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### Instinctive Plant Tolerance Towards Abiotic Stresses in Arid Regions

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#### 1. Introduction

Arid environments are extremely diverse in terms of their land forms, soils, fauna, flora, water balances, and human activities. Because of this diversity, no practical definition of arid environments can be derived. However, the one binding element to all arid regions is aridity. Aridity is usually expressed as a function of rainfall and temperature. A useful "representation" of aridity is the following climatic aridity index: p/ETP, where P = precipitation; ETP = potential evapotranspiration, calculated by method of Penman, taking into account atmospheric humidity, solar radiation, and wind. Three arid zones can be delineated by this index: namely, hyper-arid, arid and semi-arid. Of the total land area of the world, the hyper-arid zone covers 4.2 percent, the arid zone 14.6 percent, and the semiarid zone 12.2 percent. Therefore, almost one-third of the total area of the world is arid land.

Arid climate, is a climate that does not meet the criteria to be classified as a polar climate, and in which precipitation is too low to sustain any vegetation at all, or at most a very scanty scrub. An area that features this climate usually (but not always) experiences less than 250 mm (10 inches) per year of precipitation and in some years may experience no precipitation at all. In some instances an area may experience more than 250 mm of precipitation annually, but is still considered a desert climate because the region loses more water via evapotranspiration than falls as precipitation. Although different classification schemes and maps differ in their details, there is a general agreement about the fact that large areas of the Earth are arid. These include the hot deserts located broadly in subtropical regions, where the accumulation of water is largely prevented by either low precipitations, or high evaporation, or both. Abiotic disorders are associated with non-living causal factors such as weather, soils, chemicals, mechanical injuries, cultural practices and, in some cases, a genetic predisposition within the plant itself. Abiotic disorders may be caused by a single extreme environmental event such as one night of severe cold following a warm spell or by a complex of interrelated factors or events. A biotic plant problems are sometimes termed "physiological disorders" that reflects the fact that the injury or symptom, such as reduced growth, is ultimately due to the cumulative effects of the causal factors on the physiological processes necessary for plant growth and development (Schutzki & Cregg, 2007).

Abiotic stresses, such as drought, salinity, extreme temperatures, chemical toxicity and oxidative stress are serious threats to agriculture and the natural status of the environment. Increased salinization of arable land is expected to have devastating global effects, resulting

in 30% land loss within the next 25 years, and up to 50% by the year 2050. Therefore, breeding for drought and salinity stress tolerance in crop plants (for food supply) and in forest trees (a central component of the global ecosystem) should be given high research priority in plant biotechnology programs(Wang et al., 2003).

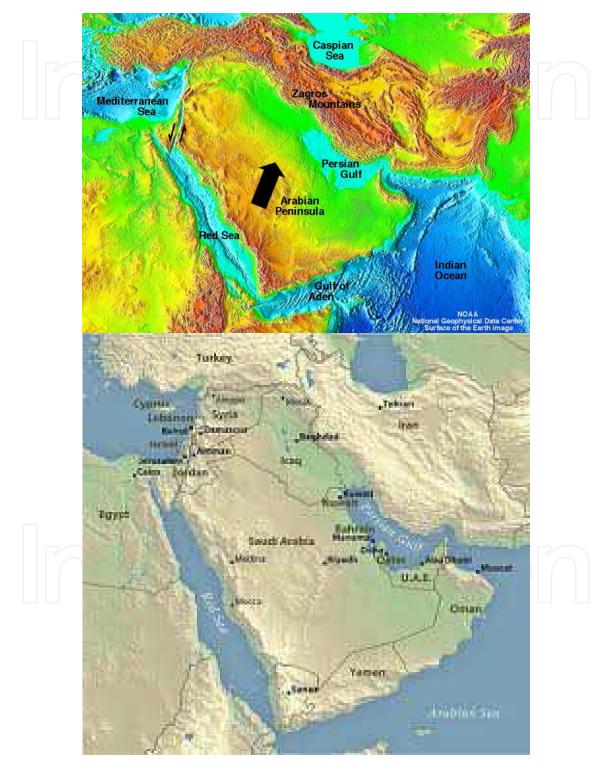


Fig. 1. The Arabian Peninsula (land-surface image formatted and labelled by Bruce Rails back); National Geophysical Data Center (NOAA).

Desert plants generally follow two main strategies i.e., they tolerate the stresses through phonologic and physiological adjustments referred to as tolerance or avoidance mechanisms contribute to the ability of a plant to survive stress but it also depends on the frequency and severity of the stress periods. Xeromorphic characteristics of desert plants have developed as the result of adaptation to drought, temperature divergence, salinity, poor nutrition, strong wind, sand movement and high light intensity (Fahn,1964,1990; Fahn and Cutler 1992; Huang et al.,1997). Plants in many habitats have various physiological mechanisms for responding to environmental changes, and the ability to tolerate environmental disturbances often contributes to their success in communities (Gutterman, 2001). In addition to genetic adaptation, the survival of a certain species is often determined by its ability to acclimate to environmental changes (Gutterman, 2002). Acclimation is known to be a widespread phenomenon in nature, and long-term responses can be observed in the course of a season.



