Journal of Pure and Applied Agriculture (2024) 9(2): 76-85
ISSN (Print) 2617-8672, ISSN (Online) 2617-8680
http://jpaa.aiou.edu.pk/

Identification of thermo-tolerant and thermo-sensitive cucumber genotypes on the basis of morpho-physiological and biochemical markers at seedling stage

Rozina Kazimi[1], Zahoor Hussain[1, 2*], Shahid Iqbal[1], Faheem Khadija[3], Muhammad Saleh Javed[2], Ahtisham Sana[2], Muhammad Safdar[2] and Iqra Arooj[4]

[1]College of Agriculture, University of Sargodha, Sargodha, Pakistan
[2]Department of Horticulture, Ghazi University, Dera Ghazi Khan, Pakistan
[3]Mango Research Station, Shujabad-Multan, Pakistan
[4]Sargodha Campus - Superior University, Lahore, Pakistan

*Corresponding author: Zahoor Hussain (zhussain@gudgk.edu.pk)

Abstract

Cucumber (*Cucumis sativus* L.) is the important vegetable crop in Pakistan. Abiotic factors such as water, salt and heat stress reduce its yield. Among these factors, temperature is a vital abiotic factor, which seriously restricts the plant growth and development. The aim of this research was to screen out the thermo-tolerant cucumber genotypes on the basis of morpho-physiological and biochemical markers at seedling stage. It was a pot culture experiment in which seeds of ten different genotypes of cucumber were sown in plastic pots and put in the growth chamber. Six seeds were sown in each pot with three replications. The plants were exposed to three different heat stresses 23˚C$\pm$2, 40˚C$\pm$2 and 45 ˚C ± 2 ˚C. The thermo-tolerance of selected genotypes was assessed at the seedling stage. In current study, it was concluded that Safoora F_1 Hybrid and Fazal F_1-Hybrid Cucumber can be categories as heat tolerant, while Khushab Local, Summer Green, Bush Green, C-200, C-400 and Cucumber Desi were regarded as heat sensitive cucumber genotypes at seedling stage because C-200 and Cucumber Desi attained minimum shoot length (0.99 cm, 0.83 cm), shoot fresh weight (9.5 g, 8.06 g), shoot dry weight (0.01 g, 0.03 g), root fresh weight (0.44 g, 0.43 g), root dry weight (0.004 g, 0.001 g), osomatic potential (12.88, 12.54 Ψ_w), Relative water content (19.87% ,15.99%), stomatal conductance (0.01 mmol m^{-2} s^{-1},0.00 mmol m^{-2} s^{-1}), photosynthesis rate (0.21 μmol CO_2 m^{-2} S^{-1}, 0.00 μmol CO_2 m^{-2} S^{-1}), proline contents (4.63 μmol g^{-1}, 4.07 μmol g^{-1}), phospours contents (1.81ppm, 1.08 ppm, and potassium contents (1.52 ppm, 1.03 ppm) at 45˚C, respectively. It was concluded that Safoora F1Hybrid and Fazal F_1-Hybrid Cucumber can be categories as heat tolerant, while Khushab Local, Summer Green, Bush Green, C-200, C-400 and Cucumber Desi were regarded as heat sensitive cucumber genotypes.

Keywords: Cucumber, Heat stress, Thermo-sensitive cucumber genotypes, Thermo-tolerant cucumber genotypes

To cite this article: Kazimi, R., Hussain, Z., Iqbal, S., Khadija, F., Javed, M. S., Sana, A., Safdar, M., & Arooj, I. (2024). Identification of thermo-tolerant and thermo-sensitive cucumber genotypes on the basis of morpho-physiological and biochemical markers at seedling stage. *Journal of Pure and Applied Agriculture*, *9*(2), 76-85.

