ISSN : 1812-5379 (Print)
ISSN : 1812-5417 (Online)
http://ansijournals.com/ja

JOURNAL OF AGRONOMY

ANSInet
Asian Network for Scientific Information
308 Lasani Town, Sargodha Road, Faisalabad - Pakistan

Journal of Agronomy 9 (2): 29-37, 2010
ISSN 1812-5379

Biochemical and Mineral Responses of Okra Seeds (*Abelmoschus esculentus* L. Variety Marsaouia) to Salt and Thermal Stresses

B.D. Besma and D. Mounir
Higher Institute of Agronomy, Chott Marièm, 4042 Sousse, Tunisia

Abstract: The present research has studied the effects of NaCl and temperature on germination and emergence of okra. Studies were carried out with seeds of a local okra variety (Marsaouia) subjected to 0 and 100 mM NaCl, performed in the dark at 10, 15, 25, 35 and 40°C at germination stage and by 12 h light at emergence stage. The cumulative germination percentage, the cumulative emergence percentage, starch content, the reducing sugars levels, total amylase activity, sodium and potassium accumulation were quantified in germinated seeds at 15°C, 25 and 35°C. Temperature presented a significant effect on salt sensitivity of this species at germination and emergence stages. Germination of okra seeds was completely inhibited at 10 and 40°C. The best germination and emergence temperature was recorded at 25°C. The adverse effect of salt was more pronounced at low and high temperature. During the salt stress treatment, the level of starch reserves was higher at 25°C and lower at 15 and 35°C, an increase in reducing sugars content in cotyledons was observed. The activity of total amylase was most intensive at lower temperature in control seeds and at higher temperature in salt treated seeds. The sodium concentration on germinated seeds increased significantly at 15 and 35°C, but potassium amount did not change regularly within thermal and salt stress interaction.

Key words: Starch, reducing sugars, amylase activity, sodium, potassium

INTRODUCTION

Successful establishment of plants was largely depends on successful germination (Tlig *et al*., 2008), since germination represents a critical event in plant's life cycle and its timing largely predetermines the chances of survival of a seedling up to maturity (Chauhan *et al*., 2009). Zadeh and Naeini (2007) suggested that one of the most sensitive phases of a plant's life to salinity is that of seed germination. In this context, Bohnert *et al*. (1995) mentioned that the metabolism of the osmoprotectants and correlating enzymes in the germinated seeds can be affected by a number of environmental factors such as irradiance, temperature, salinity and the type of present ion. Salinity is one of the serious environmental problem that cause osmotic stress and reduction in plant growth and crop productivity in irrigated areas of arid and semiarid regions (Çiçek and Çakirlar, 2002). Salts can cause reduction in water availability, sodium ions accumulation, mineral imbalances that affect seed germination and plant metabolism (Ali *et al*., 2001). Moreover, toxic effects of salts may change enzymatic activity and hormonal balance of plants (Maghsoudi and Maghsoudi, 2008). Indeed, the ecological, physiological and biochemical aspects of the germination process was also influenced by temperature. Principally, temperature may change a number of processes controlling seed germinability, including membrane permeability, the activity of membrane-bound and cytosolic enzymes (Bewley and Black, 1994; Gul and Weber, 1999). Temperature affect the final percentage, velocity of the process, water absorption and biochemical reactions. Early studies on the effect of temperature on different plant species showed that there was a broad optimum temperature for the growth of seedlings (Brar *et al*., 1991; Hucl, 1993; Lafond and Fowler, 1989). It was reported that temperature is one of the crucial factors in modulating seed germination responses under saline conditions (Khan, 1999). In fact, temperature and salinity can interact in determining salinity tolerance during germination and early seedling growth. Mosjidis and Zhang (1995) found that the optimum temperature range for germination of several Vicia species was lower than that for root growth. Although higher salinity decreased germination, the detrimental effect of salinity is generally less severe at optimum temperature (El-Keblawy and Al-Rawai, 2005). In Tunisia, regions are frequently irrigated with salt water; consequently about 10% of the whole territory and 20% of the cultivated lands are saline (Ben Ahmed *et al*., 2008).

Corresponding Author: Ben Dkhil Besma, Higher Institute of Agronomy, Chott Marièm, 4042 Sousse, Tunisia
Tel: +21697594631

Okra (*Abelmoschus esculentus* L.) Moench. Family of Malvaceae) that is widely grown in all regions of the world with a tropical or Mediterranean climate for its immature pods (Doymaz, 2005), it was a marginal crop in Tunisia. The productivity of this crop is limited because of the ignorance for its suitability with regards to soil and climate. Okra is classified as semi-tolerant vegetable crop (Maas and Holfman, 1977), water salinity retarded the growth, yield and physiological growth parameters of okra (Abid *et al*., 2002). Further, okra is sensitive to low temperatures and develop poorly below 15°C (Marsh, 1992), delayed and erratic emergence is a serious problem in okra (Anderson *et al*., 1960) that creates problems with fertilizer utilization, post emergence weed control and uniform harvesting. Temperature, salinity and the hard seed coat of okra can interfere with water up-take constraint germination, establishment and performance of seedlings.

