

Fruit quality and osmotic adjustment of four tomato cultivars under drought stress

A field experiment was conducted on loam soil to study the effect of drought stress on fruit quality and osmotic adjustment in four tomato cultivars in Bangladesh. The water stress treatments were imposed at 82-100 % (T0), 69-85% (T1), 53-67 % (T2), and 40-50 % (T3) of the field capacity. Under stress, the quality of fruits was improved as a result of the synthesis of different acids like ascorbic acid, citric acid and malic acid. The response of solute accumulation in relation to water stress revealed significant increase in glucose, fructose and sucrose in fruits and proline contents in leaves, showed the conspicuous tendency of tomato plants to adjust osmotically against water stress.

An increase of 100% (glucose), 30% (fructose) 72% (sucrose) and 345% (proline) were found at T3 treatment compared with T0. The concentration of citric acid, malic acid and ascorbic acid increased with increasing water deficit in the plants. Water stress increased sugar and different acids and consequently improved the fruit quality. No physical damage due to stress was observed in fruits, which were over 90% red.

1. Introduction

Vegetable crops play a vital role in human nutrition. Tomato (*Lycopersicon esculentum* Mill) is one of the most important and widely cultivated vegetable crop all over the world. It is one of the most popular salad vegetables in the row state and is made into soups, pickles, catchups, sauces and other products. Of more than 100 species of vegetable crops selected for intensive study in representative Asian countries, tomato ranked first (AVRDC 1977). People of Bangladesh especially in the rural areas suffer from malnutrition because of imbalanced diet. Besides majority of the people of Bangladesh suffer from nutritional deficiency particularly of vitamin A & C, iron, calcium and riboflavin.

It is also a respectable source of some key nutrients such as vitamin A, vitamin C, sugar, ascorbic acid, some protein and iron. It also possesses valuable medicinal properties and it is an excellent purifier of blood, so the crop has very diverse functions in humans.

World-wide, tomato was the seventh most valuable commodity crop in 2013, with a gross production value of over \$60 billion (FAOSTAT, 2014). The average consumption of vegetable in Bangladesh is only 82g per head per day as against the required level of 235g (Anon, 1990).

Tomato is sensitive to a number of environmental stresses, especially extreme temperature, inadequate moisture and environmental pollution, salinity, drought and there is a need to develop varieties that can withstand such environmental stresses (Kalloo, 1993). Water availability has substantial impact on the chemical composition and physical properties of plant tissues, which in turn have decisive significance on the quality and yield of plants (Kramer, 1983). Water deficits in tomato compromise fruit yield and also quality (Patane & Cosentino 2010, Nahar and Ullah 2012 and Kuscü et al 2014)

Water deficit by climate change in Bangladesh is a common phenomenon during summer and winter periods. Due to the reduction of moisture levels in this period, the growth of agriculture suffers. To this adds population problem which needs production of more food to feed for 160 million people within an area of 147570 square kilometers (Anon, 1993).

If there were adequate supply of uncontaminated water during this period, Crops can be ensured. Practically winter is the safest period for crop production where there is little chance of crop failure due to climatic reasons but unfortunately our crops suffer from drought during this period. Crops production could be enhanced either by supplying adequate water or by growing drought resistant crops. This could be overcome by selecting crops which have less demand for water or have root systems sufficient to utilize subsurface water.

Water Potential is the physiochemical availability of the water to participate in plant functions and determines the tendency for net water movement within the system. The Overall water potential of the tomato plant is a function of the combined effect of environmental factors and plant characteristics (Waister and Hudson 1970; Turner, 1981).

Generally in hot regions the amount of water necessary for obtaining a given quantity of fresh fruit (Tomato) is five times greater than in humid and cool regions. The difference becomes even greater when one considers the farming profitability of the use of water in greenhouses, where the water consumption decreases even more and the efficiency per mm of water rises further, to the point of doubling the crop per unit of water (Rendon Poblete, 1980). The judicious use of water needs to be made to obtain maximum efficiency when their supply is limited.

In this experiment we have selected tomato crop because it is less susceptible to drought and has extensive root system.

