* (2005) as a result of ionic toxicity and osmotic stress there were a decline in halophyte *Sueda* germination and when salinity stress alleviated, seed germination improved. So, water loss must be limited either by adjusting osmotic potential or regulation of transpiration. Furthermore, Ghoulam *et al.* (2002) on sugar beet varieties indicated that the relative water content decreased due to salt application. Katerji *et al.* (1997) recorded reduction in relative water content and a loss in cell turgor. Many researches had illustrated that the increase in proline concentration was noted in salt-tolerant than in salt-sensitive varieties (Kumar *et al.*, 2010 on rice; Mansour and Ali, 2017). It is evident that high content of compatible solute like proline in plant cell under salinity stress condition involved in adaptation to tolerate osmotic effects which cause reduction in the cellular water potential to a level below the external water potential (Hossain *et al.*, 2015 on soybean). They also added that proline has been reported for ROS scavenging molecule to protect plants body under abiotic stress conditions; and alleviating the bad effect of oxidative injury in plant tissues and cells. Nounjan *et al.* (2018) showed that proline accumulation involved in osmo-protection process and ROS scavenger functions.

#### 2.4 Effect of Salinity on nutrient contents and ion levels:

Under salinity conditions, uptake of  $\text{Na}^+$  and  $\text{Cl}^-$  ions antagonism with the absorption of some important elements like  $\text{K}^+$ ,  $\text{Ca}^{2+}$ , N, and P by plants, causes nutritional imbalance and reduction in yield quality and quantity (Grattan and Grieve, 1998). Several studies recorded that increase in NaCl amount in root area of different crops results in accumulation of  $\text{Na}^+$  and  $\text{Cl}^-$  ions in shoot organs and reduce in  $\text{Mg}^{2+}$ ,  $\text{Ca}^{2+}$ , and  $\text{K}^+$  levels in different crops (Bayuelo-Jimenez *et al.*, 2003). The transport of anions and cations from soil solution to the cell cytoplasm is controlled by cell membranes permeability which has protein transporters that allow the passage of ions through it (Jiménez-Casas, 2009). The passage of  $\text{Na}^+$  ion through the cell membrane is inactive process and it needs membrane proteins to make its passageway possible (Torabi *et al.* 2011). White and Broadley (2001) reported that  $\text{Cl}^-$  passage through cell membrane could be happened by both passive and active ways, and it depends on the external concentration of  $\text{Cl}^-$  ions. Active transport of  $\text{Cl}^-$  through plasma membrane happened only under the low  $\text{Cl}^-$  concentrations in the external soil solution (Munns and Tester, 2008). By increasing sodium chloride concentration, the  $\text{K}^+$  amount in plant leaves decreased hence it does not affected in sugar beet root (Ghoulam *et al.*, 2002). Several studies reported that the uptake of nitrogen is affected by salt stress so that salinity stress decreased N concentration in different crop (Silvera *et al.*, 2001). Increasing the amount of  $\text{Cl}^-$  ions in eggplant leaves was associated with decreases in the uptake of  $\text{NO}_3^-$  (Savvas and Lenz, 2000). According to growth stages, levels of salinity and plant species, the concentration of phosphorus will change under saline stress conditions. In most crops, salt stress decreasing P content in plant cells (Kaya *et al.*, 2001). The excessive accumulation of sodium ion ( $\text{Na}^+$ ) results in ion imbalance, ion toxicity and decrease in plant water uptake also it interferes with plant vital process, while the increase in potassium ion ( $\text{K}^+$ ) concentration in plant cell can reduce  $\text{Na}^+$  ion toxicity through ion balance and osmotic potential adjustment. It has