As far as this study was investigated to: (1) study the response of okra (variety Marsaouia) to a combination of temperature and salt stress at germination and emergence seedling stages, (2) to detect the effect of temperature and salinity interaction on the storage reserve mobilization along with the hydrolytic enzyme and (3) to determine the ionic differences under salt and thermal stress.

MATERIALS AND METHODS

Plant material: The experiment included a local variety of okra: Marsaouia. Seeds have been provided usually by the Baddar Company (Tunisia). Several germination experiments were conducted in the Laboratory of Agronomy of the High Institute of Agronomy during two years (2005 and 2006) from January to June.

Germination conditions: Okra seeds were first surface sterilized by immersion in 70% ethanol for 1 min, followed by 15 min in 15% sodium hypochlorite and then rinsed with sterile distilled water. Seeds were transferred to sterile Petri dishes containing two sheets of Whatman No.1 filter paper moistened with distilled water or NaCl solution (100 mM). Each Petri dish containing 25 seeds and each treatment was replicated eight times. The Petri dishes were placed in an incubator at the appropriate temperature (10, 15, 25, 35 and 40°C) in the dark. The treatments were placed in a factorial arrangement in a completely randomized design.

Seeds were considered germinated when the radicle reached 2 mm, cumulative germination percentage was determined. The germinated seeds were counted every 2 days during the 20 days-experimental period.

Germinated seeds at each treatment were sampled after 48 h from the beginning of incubation, cotyledons were separated and then placed at -20°C to stop the germination process. These samples were subject to biochemical and mineral analysis.

Emergence conditions: Only germinated seeds at 15, 25 and 35°C in distilled water and in salt solution were transferred in a plastic disposable containers filed with peat/perlite mixture (2:1 v/v) to emerge. Containers where incubated in a programmed refrigerated incubator on 12 h light: 12 h dark at 15, 25 and 35°C. The containers were irrigated continuously with distilled water or salt solution. Treatments were replicated 4 times in a factorial experiment laid out in a completely randomized design. Emergence percentages were made daily for 9 days after sowing.

Starch determination: Starch content was determined according to Allefrey and Northcote (1977) as cited by Bewley *et al*. (1993). Batches of 12 cotyledon pairs were homogenized in an ice-cold mortar and pestle in a volume of 16 mL 80% (v/v) ethanol. The homogenates were centrifuged (30000x g, 10 min at 2°C) and then perchloric acid ($HClO_4$; 6 mL, 30%, v/v) was added to solubilize starch from the pellet. The slurry was left at room temperature for 6h, starch was detected with I_2-KI reagent prepared by diluting 0.1 mL stock solution (0.06 g I_2 and 0.60 g KI in 10 mL deionized water) with 0.05 M HCl just prior to the assay. Samples of 0.5 mL starch solution were mixed with 0.5 mL I_2-KI reagent, 1 mL 30% (v/v) perchloric acid and then were vortexed and left standing at room temperature. The absorbance (620 nm) of the samples was compared to that of the standard curve of 0 to 5 mg mL^{-1} which was obtained using soluble starch dissolved in 30% $HClO_4$ and detected with the same I_2-KI reagent. The assay was conducted in triplicate for each sample.

Amylase activity: Total amylase preparations were made according to method of Liu *et al*. (2005) by grinding cotyledons of germinated seeds in a mortar and pestle with 0.05 M acetate buffer (pH 6.0). All operations were performed at 4°C. The resulting homogenate was strained through cheesecloth and then centrifuged (18000*g, 10 min) and filtered. The substrate for amylase is a 1% solution of soluble starch in 0.1 M acetate buffer (pH 5.6). The enzyme prepared to be tested was diluted to 1 mL with water and then 1.0 mL of the substrate solution was added. After 1 h incubation at 25°C, 2 mL of a 3,5-dinitrosalicylic acid was added and the tubes were placed in a boiling water bath for 5 min (Swain and Dekker, 1966) and then cooled to room temperature and diluted with 20 mL of water. The absorbance values of the resulting

colored solutions were determined by a spectrophotometer (Model camspec M330 UV/Vis). Reducing sugars content was determined too using maltose as standard.

Sodium and potassium determination: For the analysis of sodium and potassium in seeds after germination, 100 mg of dried material was placed in test tubes and digested in nitric acid 0.1 N. Sodium (Na^+) and potassium (K^+) concentrations determined using flame photometer (Eppendorf).

Statistical analysis: The data were subject to the statistical analyses. Analyse of variance was used (ANOVA) with salinity and temperature treatments as factors and their interactions was performed for the whole data set SPSS for Windows: version 13. *$p \leq 0.05$ was used to define statistical significance. If a significant difference was determined among means, a Duncan test was used for comparison of means among different temperatures within a factor.

RESULTS

Salinity and temperature effects on seed germination: The change in temperature regimes significantly affected seeds germination, indeed seeds were able to germinate at temperatures between 15 and 35°C, the optimal temperature corresponds to 25°C. Germination was completely inhibited at 10 and 40°C.