Fig. 2. Xeromorphic characteristics of desert plants in Arid environment (Ibrahim & El-Gaely, 2011)

#### 2. Convergent abiotic stress

More than one abiotic stress including drought, dust, salinity, heavy metals and UV can occur at one time. For example, high temperature and high photon irradiance often accompany low water supply, which can in turn be exacerbated by subsoil mineral toxicities that constrain root growth. Furthermore, one abiotic stress can decrease a plant's ability to resist a second stress. For example, low water supply can make a plant more susceptible to damage from high irradiance due to the plant's reduced ability to reoxidize NADPH and thus maintain an ability to dissipate energy delivered to the photosynthetic light-harvesting reaction centers (Mark & Bacic, 2005). If a single abiotic stress is to be identified as the most common in limiting the growth of crops worldwide, it most probably be low water supply (Boyer, 1982; Araus et al., 2008). The Arabian peninsula is one of the five major regions where dust originates (Idso, 1976). The Sahara and dry lands around the Arabian peninsula are the main source of airborne dust, with some contributions from Iran, Pakistan and India

into the Arabian Sea, and China's storms deposit dust in the Pacific. Dust affects photosynthesis and transpiration physically when it accumulates on leaf surfaces. Covering and plugging stomata, shading and removing cuticular wax were reported as physical effects of dust (Luis et al., 2008).

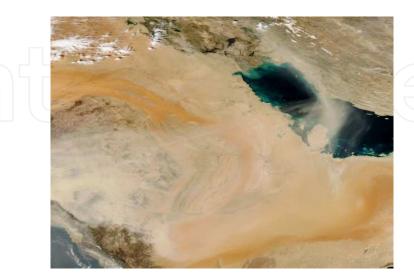


Fig. 3. Dust plumes swept across the Arabian Peninsula in early March 2009. The Moderate Resolution Imaging Spectroradiometer (MODIS) on NASA's Aqua satellite captured this image on March 11, 2009.

In arid environments, decreased water use efficiency because of dust deposition, could therefore contribute substantially to drought stress. The physical effects of dust accumulating on leaf surfaces, on leaf physiology, such as photosynthesis, transpiration, stomatal conductance and leaf temperature of cucumber and kidney bean plants were investigated by Hirano et al., 1995. It was found that dust decreased stomatal conductance in the light, and increased it in the dark by plugging the stomata, when the stomata were open during dusting. When dust of smaller particles was applied, the effect was greater (Hirano et al. 1995). However, the effect was negligible when the stomata were closed during dusting. The dust decreased the photosynthetic rate by shading the leaf surface. The dust of smaller particles had a greater shading effect. Moreover, it was found that the additional absorption of incident radiation by the dust increased the leaf temperature, and consequently changed the photosynthetic rate in accordance with its response curve to leaf temperature. The increase in leaf temperature also increased the transpiration rate (Hirano et al., 1995). Dust may allow the penetration of phytotoxic gaseous pollutants into plant leaves. Visible injury symptoms may occur and generally there is decreased productivity.

Correia et al., 2004 studied the deposition of dust on the foliar surface of the evergreen *Olea europaea* and a semi-deciduous (*Cistus laurifolius*). They found that the affect mainly on the reflectance, it increased with increasing deposition levels, causing a complementary decrease in light absorbance by the leaves of both species. As a consequence, the energy balance of the leaves and net photosynthesis may be altered, thus reducing the productivity of the affected vegetation. However, this effect seems to be more pronounced in *C. laurifolius* compared to *O. europaea*. This could mean that some species maybe more susceptible to dust pollution. In this sense, one could expect an

alteration on the specific composition of the vegetation of the affected areas in response to dust pollution (Correia et al., 2004).

On photosynthesis, however, almost all the previous studies only guessed the physical effects in their discussions. Dust deposition has been found to affect photosynthesis, stomatal functioning and productivity (Luis et al., 2008, Ibrahim & El-Gaely, 2011). Chlorophyll fluorescence, an indication of the fate of excitation energy in the photosynthetic apparatus, has been used as an early, in vivo, indication of many types of plant stress (Maxwell & Johnson, 2000, Ibrahim and Bafeel, 2008). Photoinhibition is evident through the reduction in the quantum yield of photosystem 2 (PSII) and a decrease in variable chlorophyll (Chl) a fluorescence (Demmig-Adams and Adams, 1993; Ibrahim & El-Gaely, 2011).