The aim of the present study was to find out a suitable drought resistant tomato variety out of four varieties commonly cultivated in Bangladesh, also to evaluate fruit quality and osmotic adjustment with minimum use of water.

2. MATERIALS AND METHOD

Field experiment was conducted in Dhaka, Bangladesh, geographical location is 20° 34'N-26°38'N and 88° 01'E-92°41'E, mean humidity 79.5%, annual rainfall (average) 2000 mm and maximum annual temperature 36°C and minimum 12°C. The annual precipitation varies from 1500 mm in the north to 5700mm in the northeast (Hussain, 1992) and during the periods from (November-March).

The experiment was conducted to evaluate the fruit quality and osmotic adjustment in tomato plants due to water stress. Four varieties of tomato plants namely, BR-1, BR-2, BR-4 and BR-5 were the test crops.

The seeds of four varieties namely BR-1, BR-2, BR-4 and BR-5 from Bangladesh Agriculture Research Institute (BARI) at Gazipur.

The soil used in the field experiment was of Tejgaon series under Madhupur tract (According to Reconnaissance Soil Survey report of Dhaka District, 1965 reviewed in 1987). For physical and chemical analysis soil samples were collected at a depth of 0-15 cm from experimental station of Bangladesh Agriculture Research Institute at Tejgaon, Dhaka.

Tejgaon soil has a wide range of crop potentialities and is best suited to producing dry land crops. This soil can be successfully used to study the effect of moisture levels on different cultivars of tomato.

The collected soil samples were air-dried ground to pass through 2mm sieve and then mixed thoroughly to make a composite sample. Dry grasses and other vegetative residual parts were discarded from the soil.

The general physical and chemical characteristics of the soil were:

Textural class of soil-loam, sand - 35.80%, silt - 40.20%, clay - 24.00, Moisture at field - 32%, Moisture at wilting-10%, Maximum water holding capacity-45%, Hygroscopic moisture-1.73%, Bulk density-1.39g/cc, Particle density-2.63g/cc, Porosity-47%, pH- 5.1, EC-90 μ S, OM-1.1%, CEO- 14.88 meq / 100g soil, and N-0.07%.

The experiment was carried out in a randomized complete block design with four treatments and three replications for each cultivar. Unit plot size was 1mX1m with four plants per plot.

The land was prepared well by harrowing followed by laddering. The grasses, weeds and other vegetative residual parts were removed from the land. In this experiment spacing were 75 cm between plots, 50 cm between rows and 45 cm between plants.

Cow dung was applied at the rate of 6t/ha at the time of final land preparation. N, P₂O₅ and K₂O were applied at the rate of 260-200-150 kg/ha, respectively.

The entire amount of phosphate, potash and half of the nitrogen were mixed at the time of the preparation of land. The rest half of the nitrogen was applied in two splits, one at 21 days after sowing of plants during vegetative stage and another at flowering stage.

Seeds were sown at BADC (Bangladesh Agriculture Development Corporation) and after 25 days of germination, healthy seedlings of uniform size were transplanted in the field. After transplantation, Plants had been shaded for 4 days to protect them from sunlight.

Twenty one days after transplantation, each row of tomato plant was supported with bamboo stick to prevent lodging. Weeding in the plots were done when necessary.