been indicated that high  $\text{Na}^+$  amount causes sharp reduction in  $\text{K}^+$  and  $\text{Ca}^{2+}$  content that resulted in a sharp reduction of  $\text{K}^+/\text{Na}^+$  ratio after treating plant with higher salinity level, which reveals greater leaves damage compared to those in roots (Munns 2008), leaves burning is an important symptom of salinity stress (Zhu, 2003). In salty soils, high concentration of  $\text{Cl}^-$  and  $\text{Na}^+$  ions in plant root causes strong nutritional disorder in plant. This is due to the antagonism of these ions with other important nutrients elements like, potassium calcium, magnesium, nitrogen, phosphorus, manganese, copper, iron and zinc (Siddiqi *et al.*, 2011). This confirms the view of many authors such as Fortunato *et al.* (2012) on banana plants, Sayed and Gadallah (2014) and Barbos *et al.* (2015) on *Zea mize*.

### 3. Biochemical effects of salt stress on different crops:

#### 3.1 Effect of salinity on protein content:

Salt stress enhance the formation of reactive oxygen species ROS, like hydroxyl radical, hydrogen peroxide, superoxide and single oxygen radicle. Salinity stress-stimulates production of ROS, which causes oxidative damages of macromolecules like DNA, lipids, and proteins, which disturb vital cellular functions of plants (Ahmad and Umar, 2012). Stress conditions bring about qualitative and quantitative changes in plant cell proteins. Under salt condition accumulation of protein in plant cells could play an important role in plants salt tolerance, where the proteins may use as an adjuster of osmotic potential in plants subjected to salinity or may use as a reservoir of energy (Parvaiz and Satyawati, 2008). They recorded that the proteins which formed under salinity are cytoplasmic that can induce changes in cytoplasmic viscosity of the plant cells (Hasegawa *et al.*, 2000). In general, proteins in the plant leave of most crops reduced during water deficiency as a result of the suppressed synthesis or due to its degradation (Dubey, 1999). Salinity inhibits the synthesis of new proteins and changes gene expression. Moinuddin and Chopra (2004) stated that osmotic adjustment involves an active accumulation of different types of cellular solutes such as soluble proteins within the plant cells in response to lowering of the cellular water potential and reducing the harmful effects of water deficit which caused by salt stress. As a consequence of solutes accumulation the water potential of plant cell is lowered, which attracts water into the cell and leads to maintaining its turgidity. Ashraf and Harris (2004) reported that in salt tolerant varieties of sunflower, rice and barley there were a high accumulation of soluble proteins. Osmotin is also a kind of protein that accumulated at salt stress conditions in many crop plants like triplex, tomato, tobacco, and maize (Valentovic *et al.*, 2006). Dehydrins have been the main noticeable group among the accumulated proteins under salt stress and increased in pea, barley, maize, and *Arabidopsis*. Moreover, Kshamata *et al.* (2005) on cotton (*Gossypium hirsutum*) plant indicated that LEA proteins could be considered as a novel form of molecular chaperone which help in preventing the synthesis of damaging protein aggregates under stress conditions. Several proteins were reduced by stress in maize mesocotyls (Valentovic *et al.*, 2006). Progressive water deficit as a result of salt stress increased the concentration of soluble proteins in chickpea leaves; the accumulation of the osmolytes can help the chickpea plants to maintain the structural integrity of membranes and the cell turgor (Najaphy *et al.*, 2010). These results were online with many studies done in this field e.g.,

Dasgupta and Bewley (1984) who indicated that salinity decreased protein formation in all parts of barley leaf. In addition, Vartanian *et al.* (1987) reported presence of stress specific proteins in tap root of *Brassica* species. Win and Zaw (2017) indicted on *Vigna Mungo* L. that salt stress stimulated the accumulation of salt-specific proteins and altered the protein profiles in different crops; they added that the change in protein profiles and accumulation were depended on plant genotypes, salt duration and the salt concentration. The changes of protein profiles under salinity stress were not detected in sensitive genotype, while, the accumulation of newly synthesized protein bands were detected only in salt tolerant genotype.