Improved germination of seeds was observed in non saline solutions at all temperature regimes tested and reached final germination percentages in less than 2 days mainly at 35°C.

Seed germination was significantly lower under 15°C, in the control (0 mM NaCl) germination commenced on day 6 and had about 41.5% as final germination percentage. At 100 mM NaCl, germination started on day 18, the sodium chloride treatment decreased clearly germination percentage which was about 4% at the end of the thermoperiod.

The optimal incubation temperature degree (25°C) recorded the highest seed germination percentage in the control (100%). While at higher salinity, moderate temperature treatment had better germination percentages. The decrease in seed germination percentage under this temperature was about 19.5% and it was less severe than that observed at 15°C.

Raising temperature degree was associated with a significant decrease in germination percentage. At 35°C germination started and completed on 2 days, reaching a percentage of 84.5%. At 100 mM NaCl, germination percentage stabilized after 6 days of incubation at 6% (Fig. 1a-c).

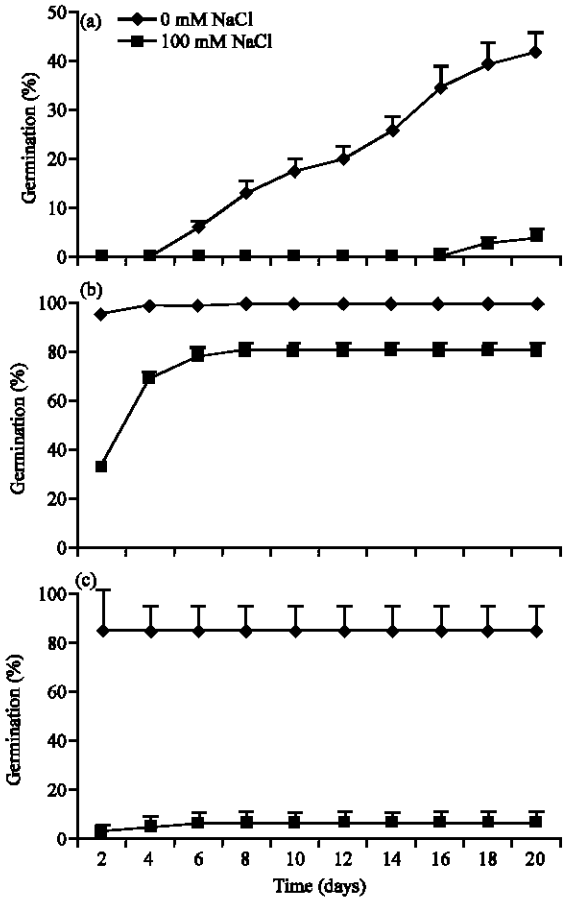

Fig. 1: Cumulative germination percentage of *Abelmoschus esculentus* seeds during 20 days as influenced by NaCl concentrations (0,100 mM) and incubation temperatures (15, 25 and 35°C). At temperature 10 and 40°C seeds germination was completely inhibited. Values are means of 8 replicates. Bars on data points are ±SE of the mean (not shown when smaller than the symbol). (a) 15°C, (b) 25°C and (c) 35°C

Salinity and temperature effects on seedling emergence: Sensibility of okra to salinity and temperature at emergence stage was also tested. At lower temperature (15°C), seedling emerged latterly after five days even those irrigated with distilled water or with saline solution. In such assertion, salinity reduced considerably final seedling emergence until 12.5%, as compared with the control seedling emergence was around 40%.

Incubation temperature of 25°C was suitable for emergence of *A. esculentus*, in the control seedling emergence initiated on day 2 with 47.5% and reached 100% on day 4. As compared with the control, salinity reduced seedling emergence until 57.5%.

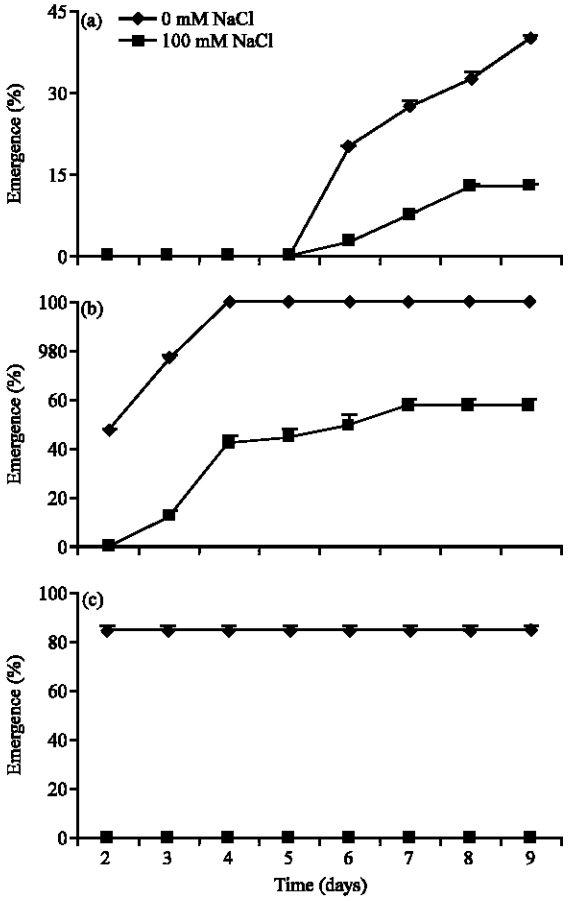

Fig. 2: Cumulative emergence percentage of *Abelmoschus esculentus* seeds during 9 days as influenced by NaCl concentrations (0,100 mM) and incubation temperatures (15, 25 and 35°C). Values are means of 4 replicates. Bars on data points are ±SE of the mean (not shown when smaller than the symbol). (a) 15°C, (b) 25°C and (c) 35°C