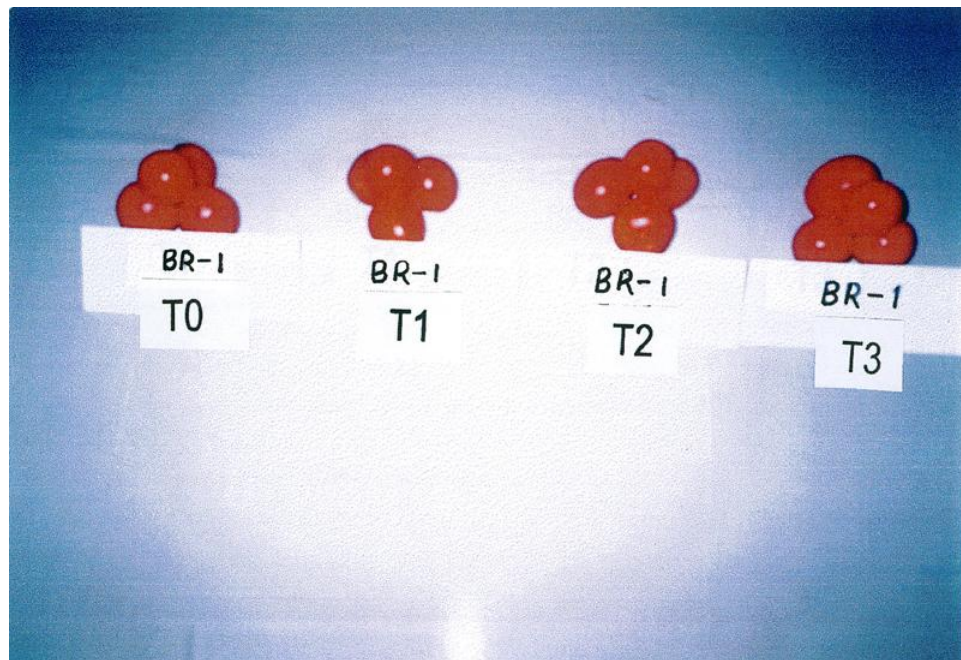
As growth progressed, the tomato plants were attacked by insects. It was therefore, necessary to spray the plants with Malathion (1mI in 1 L water) as insecticide. The insecticide was sprayed as and when required.

The stress period with the cultivars commenced from 28 days after transplantation. The water stress treatments were imposed at 82-100 % (T0), 69-85% (T1), 53-67 % (T2), and 40-50 % (T3) of the field capacity, respectively, in order to investigate the fruit quality and osmotic adjustment of plant. Soil samples were collected at 6 days intervals for measuring the soil moisture percentages from the plots and were measured gravimetrically by drying the soil samples at 105°C for 24 hours. To maintain the above mentioned moisture levels, the soil was irrigated with the amount of water lost by evaporation and transpiration. By addition of irrigation water after six days, the soil moisture levels were within the following ranges: 26-32% (T0), 22-27% (T1), 17-21 % (T2), 13-16% (T3). Water was added weekly to maintain soil moisture at 40-50%, 53-67%, 69-85% and 82-100% of the field capacity throughout the experimental period.

After the end of the experiment, the ripening of the tomatoes were observed and recorded. Young and fresh leaves were taken for biochemical analysis. Three leaves of tomato plants of each plot wrapped in aluminum foil and stored in the deep freeze. These were done just after plucking the leaves from the plants.

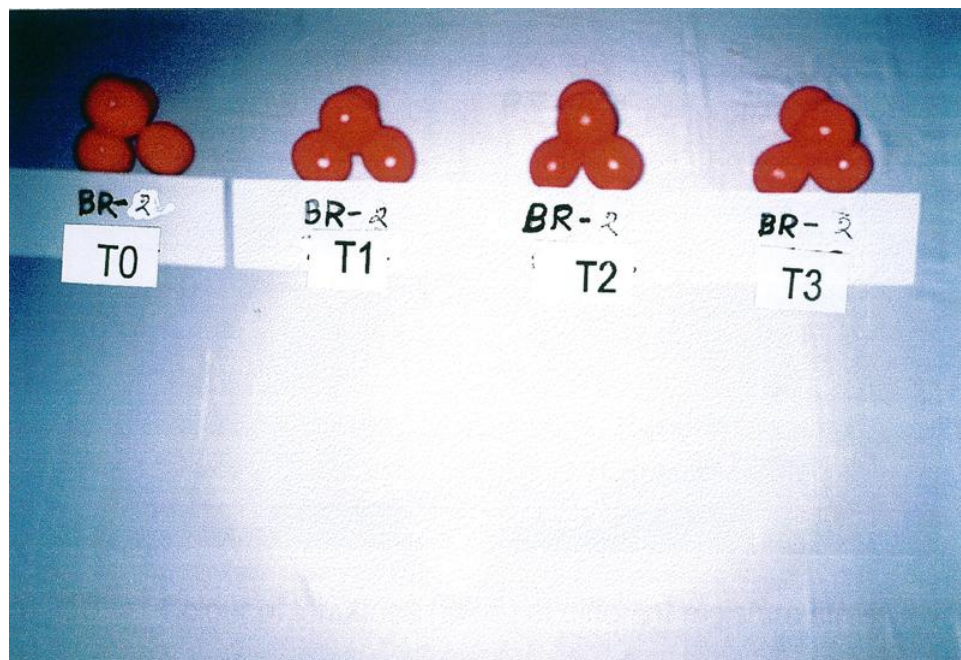
The riped tomatoes were harvested from time to time. After the harvest of the riped tomatoes, fresh weight was recorded and visual quality and physical damage of tomatoes were determined according to the rating, scale of Grierson and Kader (1986). Three tomatoes from each plot were cut into pieces for application of the rating scale for internal tissue damage due to bruising, the rest of the fruits used for other biochemical investigations.