Fig. 4. Influence of dust deposition and its physical effect on blocking stomata in some plants (A-F) in arid environment (Ibrahim & El-Gaely, 2011).

The decrease of efficiency of PSII photochemistry under stress may reflect not only the inhibition of PSII function, but also an increase in the dissipation of thermal energy (Demmig-Adams& Adams 1993), the latter is often considered as a photo-protective mechanism.



Fig. 5. Visual symptoms of some abiotic stress(including drought, dust accumulation and heavy metal pollution) on some desert plants in arid environment (Ibrahim & El-Gaely, 2011).

#### 3. Spontaneous relationship between abiotic stress and oxidative stress

The reactive oxygen species (ROS) that arise from normal metabolic processes are kept under tight control by various antioxidant mechanisms. ROS are important signal molecules that regulate many physiological processes, including environmental stress responses. Under steady state conditions, the ROS molecules are scavenged by various antioxidative defense mechanisms (Foyer & Noctor, 2005). The equilibrium between the production and the scavenging of ROS may be perturbed by various biotic and abiotic stress factors such as salinity, UV radiation, drought, heavy metals, temperature extremes, nutrient deficiency, air pollution, herbicides and pathogen attacks. The ability to utilize oxygen has provided plants with the benefit of metabolizing fats, proteins and carbohydrates for energy; however, it does not come without cost.

Oxygen is a highly reactive atom that is capable of becoming part of the potentially damaging molecules commonly called "free radicals" which appear to be a major contributor to aging and damage the cell. Fortunately, free radical formation is controlled naturally by various beneficial compounds known as antioxidants that protect cellular membranes and organelles from the damaging effects of active species. Antioxidants are the first line of defense against free radical damage, and are critical for maintaining optimum health and well being of the plant cells. The need for antioxidant becomes even more critical

with increased exposure to free radicals. Each organelle has potential targets for oxidative stress as well as mechanisms for eliminating the noxious oxyradicals. Therefore, plants are equipped with complex antioxidant systems composed of low molecular weight antioxidants non enzymatic compounds, like lipid soluble and membrane-associated tocopherol; ascorbate and glutathione (Foyer 1993), (Foyer & Noctor, 2005) as well as protective antioxidant enzymes such as superoxide dismutase (SOD, EC 1.15.1.1), catalase (CAT, EC 1.11.1.6), peroxidases (APX, EC 1.11.1.1) and glutathione reductase (GR, EC 1.6.4.2). Other components of this system, monodehydroascorbate radical reductase, and glutathione reductase serve to maintain the antioxidants in their reduced functional state (Schwanz et al.,1996) Whether this is the case or not, the antioxidant defenses appear to provide crucial protection against oxidative damage in cellular membranes and organelles in plants grown under unfavorable conditions (Smirnoff 1993 and Kocsy et al.,2000).

Ibrahim & Sameera, 2011 showed that the activity of peroxidise (POD) and CAT of *Lepidium sativum* treated with lead mainly displayed biphasic responses due to increased Pb2+ level. SOD activity under elevated lead stress was steadily stimulated with increasing metal ions level in medium up to 600 ppm. The results showed that, under high metal stress, POD and CAT activities were inhibited, while SOD activity was stimulated, indicating that those enzymes are located at different cellular sites, which had different resistance to heavy metals. Thus, the deterioration of cellular system functions by high metal stress might result in inhibition of enzyme activity (Fig. 5)

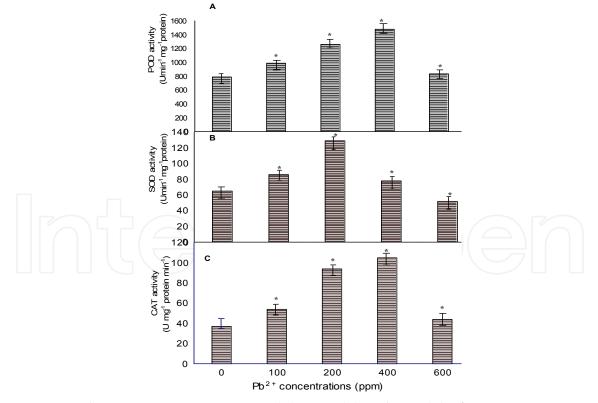


Fig. 6. Antioxidant enzyme activities POD (A), SOD (B) and CAT(C) of *Lepidium sativum* leaves subjected to various concentrations of Pb<sup>2+</sup>. Each value represents the mean ±SE of five replicates. Significant differences (P<0.05) between treatments according to LSD test are shown by an asterisk (Ibrahim & Bafeel, 2011).