At higher incubation temperature (35°C) seedling emergence started on day 2, the final emergence percentage was around 85%. Emergence was inhibited at 35°C under high salinity level (100 mM NaCl) (Fig. 2a-c).

Starch content: Changes observed in starch content in the cotyledons of germinated seeds were related both to temperature and salt concentration in the germination medium. As seen in Table 1, considerable differences were observed among temperature, the starch content was lower at 15 and 35°C (respectively 0.06 and 0.03 mg g^{-1} FW). Whereas, at 25°C starch accumulation reached its highest value (0.18 mg g^{-1} FW), okra seeds stored starch as energy source that will be useful for the maintenance

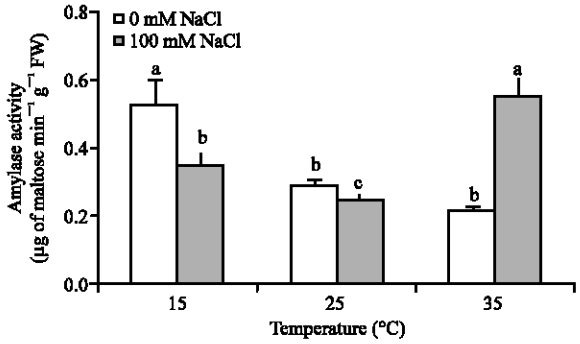

Fig. 3: The effect of temperature on total amylase activity in cotyledons of germinated seeds treated with 0 and 100 mM NaCl. Enzyme activity was expressed as µg of maltose formed min^{-1} g^{-1} FW. Means with the same letter are not significantly different at $p \leq 0.05$ according to Duncan test

Table 1: Starch content (mg g^{-1} FW) in cotyledons of germinated seeds soaked in distilled water and salt solution at 15, 25 and 35°C

	Temperature (°C)		
Treatment (mM)	15	25	35
0	0.06±0.006^{b}	0.18±0.006^{c}	0.03±0.0002^{a}
100	0.03±0.003^{a}	0.11±0.03^{b}	0.02±0.000^{a}

Values in each line followed by the same letter indicate no significant differences ($p \leq 0.05$) according to Duncan test

of developed organs. Nevertheless, unsuitable temperature accelerated the degradation of a major amount of starch reserve at germination stage.

As compared with control, imposition of NaCl resulted in a significant decrease in starch content, this decrease was more pronounced at 25°C reaching 0.11 mg g^{-1} FW. Under unfavourable temperature salinity reduced slightly the level of starch on cotyledons, the decrease rate of starch accumulation was higher at 15°C (50%) than at 35°C 34%).

Amylase activity: The total amylolytic activity of germinated seeds in 0 and 100 mM NaCl at 15°C, 25°C and 35°C is presented in Fig. 3. In the control medium (0 mM NaCl), the average total amylolytic activity was more intense at 15°C, it decreased steadily in the cotyledons after radicle protrusion at 25 and 35°C. These results suggested an increase on hydrolytic enzyme under lower temperature at germination stage; it would limit the amount of energy available to seedling during heterotrophic growth. While, salt stress caused a marked change on amylase activity in response to germination temperature, the stimulatory effect of NaCl was more pronounced at higher temperature (35°C). Under 100 mM NaCl, total amylase activity decreased slightly at 25°C and considerably at 15°C.

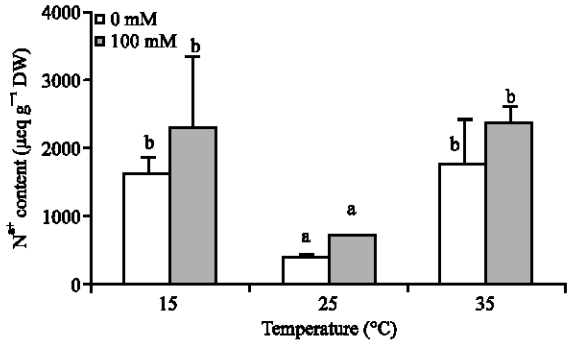

Fig. 4: The effect of increasing temperature on sodium accumulation of okra seeds soaked in distilled water and salt solution. Means with the same letter are not significantly different at $p \leq 0.05$ according to Duncan test