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Plate - 1: Colour of tomatoes (BR-1) at different moisture stress level.



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Plate - 2: Colour of tomatoes (BR-2) at different moisture stress level.

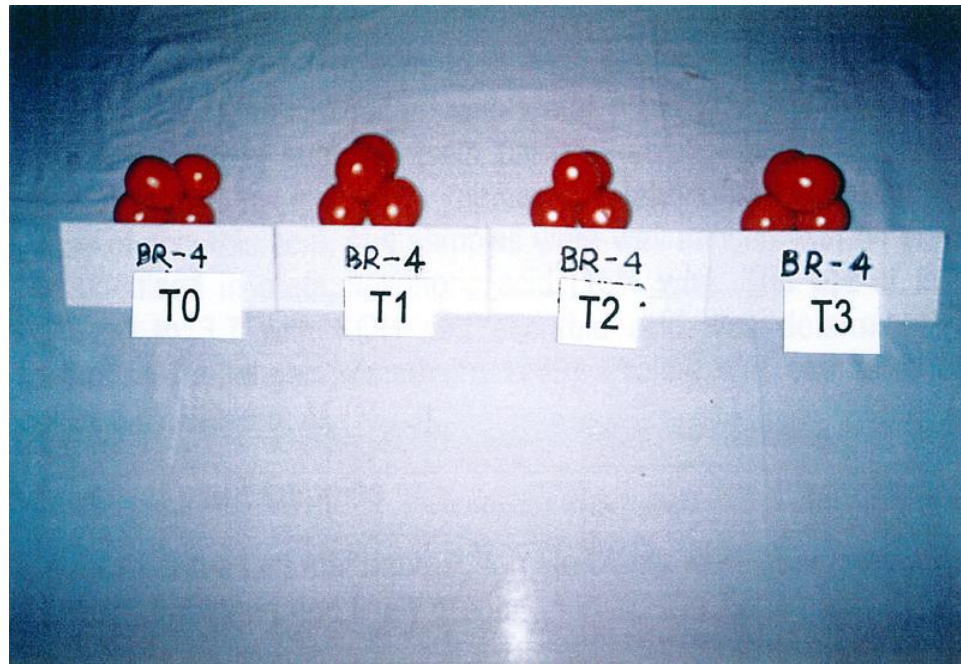


Plate - 3: Colour of tomatoes (BR-4) at different moisture stress level.