Oxygen free radicals or activated oxygen has been implicated in diverse environmental stresses in plants and animals and appears to be a common participation in most, if not all, degenerative conditions in eukaryotic cells. The peroxidation of lipid, the cross-linking and inactivation of proteins and mutations in DNA are typical consequences of free radicals, but because the reactions occur quickly and often are components of complex chain reactions, we usually can only detect their "footprints". Accumulation of ROS as a result of various environmental stresses is a major cause of loss of crop productivity worldwide (Mittler, 2002), (Apel and Hirt, 2004), (Khan & Singh, 2008), (Mahajan & Tuteja, 2005), (Tuteja, 2007; 2010).

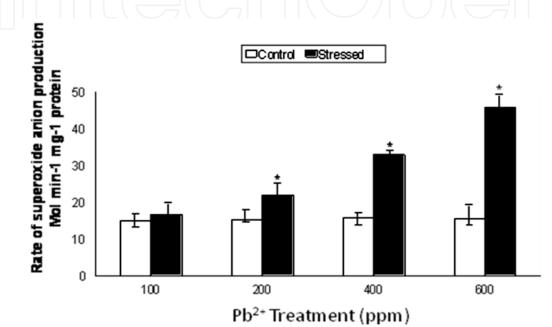


Fig. 7. Changes in the rate of superoxide production rate in roots of untreated and plants of *Lepidium sativum* subjected to various concentrations of Pb<sup>2+</sup> for 10 days (Ibrahim & Bafeel, 2009).

Oxidative stress is a condition in which ROS or free radicals, are generated extra- or intracellular, which can exert their toxic effects to the cells. These species may affect cell membrane properties and cause oxidative damage to nucleic acids, lipids and proteins that may make them non functional. It is well documented that various abiotic stresses lead to the overproduction of ROS in plants which are highly reactive and toxic and ultimately results in oxidative stress. In an environment of molecular oxygen ( $O_2$ ), all living cells are confronted with the reactivity and toxicity of active and partially reduced forms of oxygen: singlet oxygen ( $^{1}O_{2}$ ), superoxide anion ( $O_{2}$ -), hydroxyl radical (HO.), and hydrogen peroxide ( $H_2O_2$ ), which can lead to the complete destruction of cells (Mittler et al., 2004).

These reactive oxygen species (ROS) can show acute production under conditions such as ultraviolet light, environmental stress, or anthropic action through xenobiotics such as herbicides. However, their production is also directly and constantly linked with fundamental metabolic activities in different cell compartments, especially peroxisomes, mitochondria, and chloroplasts. In plants, the links between ROS production and photosynthetic metabolism are particularly important (Rossel et al., 2002).

#### 4. Examples of oxidative stress indices

#### 4.1 Lipid peroxidation

It has been recognized that during lipid peroxidation (LPO), products are formed from polyunsaturated precursors that include small hydrocarbon fragments such as ketones, malondialdehyde (MDA), etc and compounds related to them (Garg & Manchanda, 2009). Some of these compounds react with thiobarbituric acid (TBA) to form colored products called thiobarbituric acid reactive substances (TBARS) (Heath & Packer, 1968). LPO, in both cellular and organelle membranes, takes place when above-threshold ROS levels are reached, thereby not only directly affecting normal cellular functioning, but also aggravating the oxidative stress through production of lipid-derived radicals (Montillet et al., 2005).

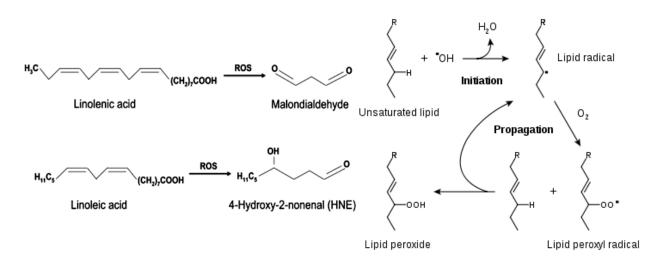


Fig. 8. Production of lipid-derived radicals via lipid peroxidation

#### 4.2 Hydrogen peroxide

Hydrogen peroxide  $(H_2O_2)$  plays a dual role in plants: at low concentrations, it acts as a signal molecule involved in acclimatory signaling triggering tolerance to various biotic and abiotic stresses and, at high concentrations, it leads to programmed cell death (PCD) (Quan et al., 2008).  $H_2O_2$  has also been shown to act as a key regulator in a broad range of physiological processes, such as senescence (Peng et al., 2005), photorespiration and photosynthesis (Noctor & Foyer, 1998), stomatal movement (Bright et al., 2006), cell cycle (Mittler et al., 2004) and growth and development (Foreman et al., 2003).