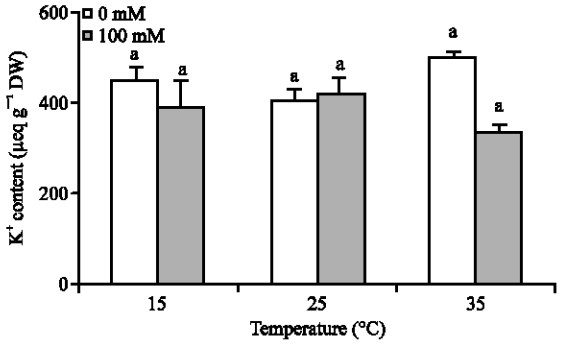

Fig. 5: Potassium concentration of okra seeds soaked in distilled water and salt solution at 15, 25 and 35°C. Means with the same letter are not significantly different at $p \leq 0.05$ according to Duncan test

Table 2: Variation of reducing sugars (mg maltose g^{-1} FW) amounts in cotyledons of germinated seeds at 15, 25 and 35°C submitted to 0 and 100 mM NaCl

	Temperature (°C)		
Treatment (mM)	15	25	35
0	2.6[a]	1.5[a]	2.8[a]
100	6.4[a]	4.8[a]	5.5[a]

Values in each line followed by the same letter indicate no significant differences ($p \leq 0.05$) according to Duncan test

Reducing sugars: As seen in Table 2, the reducing sugars content showed net variation among salt treatment. However, reducing sugars accumulation remained statistically constant through temperature treatments; it increased to a smaller extent over stressed seeds that fluctuated between 1.5 and 2.8 mg maltose g^{-1} FW in the control and between 4.8 and 6.4 mg maltose g^{-1} FW at 100 mM NaCl. Whereas, as compared with control, reducing sugars accumulation raised significantly under NaCl treatment. The lower sugar level was observed at 25°C in cotyledons of salt stressed seeds. At al circumstances, the more pronounced increase of sugars concentration particularly, at high salinity was observed at 15°C.

Sodium and potassium uptake: Analysis of variance revealed that significant differences among the temperature treatment for sodium uptake (Fig. 4). Present results also indicated that sodium amount was affected by salinity treatment. Indeed, sodium amount recorded its minimal value with the intermediate incubation temperature (25°C). Meanwhile, under the highest and lowest incubation temperature degree (15 and 35°C) seeds had higher Na^{+} concentration. However, amount of Na^{+} increased markedly in response to salinity treatment. This increase was more significant at 15 and 35°C. For example, in seeds treated with 100 mM NaCl, the sodium concentration was around 2323 and 2371 µeq g^{-1} DM, respectively at 15 and 35°C. For this treatment and at 25°C, sodium concentration was around 706 µeq g^{-1} DM.

In addition, potassium concentration was affected by temperature and salinity interaction (Fig. 5). In fact, temperature affected the potassium concentration in germinated seeds treated with 100 mM NaCl. In such assertion potassium concentration decreased markedly at 15 and 35°C, whereas, at 25°C potassium concentration increased slightly reaching the highest value which was almost 420.6 µeq g^{-1} DM. The greater accumulation of potassium was showed principally at 35°C with 0 mM NaCl, the lower accumulation was localised at the same temperature with 100 mM NaCl. Dealing with all treatments potassium content didn't change regularly.

DISCUSSION

In the present study, germination and seedling emergence of okra (variety Marsaouia) was affected by salinity and temperature. Thus, the effect of temperature on germination appeared to be dependent on the water supply from the medium, the inhibitory effect of high salinity was greater at 15 and 35°C than at 25°C. Germination declined with decreasing osmotic potential of the germination medium, the magnitude of this decline was higher at 15°C. Khan *et al.* (2001) believed that the adverse effect of high salinity is further aggravated by either an increase or decrease in temperature.

No germination occurred at 10 and 40°C, Nykiforuk and Flanagan (1999) reported that under low temperature a proportion of canola seed (about 25 %) was unable to make the transition from phase II to phase III preventing their germination. Additionally, the exposure of seeds during germination to high temperature resulted in malfunctioning of the enzymatic system. This situation would lead to limitation in much physiological process

vital for seed germination (Al-Thabet *et al*., 2004). Besides, Kursat and Kabar (2007) mentionnated that high temperature both delayed and inhibited germinations of barley and radish seeds. Okra seeds germinated better in the intermediate incubation temperature (25°C), this temperature was optimum for the final germination percentage. Results reported by Al-Thabet *et al.* (2004) concerned the effect of incubation temperature on the germination of canola seeds illustrated that the lowest incubation temperature degree (15°C) was the best which recorded the highest seed germination percentage, while the highest incubation temperature degree (35°C) recorded the lowest germination percentage.

Higher germination percentage at control treatment (0 mM NaCl) could have been due to the absence of salts in the medium and therefore seeds were able to imbibe water. Our results revealed also that at germination stage okra seems to be more tolerant to high salinity (100 mM NaCl) under the favourable temperature (25°C), where germination percentage was little reduced by salinity (19.5%). Seeds incubated under lower temperature (15°C) with 100 mM NaCl concentration seemed to be subjected to more environmental stress, which is indicated by delayed germination (Al-Khateeb, 2006). In the same meaning, Rahman *et al.* (2008) confirmed that germination was directly related to the amount of water absorbed and the delay in germination was associated to the salt concentration of the medium.