Introduction

Cucumber is an important summer vegetable crop in Pakistan. It is an important member of the family Cucurbitaceae. It is native to Asia, and cultivated history is about 3000-year ago (Siddique et al., 2017; Ammar and Arif, 2019). Now a day cucumber is grown all over the world (Ali, 2018; Ali et al., 2021). Its fruit approximately tubular and it has elongated shape. Its fruit can attain the length of 60cm. It is cultured all over the place in the world and used as fresh vegetable. Cucumber's foliage and mature fruit are used as salads in some area of the world (Ajayeoba et al., 2015). This fruit is consumed mostly for preserving and for carving in Asia. Its young branches are consumed as soup and seed are also used as drawing out of oil. Cucumber's raita is mixture of cucumber and curd which is further general recipe in India. Also being extensively used for cooking purposes it is used in facemask creams, ointments, and antiseptics. This anti-inflammatory mediator is known for its acerbic and peaceful properties. In several countries it is also used to reduce the sickness in the body (Grubben & Deton, 2004).

Cucumber is nutritious vegetable which is mostly used as fresh as salad and pickles. However, sometime its fruit has bitter taste which is due to cucurbitacin. It also has cooling effect and keeping the bodies cool. It is good source of different mineral and vitamins such as vitamin A, vitamin C, as well as folic acid. The fruit peel is very important, and it is a rich source of fibres and minerals including potassium, magnesium, and molybdenum (Hameed et al., 2018). The annual world cucumber production more than 71.70 MT (Khater, 2017).

In Pakistan, cucumber is grown in all provinces with area 33,67 hectares and production of 68,664 tones in Pakistan (GOP, 2018-2019). China is the main product of cucumber in the world with respect to area and production and has 62.7% share in total area while 63.59% in whole world's production.

However, the Pakistan share in the world production is negligible. The cucumber is a summer crop which requires 20 - 32 ˚ C temperature for its growth. There are so many factor effects on the production of cucumber such as abiotic and biotic factors weed, insect's pests and diseases attack as well as environmental issue like drought, and temperature stress. However, climate change such as increase in temperature is the main factor which impact on the vegetable production (Ramakrishna & Gokare, 2011). The current study focused on the identification of thermo-tolerant and thermo-sensitive cucumber genotypes on the basis of morpho-physiological and biochemical characteristics at seedling stage.

Materials and Methods

Growth attributes

Leaf area of the fully expanded leaves from each replication was measured with the help of digital leaf area meter (Ll-3100; LI-COR Inc., Lincoln, NE, USA). However, the fully mature leaves were used to measure the leaf area. Similarly, Chlorophyll content was calculated by the help of chlorophyll meter (CCM-200plus; Opti-Sciences, Hudson, NH, USA) in this process we take two readings of each replication then average use for the calculation of contents**.** However, Free proline contents in the leaves were determined following the method of Bates et al. (1973). Leaf samples 0.5g were homogenized in 4 mL of salicylic acid (3%) using a pestle and mortar filtered with Whatman No. 1 filter paper and the volume was made up to 10 mL with sulfo salicylic acid. 2 mL extract was taken in test tubes along with 2 mL of glacial acetic acid and 2 mL ninhydrin reagent (1.25 g ninhydrin dissolved in 30ml glacial acetic acid and 20 mL ortho phosphoric acid) were added. The reaction mixture was boiled in a water bath at 100°C for 30 min. 5ml of toluene was added into it after cooling the reaction mixture and after thorough mixing, the toluene was separated and absorption read at 520 nm in spectrophotometer.

Physiological parameters

Physiological parameters such as photosynthesis and transpiration rate (m mol m-2 S-1) were measured with the help of portable (IRGA) infrared gas Analyzer (LCi-SD; ADC Bioscientific Ltd, Hoddesdon, UK). Dried ground substance of (0.5 g) was use in the digestion tubes then the concentrations of 5 mL of H_2SO_4 were added in each test tube (wolf, 1982). All the test tubes keep warm during the night at room temperature then by using the 0.5ml of H_2O_2 (35%) dispense down the surface of the digestion test tube, placed the tube in the digestion block and heated at the temperature of 35°C until fumes were formed. These were constant to heat for a further 30 minutes the digestion tubes were detached from the block and then cooled 0.5 mL of H_2O_2 as gradually added and positioned the tube backside into the digestion block. The above step was repeated until the cooled digestion substance become colorless. The amount of the extract was maintained up to 50 mL in volumetric flasks. The takeout was clean and used for the purpose of determination of the K and P. Water use efficiency (WUE) was calculated by the following method by IRGA (LCi-SD; ADC Bio-scientific Ltd, Hoddesdon, UK), for water use efficiency (μmol CO_2 mmol^{-1} H_2O) was measured by dividing photosynthetic rate (A) and transpiration rate (E).