Also,  $H_2O_2$  is starting to be accepted as a second messenger for signals generated by means of ROS because of its relatively long life and high permeability across membranes (Quan *et al.*, 2008). In an interesting study the response of pre-treated citrus roots with  $H_2O_2$  (10 mM for 8 h) or sodium nitroprusside (SNP; 100 mM for 48 h) was investigated to know the antioxidant defense responses in citrus leaves grown in the absence or presence of 150 mM NaCl for 16d (Tanoua *et al.*, 2009). It was noted that  $H_2O_2$  and SNP increased the activities of leaf antioxidant enzymes such as, superoxide dismutase (SOD), catalase (CAT), ascorbate peroxidase (APX) and glutathione reductase (GR) along with the induction of relatedisoform(s) under non-NaCl-stress conditions.

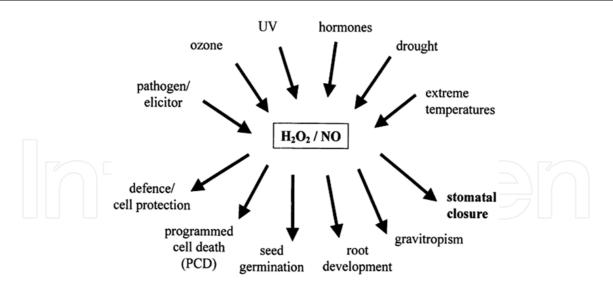


Fig. 9. Involvement of  $H_2O_2$  and NO in cellular responses to various stresses and stimuli(Desikan et al., 2004).

#### 4.3 Protein oxidation

Protein oxidation is defined as covalent modification of a protein induced by ROS or byproducts of oxidative stress. Most types of protein oxidations are essentially irreversible, whereas, a few involving sulfur-containing amino acids are reversible (Ghezzi &Bonetto, 2003). Protein carbonylation is widely used marker of protein oxidation (Moller et al., 2007) and (Job et al., 2005). The oxidation of a number of protein amino acids particularly Arg, His, Lys, Pro, Thr and Trp give free carbonyl groups which may inhibit or alter their activities and increase susceptibility towards proteolytic attack (Moller et al., 2007). Protein carbonylation may occur due to direct oxidation of amino acid side chains (e.g. proline and arginine to  $\gamma$ -glutamyl semialdehyde, lysine to amino adipic semialdehyde, and threonine to aminoketobutyrate) (Shringarpure& Davies, 2002).

## 5. Physiological, biochemical and molecular responses of plant to abiotic stresses

#### 5.1 Photosynthetic responses toward oxidative stress

In higher plants, photosynthesis takes place in chloroplasts, which contain a highly organized thylakoid membrane system that harbours all components of the light-capturing photosynthetic apparatus and provides all structural properties for optimal light harvesting. Oxygen generated in the chloroplasts during photosynthesis can accept electrons passing through the photosystems, thus forming  $O_2^-$ . Through a variety of reactions,  $O_2^-$  leads to the formation of  $H_2O_2$ , OH and other ROS. The ROS comprising  $O_2^-$ ,  $H_2O_2$ ,  $IO_2$ ,  $HO_2^-$ , OH, ROOH, ROO, and RO are highly reactive and toxic and causes damage to proteins, lipids, carbohydrates, DNA which ultimately results in cell death (Bryan , 1996; Downs et al., 1999). In chloroplast activated oxygen species can be generated by direct transfer of excitation energy from chlorophyll to produce singlet oxygen, or by univalent oxygen reduction at PSI, in the Mehler reaction (Asada et al.,1998). The latter process results in the formation of the superoxide anion radical (O<sub>2</sub>.-), singlet oxygen (<sup>1</sup>O<sub>2</sub>) and eventually H<sub>2</sub>O<sub>2</sub> and the highly toxic hydroxyl radical (OH). It is well known that Cu<sup>2+</sup> catalyze the formation of OH. from the non-enzymatic chemical reaction between superoxide and H<sub>2</sub>O<sub>2</sub>.

Thylakoids are considered to be one of the major sites of superoxide production because of the simultaneous presence in chloroplasts of a high oxygen level and an electron transport system. Most of the superoxide is produced by photosystem I via the univalent reduction of oxygen through the ferredoxin / ferredoxin NADP+ oxidoreductase system (Mehler reaction). The use of DCMU, the known inhibitor of photosynthetic electron transport, and the use of the new spin trap DEPMPO have demonstrated that photosystem II also contributes to superoxide production (Navari-Izzo et al.,1998).

The modifications of the chloroplast in response to various environmental stresses have been widely studied in different laboratories and, thus the literature in the area is vast. The stress is sensed at the levels of pigment composition, structural organization, primary photochemistry and the  $CO_2$  fixation(Biswal et al., 2003; Biswal, 2005).