Our results proved that seedling emergence was significantly affected by temperature, it decreased substantially in both saline and non saline treatments at 15°C. Besides, 25°C was the appropriate temperature for final emergence percentage on the establishment of okra seedlings. Other researcher suggested that 15°C was the optimum temperature for final germination percentage of three cultivars barley seeds and 15 and 21°C for germination rate. The optimum temperatures for early root growth were 21 and 27°C regardless of water stress level (Ghazi *et al*., 2007).

High temperature regime at higher salinity concentration inhibited seed emergence. Lower temperature regime decreased as well as delayed seedling emergence, the salt treatment reduced the final percentage of seed emergence. According to Rahman *et al.* (2008) salinity affects the seedling growth of plants by slow or less mobilization of reserve foods, suspending the cell division, enlarging and injuring hypocotyls. At 25°C, salt treatment decreased final emergence percentage.

In germinating seeds, starch degradation is initiated by α-amylase (Yamasaki, 2003), that produces soluble oligosaccharides from starch and these are then hydrolysed by β-amylase to liberate maltose.

Our results indicate a lower content of starch in the cotyledons of germinated seeds at 15 and 35°C compared to those germinated at 25°C. It is mentioned that accumulated starch is probably an energy reserve for the high-energy process of organogenesis and provides for osmotic agents in the form of free sugars. Whereas, thermal stress appear to be associated to the degradation and mobilization of starchy cotyledon reserves and seeds needs high amounts of stored carbohydrates for maintenance of developed organs. Salt stress caused a decrease in starch content and an increase in sugar content of okra cotyledons. It has been reported that salt stress affected an active conversion of starch to sugars in germinated sorghum seeds (Thakur and Sharma, 2005). Similar studies with a variety of plants demonstrated that water stress induced conversion of hexoses and other carbohydrates, such as starch, into sugar alcohols (polyols) and proline (Perez-Alfocea and Larher, 1995; Wang *et al*., 1996). Singh (2004) suggested that the greater accumulation of sugar lowers the osmotic potential of cells and reduces loss of turgidity in tolerant genotypes. The increase in sugar levels accompanied by decrease in starch content in cotyledons was not directly linked to the activity of α-and β-amylases in the cotyledons, which is in disagreement with the existing reports (Thakur and Sharma, 2005; Monerri *et al*., 1986; Gupta *et al*., 1993). Furthermore, our results showed a decrease on total amylases activity under NaCl treatment at the suitable temperature. This suggested that total amylases activity was not associated with both reducing sugars and starch remobilization, the fast hydrolysis of starch under salt stress is mainly attributed to the enhanced lipase or protease activity in cotyledons.

Whereas, at lower temperature (15°C) total amylases activity was higher, in spite of the delays in germination and the reductions in emergence that occurred. This result is in disagreement with previous reports providing that low temperature caused conventionally as a lack of enzyme biosynthesis, leads to reduced activity and may contribute to poor lipid mobilization in canola seeds (Nykiforuk and Flanagan, 1994).

Additionally, total amylase activity was reduced by higher temperature (35°C), at which germination occurred rapidly. Under salt treatment, total amylase activity increased considerably. Indeed, the relationship between the speed of germination and amylase expression in germinating okra seeds was not clear at this stage. Sultana *et al.* (2000) have reported that α-amylase plays an important role in the germination process of wheat seeds. Maximum expression occurred at 29°C, although a high temperature of 38°C prevented the synthesis of α-amylase.

Meanwhile sodium concentration increased in okra germinated seeds soaked in salt solution considerably at 15 and 35°C. Sodium chloride treatment decreased germination percentage, Almodares *et al.* (2007) mentioned that germination process might have been stopped as a result of contact of the seeds with high concentration of Na^+ and Cl^- ions.

High concentration of NaCl in the solution might have increased osmotic potential; hence the seeds were unable to imbibe the water required for germination (Nyagah and Musyimi, 2009). High levels of Na^+ inhibit the K^+ up-take, this inhibition was accentuated at 15 and 35°C. Present results agree with those of Alpaslan *et al.* (1999), Grieve and Poss (2000), whose proved in similar research the antagonistic interactions between sodium and potassium, especially under salinity stress conditions.

High ion concentration (Na^+) reported under the high incubation temperature (35°C) could be attributed to the high membrane permeability. Al-Khateeb (2006) suggested that membrane permeability due to high temperature favoured Na^+ uptake in contrast to K^+.

These results confirmed once more that okra was tolerant to salinity at 25°C since it was able to accumulate lower Na^+, potassium content increased slightly at high level salinity.

Under salt stress, plants maintain high concentrations of K^+ and low concentrations of Na^+ in the cytosol. They accomplished this by regulating the expression and activity of K^+ and Na^+ transporters and H^+ pumps that generate the driving force for ion transport (Zhu, 2003).