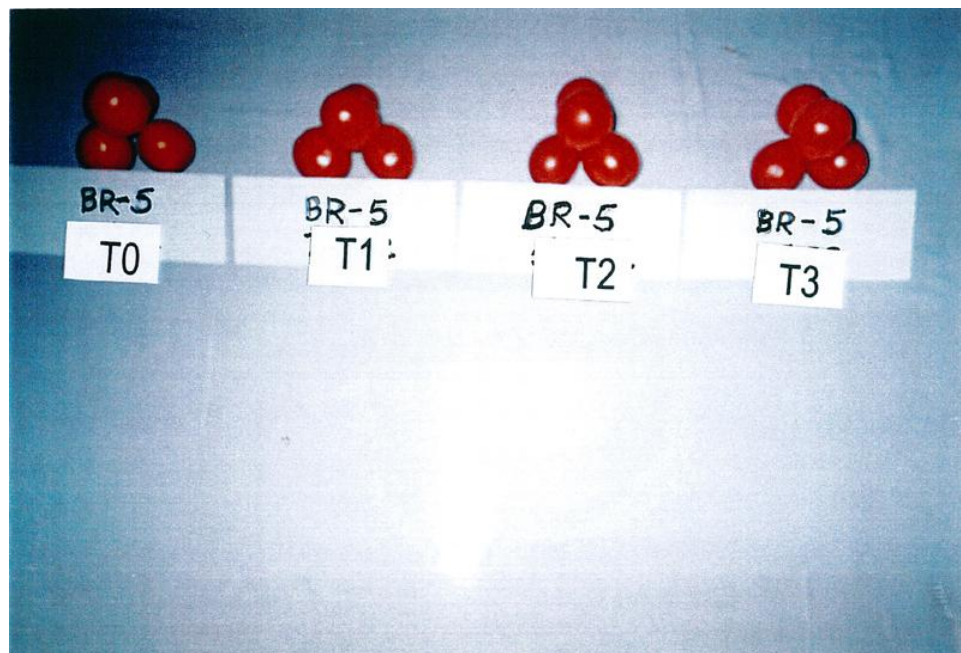


Plate - 4: Colour of tomatoes (BR-5) at different moisture stress level.

Organic solutes like Glucose, fructose, sucrose, malic acid, L-ascorbic acid and citric acid in tomatoes were determined by enzymatic methods described by Boehringer Mannheim (1989). Proline in leaves was estimated by the method outlined by Bates et al. (1973).

Three tomatoes from each plot were minced separately by an electric mixture and extracted with water (60°C). In the extract the contents of glucose, fructose, sucrose, (with carrez - solutions) citric acid and malic acid were analyzed by enzymatic methods (Boehringer- Mannheim 1989). For the assay of ascorbic acid, fruit samples were well minced with an electric mixer and homogenized in metaphosphoric acid (15% w/v). The pH of the mixture was adjusted to 3.7 with KOH and ascorbic acid was determined by enzymatic methods (Boehringer- Mannheim 1989). Proline was estimated by the method outlined by Bates et al. (1973).

For determination of proline in tomato leaves, Purified Proline was used to standardize the sample values.

Reagents:

Acid ninhydrin was prepared by warming 1.25 g ninhydrin in 30 ml glacial acetic acid and 20 ml 6M phosphoric acid, with agitation, until dissolved. Kept cold (Stored at 4°C). The reagent remains stable for 24 hours.

Procedure:

- (a) Approximately 0.5 g of plant material was homogenized in 10 ml of 3% aqueous sulfosalicylic acid and the homogenate filtered through whatman # 2 filter paper.
- (b) Two ml of filtrate was reacted with 2ml acid ninhydrin and 2ml of glacial acetic acid in a test tube for 1 hour at 100°C and reaction terminated in an ice bath.
- (c) The reaction mixture was extracted with 4-ml toluene, mixed vigorously with a test tube stirrer for 15-20 sec.
- (d) The chromophore containing toluene was aspirated from the aqueous phase, warmed to room temperature and the absorbance read at 520 nm using toluene for a blank.
- (e) The proline concentration was determined from a standard curve and calculated on a fresh weight basis.
- (f) To evaluate the quality parameters of plant, enzymatic methods were used (Boehringer — Mannheim, 1989)

For determination of glucose, fructose, sucrose, malic acid and citric acid in tomato fruits, following techniques are used for sample preparation.

The sample was homogenized using a mortar, A well mixed sample was accurately weighed and extracted with hot water (60°C). The extract was transferred quantitatively to a volumetric flask and filled up to the mark with redistilled water. Filtered and used the clear solution for the assay. For clarification (glucose, fructose and sucrose) the following solutions are used 5 ml carrez-I solution (360g potassium hexacyanoferrate-II, $K_4[Fe(CN)_6] \cdot 3H_2O/100ml$. 5ml carrez-II solution (7.20g of $ZnSO_4 \cdot 7H_2O/100ml$ and 10ml NaOH(0.1 mol/L).