Spatial and temporal complexity of photosynthesis makes photostasis prone to stress. The sequence of photosynthesis is known to cover a wide time-span and begins with photophysical and photochemical events, i.e. light absorption, excitation energy transfer and charge separation in the timescale of femtoseconds (10-15 s) to nanoseconds (10-9 s). This is followed by electron transport in the microseconds (10-6 s) to milliseconds (10-3 s) range, and finally by enzyme mediated reactions in the milliseconds to seconds range. Relatively slow reactions are rate-limiting and thus, incompatible with the fast reactions. Further, the fast primary photochemical reactions are relatively stress-resistant compared to temperaturedependent, slow, enzyme-mediated reactions associated with the electron transport system and carbon dioxide fixation in the Calvin–Benson cycle (Krause & Jahns, 2004). This results in the development of excitation pressure at the source. Since plants are photoautotrophs, light at any intensity in combination with other environmental stresses can bring a change in photostasis in terms of accumulation of excess unutilized quanta because of weakened sink demand induced by stress. In addition, high light always accumulates excess energy at the 'source'. NPQ of excess quanta at the source is one of the major processes for restoration of the balance and maintenance of photostasis(Biswal et al., 2011).

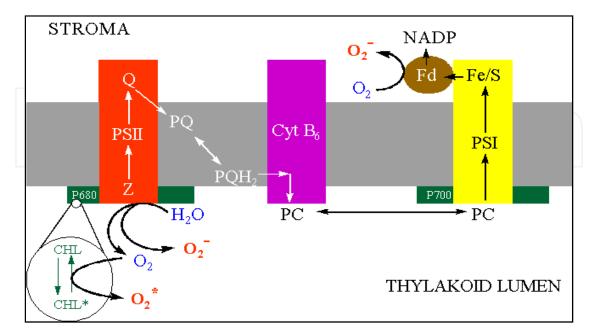


Fig. 10. Electron transport system in the thylakoid membrane showing three possible sites of activated oxygen production (Elstner, 1991; Bryan, 1996).

#### 5.2 Plant responses toward temperature divergence

The climatic pattern in the arid zones is frequently characterized by a relatively "cool" dry season, followed by a relatively "hot" dry season, and ultimately by a "moderate" rainy season. In general, there are significant diurnal temperature fluctuations within these seasons. Quite often, during the "cool" dry season, daytime temperatures peak between 35 and 45 centigrade and fall to 10 to 15 centigrade at night. Daytime temperatures can approach 45 centigrade during the "hot" dry season and drop to 15 centigrade during the night. During the rainy season, temperatures can range from 35 centigrade in the daytime to 20 centigrade at night. In many situations, these diurnal temperature fluctuations restrict the growth of plant species.



Fig. 11. Different kinds of desert plants in arid environment (Ibrahim 2011).

Arid region plants are adapted to cope with temperature divergence between the prolonged annual hot and dry period in summer and the cooled winter. Plants evolved different survival mechanism including activation of antioxidant system, up-regulation of early lightinduced proteins (ELIPs), and xanthophyll-cycle-dependent heat energy dissipation, among others (Demmig-Adams and Adams, 1993; Verhoeven et al., 2005). Increases in temperature raise the rate of many physiological processes such as photosynthesis in plants, to an upper limit. Extreme temperatures can be harmful when beyond the physiological limits of a plant. Decreasing photosynthesis seems to be the major cause of the chill induced reduction in the growth of plant in temperate climates (Baker et al., 1994). Several indicators support this assumption: periods of low temperature were accompanied by a lower chlorophyll content (Leipner et al., 1999; Fryer et al., 1998), an increased pool size of xanthophyll cycle pigments, reduced photosynthetic capacity (Baker et al., 1994; Fryer et al., 1998).

Leaf antioxidant systems can prevent or alleviate the damage caused by reactive oxygen species (ROS) under stress conditions, and include enzymes such as superoxide dismutase (SOD), catalase (CAT), peroxidase (POD), ascorbate peroxidase (APX), and metabolites including ascorbate acid (AsA) and glutathione (GSH) (Asada, 1999; Xu *et al.*, 2008). Phenolics are ubiquitous secondary metabolites in plants including large group of biologically active components, from simple phenol molecules to polymeric structures with molecular mass above 30 kDa (Dreosti, 2000, Ibrahim *et al.*, 2011).

*Artemisia monosperma* showed the lowest activities for Guaiacol peroxidase(GuPx) and polyphenol oxidase (PPO) at 38°C and at 47°C in comparison with activities on plants collected at 9 and 15 °C (Table 1). Moreover, the relationship between GuPx and PPO activities and soluble phenolics concentration in A. monosperma plants appear to indicate that 47°C and 9°C caused heat and cold stress, by subjecting the plants to a super-optimal and suboptimal temperatures respectively(Ibrahim et al., 2011).

The metabolism of phenolic compounds includes the action of oxidative enzymes such as GuPx and PPO, which catalyze the oxidation of phenols to quinones (Thypyapong et al., 1995; Vaughn and Duke, 1984). Some studies have reported that these enzyme activities increase in response to different types of stress, both biotic and abiotic (Ruiz et al., 1998, 1999). More specifically, both enzymes have been related to the appearance of physiological injuries caused by thermal stress (Grace et al., 1998).