Ion analysis

The digested substance (5mL) was in use in the tubes of Kjeldhals, tubes were positioned on the Kjeldhals ammonia distillation part and 5 ml of 40% NaOH were further to every tube boric acid solution (5 mL) was taken in a conical flask with a small number of drops of assorted indicator. While the distillate was about 40 ml, the distillation was stopped up. the distillate was cooled for little time and titrated it with 0.01 N average. H_2SO_4 till the solution curved in pink color. A blank was run for the whole process.

Nitrogen was predictable by the below formula:

N % age = (V2 – V1) × N × 0.014 ×100
Whereas
V1 = volume of average H_2SO_4 necessary to titrate the Blank solution.
V2 = volume of average H_2SO_4 necessary to titrate the sample solution.
N = Normality of H_2SO_4
W = Weight of the sample

Statistical analysis

Data were analyzed with the help of help of Statistix 8.1 and least significant difference (LSD) test at 5 percent probability level was used to compare the differences among treatments (Steel et al.,1997).

Results

Effect of different heat stress on morphological attributes

Effect of heat stress on shoot length (cm), shoot fresh weight (g) and shoot dry weight (g) at seedling stage in different cucumber genotypes

Table 1 showed the significant interact in heat stress with ten genotypes at $p < 0.05$ in relation to shoot length), shoot fresh weight (g) and shoot dry weight (g) at seedling stage in different cucumber genotypes. Safoora F_1-Hybrid had maximum shoot length (38.70 cm) at 23 ˚C. On the other hand, the less interaction was recorded in Cucumber Desi at 40 ˚C and at 45 ˚C leaving shoot length of 5.54 cm and 0.83 cm, respectively. It was found that cucumber Safoora F_1 Hybrid and Fazal F_1-Hybrid were more heat tolerant through keeping the maximum values 38.70 cm and 36.84 cm, respectively of shoot length.

Cucumber Ana, Khushab Local, C-200 and C-400 had the range 0.9-3.70 cm of shoot length at 45 ˚C and were heat sensitive. It was observed that mortality occurred in C400 and C-200 at maximum heat 45 ˚C. The treatments effect on shoot fresh weight was found significant and variation among ten genotypes was also significant at maximum heat stress (45 ˚C). The maximum shoot fresh weight (SFW) was observed in control in Safoora F_1-Hybrid (24.44 g), while the minimum SFW was recorded at 45 ˚C in Cucumber Desi (0.23 g). Similarly, Cucumber Ana, Green long, Khushab Local and Bush Green genotypes maintained 6.70 g, 4.68 g, and 3.68 g shoot fresh weight, respectively. Summer Green, C-400 and C-200 achieved 1.53 g, 0.23 g, and 0.44 g, respectively of SFW at 45 ˚C. These genotypes were classified as semi-tolerant at maximum heat. However, it was found that maximum level of heat stress (50 ˚C) cause mortality of plants. Statistical analysis showed a significant interaction of Safoora F_1 Hybrid cucumber genotype and heat stress levels at $p < 0.05$ in terms of plant dry weight. The highest interaction dry weight (0.92 g) was observed in Safoora F1-Hybrid at 45 ˚C. On the other hand, the minimum values of the dry weight recorded in C-200 (0.01 g).