L. Ascorbic acid: For the assay of ascorbic acid the tomatoes were well minced with an electric mixture and homogenized with metaphosphoric acid (15%WN). After mincing the pH of the mixture was adjusted to 3.7 with KOH solution.

Finally the results were analyzed statistically employing the Duncan's New Multiple Range Test (DMRT).

3. RESULTS AND DISCUSSION

Effect of water stress on Concentrations of Proline, Glucose, Fructose, Sucrose, Malic acid, Citric acid and L-Ascorbic acid (osmotic adjustment and quality parameters):

Results

Results of these parameters among varieties and treatments are given in table 1-2

Proline:

The proline contents in tomato leaves showed significant difference among the cultivars. It was the highest in BR-2 and the lowest in BR-5. There was no significant difference between BR-4 and BR-1 (Table 1). The concentration of proline increased significantly with increasing water stresses. The highest concentration of 9.16% was observed at T3, which was about 345% higher than that of control (T0) treatment. (Table-2).

Table-1 Content of organic solutes in different cultivars

Cultivars	% Proline	% Glucose	% Fructose	% Sucrose	% Ascorbic acid	% Malic acid	% Citric acid
BR-1	5.21ab	0.66b	0.93ab	1.11b	0.049a	0.32c	0.66a
BR-2	5.79a	0.92a	0.97a	1.84a	0.050a	0.36c	0.70a
BR-4	5.53ab	0.80ab	0.91ab	1.29b	0.051a	0.50a	0.70a
BR-5	4.69b	0.71b	0.86b	1.22b	0.053a	0.45b	0.68a

In a column, means followed by a common letter are not significantly different at the 5% level by DMRT.

Table-2.Effect of different water stress treatments on organic solutes content in plants.

Treatment	% Proline	% Glucose	% Fructose	% Sucrose	% Ascorbic acid	% Malic acid	% Citric acid
T0	2.06d	0.53c	0.79b	0.99b	0.028c	0.26d	0.42d
T1	3.89c	0.67c	0.97a	1.84a	0.050a	0.36a	0.70a
T2	6.12b	0.83b	0.93a	1.47ab	0.059b	0.47b	0.81b
T3	9.16a	1.06a	1.03a	1.71a	0.077a	0.54a	0.94a

In a column, means followed by a common letter are not significantly different at the 5% level by DMRT.

Concentration of Glucose:

The glucose concentration of fruits among the cultivars differed significantly and was found the highest in BR-2 followed by BR-4, BR-5 and BR-1 (Table 1). The contents of glucose in tomato fruits increased significantly with the increase in water stress (Table 2). There was about 100% increase in glucose contents at T3 compared with T0 treatment.

Concentration of Fructose:

Like glucose, fructose contents in tomato fruits were also affected by water stresses. The lowest concentration of fructose was observed at T0 (Table 2), which had about 30% lower fructose content than that of T3 treatment.

Concentration of fructose is also dependent on variety and was found the highest in BR-2 and the lowest in BR-5. There was no significant difference between BR-1 and BR-4 (Table 1).

Concentration of sucrose:

The result of this experiment demonstrated that the sucrose contents in fruits were much higher than glucose and fructose. The concentration was highest in BR-2. However there was no significant variation among the three varieties (Table 1).

The results also revealed that water stress increased the concentration of sucrose than glucose and fructose. The highest concentration was measured at T3 and the lowest at T0 treatment (Table 2). More than 72% increase in sucrose was notice at T3 compared with that on the control (T0).

Malic acid concentration:

Malic acid concentration of fruits among the cultivars differed significantly. The highest concentration was found in BR-4 followed by BR-5. There was no significant difference between BR-2 and BR-1 (Table 1). Malic acid concentration was also affected by water stresses. Increased water stress also increased the synthesis of malic acid. The highest concentration was observed at T3 and the lowest was measured at T0 treatment (Table 2). An increased of 100% malic acid concentration was observed at T3 compared with T0 treatment.

Ascorbic concentration:

The result of ascorbic acid concentration showed that there was no significant different among the cultivars (Table 1). However the treatments differed significantly. Its concentration increased with increasing water stress.