Phenylalanine ammonia-lyase (PAL) is considered to be the principal enzyme of the phenylpropanoid pathway (Kacperska, 1993) catalyzing the transformation, by deamination, of L-Phenyalanine into *trans*-cinnamic acid, which is the prime intermediary in the biosynthesis of phenolics (Levine et al., 1994). This enzyme increases in activity in response to thermal stress and is considered by most authors to be one of the main lines of cell acclimation against stress in plants (Leyva et al., 1995). Phenols are oxidized by peroxidase (POD) and primarily by polyphenol oxidase (PPO), this latter enzyme catalyzing the oxidation of the *o*-diphenols to *o*-diquinones, as well as hydroxylation of monophenols(Thypyapong et al., 1995). These activities of enzymes increase in response to different types of stress, both biotic and abiotic (Ruiz et al., 1998, 1999). More specifically, both enzymes have been related to the appearance of physiological injuries caused in plants by different stress (Grace et al., 1998; Ruiz et al., 1998; Ibrahim et al., 2011).

| Sampling<br>date | Temp. | U mg protein min <sup>-1</sup> |           |            | PPO           | POD                      |
|------------------|-------|--------------------------------|-----------|------------|---------------|--------------------------|
|                  |       | SOD                            | АРХ       | CAT        | µmol caffeic  | µmol guaiacol            |
|                  |       |                                |           |            | acid mg-1     | mg <sup>-1</sup> protein |
|                  |       |                                |           |            | protein min-1 | min <sup>-1</sup>        |
| 15 June          | 38°C  | 14.7±1.12                      | 4.32±0.66 | 3.27±0.06  | 11.4±0.83     | 12.7±0.99                |
| 15 Aug           | 47°C  | 16.6±1.22                      | 5.31±0.71 | 4.87±0.07  | 22.3±2.30     | 16.9±1.23                |
| 15 Dec           | 9°C   | 22.9±2.08                      | 20.8±2.14 | 18.74±1.32 | 38.8±3.86     | 37.3±2.98                |
| 15 Feb           | 15°C  | 19.0±2.03                      | 17.5±1.65 | 13.58±0.98 | 28.2±2.19     | 25.3±2.07                |

Table 1. Variation of antioxidant enzymes activities( superoxide dismutase, SOD; ascorbate peroxidase, APX; catalase, CAT; phenol peroxidase, PPO and guaiacol peroxidase, POD in *Artemisia monosperma* plant in response to temperature divergence in Riyadh (Saudi Arabia)( Ibrahim et al., 2011).

Over-expression of ROS scavenging enzymes like isoforms of SOD (Mn-SOD, Cu/Zn-SOD, Fe-SOD), CAT, APX, GR, DHAR, GST and GPX resulted in abiotic stress tolerance in various plants due to efficient ROS scavenging capacity. Pyramiding of ROS scavenging enzymes may also be used to obtain abiotic stress tolerance plants. Therefore, plants with the ability to scavenge and/or control the level of cellular ROS may be useful in future to withstand harsh environmental conditions.

#### 5.3 Osmotic adjustment in stressed plants

Osmotic response and their adjustment was considered as a biochemical marker in plants subjected to abiotic stress such as salinity can occur by the accumulation of high concentrations of either inorganic ions or low molecular weight organic solutes. Although both of these play a crucial role in higher plants grown under saline conditions, their relative contribution varies among species, among cultivars and even between different compartments within the same plant (Greenway & Munns, 1980). The compatible osmolytes generally found in higher plants are low molecular weight sugars, organic acids, polyols, and nitrogen containing compounds such as amino acids, amides, imino acids, ectoine (1,4,5,6-tetrahydro-2-methyl-4-carboxylpyrimidine), proteins and quaternary ammonium compounds. According to Murakeozy et al.(2003), of the various organic osmotica, sugars contribute up to 50% of the total osmotic potential in glycophytes subject to saline conditions. The accumulation of soluble carbohydrates in plants has been widely reported as a response to salinity or drought, despite a significant decrease in net  $CO_2$  assimilation rate (Carm, 1976; Popp & Smirnoff, 1995).