CONCLUSION

Considering all the results obtained from this study, we concluded that combined salinity and temperature had both additive and interaction effects on okra plants even at germination than at seedling emergence stage. Salinity had a minimal effect on germination of okra under the suitable temperature. Temperature had a dramatic effect on the seedling emergence percentage, particularly at lower and higher temperature. Seeds incubated under low temperature with high NaCl concentrations seemed to be subjected to more environmental stress. Emergence data showed that okra was more sensitive to salt stress at this stage.

Low and high temperature decreased seed germination at 100 mM NaCl as consequence of a decrease on starch level, an increase on reducing sugars content in cotyledons and an accumulation of sodium. At 25°C, germination requires to the embryonic axis to develop a high growth potential which was less affected by salinity.

REFERENCES

Abid, M., S.A. Maleek, K. Bilal and R.A. Wajid, 2002. Response of Okra (*Abelmoschus esculentus* L.) to EC and SAR of irrigation water. Int. J. Agric. Biol., 4: 311-314.

Al-Khateeb, S.A., 2006. Effect of salinity and temperature on germination, growth and ion relations of *Panicum turgidum* forssk. Bioresour. Technol., 97: 292-298.

Al-Thabet, S.S., A.A. Leilah and I. Al-Hawass, 2004. Effect of NaCl and incubation temperature on seed germination of three canola (*Brassica napus* L.) cultivars. Sci. J. King Faisal Univ. Basic Applied Sci., 5: 81-92.

Ali, H., T.C. Tucher, T.L. Thompson and M. Salim, 2001. Effects of salinity and mixed ammonium and nitrate nutrition on the growth and nitrogen utilization of barley. J. Agron. Crop Sci., 186: 223-228.

Allefrey, J.M. and D.H. Northcote, 1977. The effects of the axis and plant hormones on the mobilization of storage materials in the ground-nut (*Arachis hypogaea*) during germination. New Phytologist, 78: 547-563.

Almodares, A., M.R. Hadi and B. Dosti, 2007. Effects of salt stress on germination percentage and seedling growth in sweet sorghum cultivars. J. Boil. Sci., 7: 1492-1495.

Alpaslan, M., A. Gunes and S. Taban, 1999. Salinity resistance of certain rice (*Oryza sativa* L.) varieties. Turk. J. Biol., 23: 499-506.

Anderson, W.H., R.I. Carolus and D.P. Watson, 1960. The germination of okra seed as influenced by treatment with acetone and alcohol. Proc. Am. Soc. Hort Sci., 62 : 427-432.

Ben Ahmed, H., A. Manaa and E. Zid, 2008. Salt tolerance of *Setaria verticillata* L.: A short-cycle poaceae. Comptes Rendus Biologies, 331: 164-170.

Bewley, D., D.W.M. Leung, S. Macisaac, J.S.G. Reid and N. Xu, 1993. Transient starch accumulation in the cotyledons of fenugreek seeds during galactomannan mobilization from the endosperm. Plant Physiol. Biochem., 31: 483-490.

Bewley, J.D. and M. Black, 1994. Seeds, Physiology of Development and Germination. 2nd Edn., Springer-Verlag, New York, ISBN-13: 9780306447488, pp: 460.

Bohnert, H., D.E. Nelson and R.G. Jensen, 1995. Adaptations to environment stresses. Plant Cell, 7: 1099-1111.

Brar, G.S., J.F. Gomez, B.L. McMichael, A.G. Matches and H.M. Taylor, 1991. Germination of twenty forage legumes as influenced by temperature. Agron. J., 83: 173-175.

Chauhan, J.S., Y.K. Tomar, A. Badoni, N.I. Singh and A. Seema, 2009. Morphology, germination and early seedling growth in *Phaseolus mungo* L. with reference to the influence of various plant growth substances. J. Am. Sci., 6: 34-41.

Çiçek, N. and H. Çakirlar, 2002. The effect of salinity on some physiological parameters in two maize cultivars. BULG. J. Plant Physiol., 28: 66-74.

Doymaz, I., 2005. Drying characteristics and kinetics of Okra. J. Food Eng., 69: 275-279.

El-Keblawy, A. and A. Al-Rawai, 2005. Effects of salinity, temperature and light on germination of invasive *Prosopis juliflora* (Sw.) D.C. J. Arid Environ., 61: 555-565.

Ghazi, N., Al-Karaki, A.J. Asadulla and Y. Othman, 2007. Seed germination and early root growth of three barley cultivars as affected by temperature and water stress. Am. Eurasian J. Agric. Environ. Sci., 2: 112-117.

Grieve, C.M. and J.A. Poss, 2000. Wheat response to interactive effects of boron and salinity. J. Plant Nutr., 23: 1217-1226.

Gul, B. and D.J. Weber, 1999. Effects of salinity, light and temperature on germination in *Allenrolfea occidentalis*. Can. J. Bot., 77: 240-246.

Gupta, A.K., J. Singh, N. Kaur and R. Singh, 1993. Effect of polyethylene glycol induced–water stress on germination and reserve carbohydrate metabolism in chickpea cultivars differing in tolerance to water deficit. Plant Physiol. Biochem., 31: 369-378.