The highest amount of ascorbic acid was found at T3 treatment, while the lowest was at T0 treatment (2). Water deficit significantly increased acid contents in tomato fruits to more than 175% at T3 compared with T0 treatment.

Citric acid concentration:

Citric acid concentrations in tomato fruits showed that there was no significant difference among the cultivars, but the treatments differed significantly from each other.

The highest concentration was found at T3 while the lowest was at T0 treatment. There was an increase of about 124% at T3 compared with T0 treatment. The results also indicate that tomato fruits accumulated more citric acid than malic and ascorbic acids (Table 1).

Ripeness classes of tomatoes were also determined according to Grieson and Kader (1986). The tomatoes were red over 90%, classified as red scored 6 of Grierson and Kader's Table 6.5 in all treatments.

Regarding the internal tissue damage due to bruising, no degree to severity and no visible internal tissue damage were observed (Score Table 6.6 of Grierson and Kader, 1986). in all treatments. Overall visual quality of the fruits was found excellent and essentially no symptoms of deterioration were noticed (Score 9 of Table 6.7; Grierson and Kader, 1986). in all treatments. No symptom of physical damage in any of the treatments could be detected (Score 1 of Table 6., Grierson and Kader, 1986). Ripening and fruit quality studies showed that none of the stress treated tomatoes deteriorated in quality (Plate 1-4). On the other hand water stress enhanced the sweetness of the tomatoes by increasing their glucose, fructose, and sucrose contents and improved the quality by increasing the amount of important acids such as ascorbic acid, malic acid and citric acid.

It was found that the plants had a tendency to adjust against drop in potential in soil by producing organic solutes such as glucose, fructose, sucrose and proline. An increase of 100% (glucose), 30% (fructose) 72% (sucrose) and 345% (proline) was found at T3 treatment compared with T0. The quality of fruits was improved as a result of the synthesis of ascorbic acid, citric acid and malic acid. No physical damage due to stress was observed in fruits, which were over 90% red.

Discussion:

Osmotic adjustment is a key mechanism by which plants adapt to water shortages resulting from an increased solute concentration of cells in order to maintain the water potential gradients needed to ensure continued uptake of water during the stress period. In addition, osmotic adjustment allows cell to maintain the turgor, which is essential for plant growth and various other physiological processes.

According to Greenway and Munns (1980), Flowers et al. (1977), McCree (1986), Torrecillas et al (1995), Ullah et al (1993, 1994, 1997), Nahar and Gratzmacher (2002), Nahar et al (2011) and Dekoum et al (2016) plants synthesize and accumulate organic molecules such as glucose, fructose, proline etc, which act as osmotica and play important role in osmotic adjustment in plants at reduced potential.

In this experiment, the proline concentration in tomato leaves increased with increasing water stress. This result is in agreement with others (Aloni and Rosenstein 1984, Nahar et al 2014) who reported that proline accumulation during water stress was the greatest in tomato varieties.

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In this experiment, the content of glucose, fructose, sucrose, ascorbic, malic and citric acid in tomato increased significantly with water stress. This result with the findings of Ullah et al. (1993, 1994, 1997), who reported a significant increase in glucose, fructose, in some cases sucrose, acids and proline contents in faba beans and tomato by salt stress improving fruit quality.

Ripeness classes of tomatoes were determined according to Grierson and Kader (1986). The tomatoes were red over 90%, classified as red scored 6 of Grierson and Kader's table 6.5 in all treatments. Regarding the internal tissue damage due to bruising, no degree to severity and no visible. internal tissue damage was observed (Score Table 6.6 of Grierson and Kader, 1986). in all treatments. Overall visual quality of the fruits was found excellent and essentially no symptoms of deterioration were noticed (Score 9 of Table 6.7; Grierson and Kader 1986). No symptom of physical damage in any of the treatments could be detected (Score 1 of Table 6., Grierson and Kader, 1986). Ripening and fruit quality studies showed that none of the stress treated tomatoes deteriorated in quality. On the other hand water stress enhanced the sweetness of the tomatoes by increasing their glucose, fructose, and sucrose contents and improved the quality by increasing the amount of important acids such as ascorbic acid, malic acid and citric acid.

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