### 3.2 Effect of salinity on plant amino acids:

Some amino acids such as alanine, proline, arginine, leucine, glycine serine, valine, and amides (asparagines and glutamine) as well as the non-protein amino acids (ornithine and citrulline) accumulate in plants exposed to salt stress (Torabi *et al.*, 2010). In addition, Hussain *et al.* (2016) indicated that the accumulation of total free amino acids were greater in leaves of salt tolerant plants than in salt sensitive lines. Proline content is considered to be the first response of plants under salt stress to decrease the leaf osmotic potential. Abdalla and El-Khoshiban (2007) noted that there was a direct proportional relationship between the proline accumulation in roots and shoots of two *Triticum aestivum* cultivars and the duration and severity of stress compared with control varieties. Also, tolerant varieties had more proline content than sensitive ones. They returned the increments in proline concentrations under stress conditions to the defense mechanism that stressed plants take to decrease the cell osmotic potential, thus promoting inward water uptake with the increases in both cell activity and its turgidity. These findings were confirmed by many studies done in this field e.g., El-Tayeb and Hassanein (2000) on *Vicia faba*; Sawhney and Singh (2002) on wheat and Zhang *et al.* (2006) on soybean. Moreover, Yoshida *et al.* (1997) and Phutela *et al.* (2000) on *Brassica juncea* mentioned that proline concentrated in cells of stressed plants as a result of the decreased rate of its degradation by proline oxidase enzyme and the increased rate of its synthesis by pyrroline-5-carboxylate synthetase.

### 3.3 Effect of Salinity on plant carbohydrates:

The increase in soluble carbohydrates accumulation in tissue and cells of different crops had been greatly recorded as a response to salt stress, which cause a significant reduction in CO<sub>2</sub> assimilation rate (Murakezy *et al.*, 2003). When the plant is treated with high salinity levels, the increment in reducing sugars reaches to 50% increase (Parvaiz and Satyawati, 2008). Parida *et al.* (2002) mentioned that soluble sugars like mono and disaccharides (fructose, sucrose glucose) and polysaccharides such as starch are increased in amount under salinity stress conditions and had an important role in osmoprotection, radical scavenging, osmotic adjustment, and carbon storage. Parida *et al.* (2002) illustrated on *Bruguiera parviflora* leaves that salinity stress increased reducing and non-reducing sugars and reduced starch. In corroboration, Khavari-Nejad and Mostofi (1998) reported that the concentration of total saccharides and soluble carbohydrates are increased significantly, while the starch concentration was not changed in leaves of tomato. Singer and Lindquist (1998) in a

research on abiotic stresses recorded that cell content of trehalose which is considered as disaccharide accumulates under different abiotic stresses conditions and protects cell proteins and cell membranes when plant treated with salt stress that led to water deficit and decreased aggregation of denatured proteins. In addition, trehalose prevents cell death and there are some evidence that trehalose is present in tiny amounts in vascular plants (Yamada *et al.*, 2003). Sami *et al.* (2016) contribute the increase in reducing sugars to the osmotic potential of plant tissues and the osmotic adjustment process. The accumulation of reducing sugars in many plants under stress conditions were on line with those recorded by He *et al.*, 2006; Sami *et al.*, 2016 and Yassen *et al.*, 2018.