Hucl, P., 1993. Effects of temperature and moisture stress on the germination of diverse common bean genotypes. Can. J. Plant Sci. Technol., 73: 697-702.

Khan, M.A., 1999. Comparative Influence of Salinity and Temperature on the Germination of Subtropical Halophytes. In: Halophyte Uses in Different Climates I: Ecological and Ecophysiological Studies, Lieth, H., M. Moschenko, M. Lohman, H.W. Koyro and A. Hamdy (Eds.). Backhuys Publishers, Leiden, pp: 77-88.

Khan, M.A., B. Gul and D.J. Weber, 2001. Effect of salinity on the growth and ion content of *Salicornia rubra*. Commun. Soil Sci. Plant Anal., 32: 1532-2416.

Kursat, C. and K. Kabar, 2007. Comparative effects of some plant growth regulators on the germination of barley and radish seeds under high temperature stress. Eur. Asian J. Bio. Sci., 1: 1-10.

Lafond, G. and D. Fowler, 1989. Soil temperature and moisture stress effects on kernel water uptake and germination of winter wheat. Agron. J., 81: 447-450.

Liu, X., S. Zhang, X. Shan and Y.G. Zhu, 2005. Toxicity of arsenate and arsenite on germination, seedling growth and amylolytic activity of wheat. Chemosphere, 61: 293-301.

Maas, E.V. and G.F. Holfman, 1977. Crop salt tolerance–current assessment. J. Irrigation Drainage Div., 103: 115-134.

Maghsoudi, A. and K. Maghsoudi, 2008. Salt stress effects on respiration and growth of germinated seeds of different wheat (*Triticum aestivum* L.) cultivars. World J. Agric. Sci., 4: 351-358.

Marsh, L., 1992. Emergence and seedling growth of okra genotypes at low tempetaures. Hortscience, 27: 1310-1312.

Monerri, C., A. Garcia-Luis and J.L. Guardiola, 1986. Sugar and starch changes in pea cotyledons during germination. Physiologia Plantarum, 67: 49-54.

Mosjidis, J.A. and X. Zhang, 1995. Seed germination and root growth of several *Vicia* spcies at different temperatures. J. Seed Sci. Technol., 23: 749-759.

Nyagah, A.W. and D.M. Musyimi, 2009. Effects of sodium chloride solution stress on germination and growth of passion fruits seedlings. ARPN J. Agric. Biol. Sci., 4: 49-53.

Nykiforuk, C.L. and A.M.J. Flanagan, 1994. Germination and early seedling development under low temperature in canola. Crop Sci., 34: 1047-1054.

Nykiforuk, C.L. and A.M.J. Flanagan, 1999. Storage reserve mobilization during low temperature germination and early seedling growth in *Brassice napus*. Plant Physiol. Biochem., 37: 939-947.

Perez-Alfocea, F. and F. Larher, 1995. Effects of Phlorizin and p chloromercuribenzenesulfonic acid on sucrose and praline accumulation in detached tomato leaves submitted to NaCl and osmotic stresses. J. Plant Physiol., 145: 367-373.

Rahman, M., U. Soomro, M. Zahoor-ul-Hag and S. Gul, 2008. Effects of NaCl salinity on wheat (*Triticum aestivum* L.) cultivars. World J. Agric. Sci., 4: 398-403.

Singh, A.K., 2004. The physiology of salt tolerance in four genotypes of chickpea during germination. J. Agric. Sci. Technol., 6: 87-93.

Sultana, N., K. Ikeda and T. Mitsui, 2000. GA3 and proline promote germination of wheat seeds by stimulating α-amylase at unfavourable temperatures. Plant Prod. Sci., 3: 232-237.

Swain, R.R. and E.E. Dekker, 1966. Seed germination studies I. Purification and properties of an α-amylase from the cotyledons of germinating peas. Biochimica Biophysica Acta (BBA)–Enzymol. Biol. Oxidation, 122: 75-86.

Thakur, M. and A.D. Sharma, 2005. Salt stress and phytohormone (ABA)-induced changes in germination, sugars and enzymes of carbohydrate metabolism in *Sorghum bicolor* (L.) Moench seeds. J. Agric. Soc. Sci., 1: 89-93.

Tlig, T., M. Gorai and M. Neffati, 2008. Germination responses of *Diplotaxis harra* to temperature and salinity. Flora, 203: 421-428.

Wang, Z., B. Quebedeaux and G.W. Stutte, 1996. Partitioning of (14 C) glucose into sorbitol and other carbohydrates in apple under water stress. Aust. J. Plant Physiol., 23: 245-251.

Yamasaki, Y., 2003. α-Amylase in germinating millet seeds. Phytochemistry, 64: 935-939.

Zadeh, H.M. and M.B. Naeini, 2007. Effects of salinity stress on the morphology and yield of two cultivars of canola (*Brassica napus* L.). J. Agron., 6: 409-414.

Zhu, J.K., 2003. Regulation of Ion homeostasis under salt stress. Curr. Opin. Plant Biol., 6: 441-445.