#### 5.4 Role of amino acids and amides on the avoidance of abiotic stress

Amino acids have been reported to accumulate in higher plants under salinity stress (Bielski, 1983; Moller, 2001; Mahajan and Tuteja, 2005). The important amino acids include alanine, arginine, glycine, serine, leucine, and valine, together with the imino acid, proline, and the non-protein amino acids, citrulline and ornithine (Mahajan and Tuteja, 2005, Hu, 2007) .Proline, which occurs widely in higher plants, accumulates in larger amounts than other amino acids in salt stressed plants (Bielski et al., 1983; McDowell and Dangl, 2000; Navrot et al., 2007; Pastore et al., 2002; Reumann et al., 2004). Proline accumulation is one of the common characteristics in many monocotyledons under saline conditions (Dybing et al., 1978; Grant and Loake, 2000), although in barley seedlings, NaCl stress did not affect proline accumulation (Bolwell & Woftastek, 1997). However, proline accumulation occurs in response to water deficit as well as to salt. Thus, synthesis of proline is a non-specific response to low growth medium water potential (Navrot et al., 2007). Proline regulates the accumulation of useable N, is osmotically very active (Bielski et al., 1983; Moller, 2001), contributes to membrane stability (Heath, & Packer, 1968; Garg and Manchanda, 2009; Montillet et al., 2005) and mitigates the effect of NaCl on cell membrane disruption (Fam and Morrow, 2003). Even at supra-optimal levels, proline does not suppress enzyme activity (Hayashi and Nishimura, 2003; Moller et al., 2007).

#### 6. Conclusion

According to our investigations, Ibrahim & Bafeel, 2008 concluded that dark chilling imposes metabolic limitation on photosynthesis and ROS are involved, to some degree, in

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the limiting photosynthetic capacity of alfalfa leaves. After recovery period the alfalfa plants showed physiological and biochemical changes that contribute to its superior dark chilling resistance and prevent the leaves from undergoing photooxidation damage and eventual death. Also our results showed that high cellular levels of  $H_2O_2$  accumulated during the dark chilling treatment can induce the activation of a defense mechanism against chilling stress or programmed cell death. The accumulation of  $H_2O_2$  can be induced by the increase in SOD activity. Therefore, during the recovery treatment the accumulated  $H_2O_2$ , in turn, may activate a protective mechanisms that increase the activities of several antioxidant enzymes such as APX, CAT and GR .Also induce alterations in the relative concentration of several non-enzymatic antioxidant compounds such as phenolics and tocopherols. (Bafeel & Ibrahim, 2008).

Results reported by Ibrahim & Alaraidh, 2010 demonstrated that changes in gene expression do occur in the two cultivars of Triticum aestivum in response to drought, and these differentially expressed genes, though functionally not known yet, may play important roles for cultivars to exhibit its response to drought stress before and after rehydration. Moreover, Ibrahim & Bafeel, 2009 concluded that prolonged stress induced by Pb<sup>2+</sup> concentrations, can result into the activation of antioxidative enzymes and also enhance the gene expression of these antioxidant enzymes.

Although oxidative stress is potentially a lethal situation, it is also clear that plant systems exploit the interaction with oxygen. The production and destruction of active oxygen species is intimately involved with processes such as the hypersensitive responses and the regulation of photosynthetic electron flow. There are numerous sites of oxygen activation in the plant cell, which are highly controlled and tightly coupled to prevent release of intermediate products. Under stress situations, it is likely that this control or coupling breaks down and the process "dysfunctions" leaking activated oxygen. This is probably a common occurrence in plants especially when we consider that a plant has minimal mobility and control of its environment. Activated forms of oxygen are important in the biosynthesis of "complex" organic molecules, in the polymerization of cell wall constituents, in the detoxification of xenobiotic chemicals and in the defense against pathogens. Thus, the plant's dilemma is not how to eliminate the activation of oxygen, but how to control and manage the potential reactions of activated oxygen.

Genetic engineering also offer advantages in terms of the study of the physiological roles of enzymes where a classical genetic approach, such as selection of enzyme-deficient mutants, is difficult or almost impossible to carry out. In plant systems, the situation is often considerably complicated by the presence of a large number of isoenzyme forms, for example, the large GR and SOD families of isoenzymes, encoded by different genes. In the future, however, the use of antisense technology combined with selection of specific cDNA clones for isoenzymes may facilitate investigation of such enzyme-deficient mutants. Current observations suggest that increasing the level of stress tolerance by reinforcing the plant's defense system with new genes is an attainable goal.

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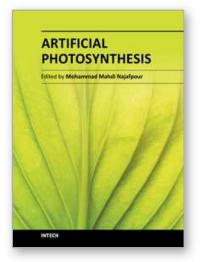
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Photosynthesis is one of the most important reactions on Earth, and it is a scientific field that is intrinsically interdisciplinary, with many research groups examining it. We could learn many strategies from photosynthesis and can apply these strategies in artificial photosynthesis. Artificial photosynthesis is a research field that attempts to replicate the natural process of photosynthesis. The goal of artificial photosynthesis is to use the energy of the sun to make different useful material or high-energy chemicals for energy production. This book is aimed at providing fundamental and applied aspects of artificial photosynthesis. In each section, important topics in the subject are discussed and reviewed by experts.

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