### 3.4 Effects of salinity on plant lipids:

Abiotic stress may alter the association between membrane proteins and lipids as well as permeability capacity of the cell membranes and enzymes activity. Lipids, considered as one of the major components of the cell membrane, which affected by stress. In plant tissue, polar acyl lipids are the major lipids formed the cell membranous structures (Bishop, 1983). Glycolipids are the main component of chloroplasts membranes which formed about 60% of the membrane and phospholipids are the main important plasma membrane and mitochondrial lipids (Harwood, 1980). Where, triglycerides (fats and oils) are an effective storage form of carbon, at different growth stages especially in seeds (Taiz and Zeiger, 1991). The role of membrane proteins are affected by the lipid bi-layer, where they are either bound at the surface or embedded in. The structure of lipid altered by the environmental conditions are always depend on the physical characters of the lipids unclouded in the cell membrane structure, like fluidity and temperature, that could affect the transport capacity of bio membranes (Gigon *et al.*, 2004). Many workers had reported the effect of various levels of salinity on lipid composition and content in different parts of plants. Wang *et al.* (2002) demonstrated that the stress conditions (water deficiency) could led to membrane dehydration, increase membrane transport capacity and the accumulation of free radicals in maize plants. GeTi-da *et al.* (2006) noted on maize plant that salt stress acted directly on roots, which suffered severe damage because of soil drought and salinity as MDA (the product of lipid peroxidation) accumulated, cell membranes broke down as well as decreases in endogenetic protective enzyme activities. Moreover, Júnior *et al.* (2008) on *Phaseolus vulgaris* plant demonstrated that the increment in the transport capacity of the membrane due to water deficit may happened in response to an increased in passive diffusion, which may cause by the lipids membranes damage, the salinity stress led to some fatty acid contents modifications, inducing unsaturation index decreases and saturation increases, the polar lipids damage which related to the change in the saturated/unsaturated relation and a decrease in unsaturated index explain the biggest loss of electrolytes and consequent dehydrated plants cellular membranes. Similar findings were observed in lipid analysis in leaves of cotton under stress conditions (Pham Thi *et al.*, 1987), the plants *Ramonda nathaliae* and *Ramonda serbica* (Stevanovic *et al.*, 1992); the cowpea plants (Monteiro de Paula *et al.*, 1993); the coconut tree (Repellin *et al.*, 1997), rape plant (Benhassaine-Kesri *et al.*, 2002) and on halophytes (Vizetto-Duarte *et al.*, 2019).

### Plants tolerance to salt stress

Plants enhance the metabolism, physiology and morphology of their tissues and organs to increase its yield and productivity at saline conditions. The reactions of the plants to salinity differ significantly according to duration and intensity of stress as well as plant species and according to the various organizational levels and upon its stage of development. Stress resistance in different crops is classified to two groups, which are stress avoidance and tolerance. Stress avoidance in plants is the ability of plant to keep the cellular water potential high in their tissue and organs under stress conditions, science stress tolerance is the ability of the plant to keep their vital functions even when the tissue water potentials is low. Stress tolerance is happened by different cells and tissues by undergoing certain molecular, biochemical and physiological mechanisms that involved accumulation of certain proteins and specific gene expression. While stress avoidance is always happened via morphological modifications, including decreased leaf area reduced stomatal conductance, increased root/shoot ratios and development of extensive root systems (Nagarajan and Nagarajan, 2010).

### Biotechnology and abiotic stress

Great efforts of breeders and plant physiologists during the last 30 years, had concentrated on improving the stress tolerance of some horticultural and agricultural crops. It is obviously that, with the greater world need for food, there is a requiring of immediate improves of the tolerance of many crop plants and to develop more management mechanisms and techniques to maintain food demands at levels near to that needed in spite of limited resources of water and land. Borlaug and Dowsell (2005) stated that the crop production should be doubled to keep food production at levels near to demand, by increasing crop productivity from per hectare or expanding land area for cultivation. Various approaches had been done to improve stress tolerant crops, using modern genetic technics and to improve plant breeding methods. An important approach to improve crop performance and plant resistance is to select gens that have increased yield in dry or salted lands and environments. The technics of gene transfer from more tolerant wild crop plants to the less tolerant plants by using classical genetic methods has a limited success. A partial list for the important traits for plant breeding might include water-use efficiency, hydraulic conductance, water-extraction efficiency, elastic and osmotic adjustments, and modulation of leaf area. For enhance plant tolerance, it is mainly based on the manipulation of genes that maintain and protect the structure and function of cellular components. Present engineering strategies based on the transfer of one or several genes responsible for stress responsive pathways. Although the current efforts are concentrated in improve plant stress tolerance by gene transformation and it has resulted in important achievement (Cherian *et al.*, 2006).

### Stress alleviation by using organic and soil microbes or bio fertilizers:

Plants have complex mechanisms to tolerate abiotic stresses caused by various ecological factors, including salinity and drought. Fertilizers are used to improve tolerance of most crops and soil fertility but excess use of inorganic fertilizer in agriculture causes unrecoverable environmental pollution and so many health problems. Inorganic fertilizers are known for their negative environmental effects and their

high cost (Morris *et al.*, 2007). In this regard, attempts have recently been made towards the production of high quality nutrient rich fertilizer (Bio and organic fertilizer) to ensure bio-safety. The choices of suitable fertilizer are controlled by different factors such natural conditions, locality, climate, and soil type and their suitability for crops cultivation. The leafy vegetables, cereal crops, and fruits are efficient source of basic nutrients such as phosphorus, nitrogen, potassium, and micronutrients such as magnesium, boron, calcium, and manganese. To eliminate the bad effects of chemical fertilizers on the environment, and human health, recently a new agricultural practice developed known as sustainable agriculture, ecological agriculture or organic agriculture (Chowdhury, 2004). Organic farming is one of most effective strategies that not only ensure biodiversity of soil but also food safety (Khosro and Yousef, 2012). Organic fertilizers are primarily cost effective (Solomon, *et al.*, 2012). Organic fertilizers are the basis of soil fertility. Organic fertilizers are come from living or biological materials. To release the nutrient in the soil from organic fertilizers long time is needed. Organic fertilizers are different forms like: Manure which come from livestock like goats, chickens, and cows. Green manure derived from plants remains, especially different type of legumes. Compost, which obtained from agricultural waste organic material like decomposed waste, corn stalks or straw. Furthermore, bio fertilizers are non-bulky, environment friendly, cost effective that plays important role in plant nutrition (Mahajan *et al.*, 2008). In addition, Bio-fertilizer considered as an alternative to chemical fertilizer to improve crop production and soil fertility in sustainable agriculture. These biological fertilizers could act as the key role that increasing productivity and sustainability of soil protect the environment as eco-friendly for the farmers and cost effective inputs. Production of tolerant crop plants under stress conditions can be also increase by using soil microorganisms (biofertilizers), which seems to be effective on the increasing tolerance of different crop plants under stress conditions. Plant associated microbe (including bacteria and fungi) in the soil alleviate the adverse effects of stresses in a more cost-effective and time sensitive manner. Research directed towards the application of bio-fertilizers in salt and drought-affected fields, which encourages commercialization of inoculants for stress resistance (Babalola, 2010). Bio-fertilizers are compounds composed of living cells of different types of microorganisms that are able to convert important nutritionally elements from unavailable to available form via different biological mechanisms (Vessey, 2003). Under salinity stress conditions, Bacteria decrease plant salt uptake by altering root structure with extensive rhizosheaths, trapping cations in the exopolysaccharide matrix, and regulating ion transporters. Microorganisms have been known to regulate nutrient imbalance caused by the high accumulation of toxic ion such as  $\text{Cl}^-$  and  $\text{Na}^+$  ions and increase the mineral nutrient exchange of both micro and macronutrients in plant body. Microbial induced nutrient cycling (mineralization), rhizosphere pH changes (organic acids), and metal chelation (siderophores) which improve plant nutrient availability in the soil (Lugtenberg *et al.*, 2013). Microbes help controlling ion homeostasis and keeping  $\text{K}^+/\text{Na}^+$  ratios high in shoots by reducing  $\text{Na}^+$  and  $\text{Cl}^-$  accumulation in leaves, increasing  $\text{Na}^+$  exclusion by roots, and increasing the activity of high affinity  $\text{K}^+$  transporters. So the use of bio and organic fertilizers seems to be an effective method to increase plant tolerant and

alleviate the adverse effects of stresses in a more cost-effective, time sensitive manner and without negative environmental effects.

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