Effect of heat stress on root fresh weight (g), root dry weight (g) and on leaf osmotic potential (Ψ_w) at seedling stage in different cucumber genotypes

Results showed that increasing levels of heat stress related significantly ($p< 0.05$) with cucumber genotypes at in terms of root fresh weight (Table 2). Data regarding root fresh weight influenced by different treatments of heat stress. It was described from the data that the maximum root fresh weight was observed in Safoora F_1Hybrid (6.58 g) at 23 ˚C, while minimum root fresh weight (0.43 g) was revealed C-200 at 45 ˚C. Heat stress has significant effect on root dry weight of all cucumber genotypes. Safoora F_1 Hybrid and Fazal F_1 Hybrid showed maximum values of root dry weight (2.69 g and 2.53 g, respectively) at 23 ˚C. Similar, findings were observed at 40 and 45 ˚C. However, Cucumber Desi, C-400 and C-200 showed the lowest root dry weight at 40 and 45 ˚C than all other genotypes. Different treatments of heat stress significantly ($p< 0.05$) effect on leaf osmotic potential. The maximum leaf osmotic potential was observed in Safoora F_1-Hybrid, and Cucumber Ana at 23 ˚C. However, the highest leaf osmotic potential of (16.36 Ψ_w) was observed in Safoora F_1 Hybrid at 40 ˚C and it was heat tolerant genotype compared all other genotypes (Table 2).

Table 1 Effect of heat stress on shoot length (cm), shoot fresh weight and shoot dry weight and at seedling stage in different cucumber genotypes

Genotypes	Shoot length (cm)			Shoot fresh weight (g)			Shoot dry weight (g)		
	23˚C±2	40˚C±2	45˚C±2	23˚C	40˚C	45˚C	23˚C	40˚C	45˚C
Safoora F_1 Hybrid	38.70^a ±2.98	32.56^a±0.78	14.50^a±1.02	24.438^a	12.83^a	9.504^a	4.57^a	1.82^a	0.92^a
Fazal F_1-Hybrid	36.84^b ±2.81	29.62^b±1.04	12.06^b ±0.51	21.20^b	8.20^b	8.06^{ab}	4.29^{ab}	1.61^{ab}	0.80^a
Cucumber Ana	34.42^c ±2.51	9.42^d±0.48	3.70^d ±2.79	19.17^{bc}	6.99^d	6.70^b	4.15^{ab}	1.37^{abc}	0.58^a
Green Long	35.40^{bc}±1.92	10.00^c±2.44	3.88^c ±1.36	17.63^{cd}	6.22^c	4.68^c	3.67^{abc}	1.01^{abc}	0.32^a
Khushab Local	31.85^d ±1.11	5.99^{cd}±1.52	2.93^{cd} ±2.27	17.30^{cde}	3.27^d	3.68^{cd}	3.42^{bcd}	0.79^{abc}	0.15^{ab}
Bush Green	30.92^e ±0.68	5.68^e ±1.12	2.80^e ±1.64	17.04^{cde}	3.25^d	2.80^{de}	3.39^{bcd}	0.53^{abc}	0.10^{ab}
Summer Green	31.33^{de}±1.13	5.72^{de} ±2.15	1.83^f ±2.37	16.54^{de}	2.74^d	1.53^{ef}	2.86^{bcd}	0.29^{abc}	0.05^{ab}
Cucumber Desi	29.35^f ±1.73	5.54^f ±2.34	0.83^{fg}±1.60	15.05^{ef}	1.58^d	0.23^f	2.41^{bcd}	0.13^{bc}	0.03^b
C-400	31.77^{de}±2.30	5.84^{ef} ± 0.95	0.94^g ±2.88	15.47^f	2.10^d	0.44^f	1.98^{cd}	0.10^c	0.02^b
C-200	30.01^{ef}±3.03	6.03^{cd} ±1.98	0.99^g ±2.27	16.58^f	2.04^d	0.440^f	1.73^d	0.09 ^{c}	0.01^b

Mean sharing similar letter in a row or in a column are statistically non-significant, while different lettering shows significant difference at $P \leq 0.05$.

Effect of heat stress on relative water content (RWC), stomatal conductance (mmol m^{-2} s^{-1}) and net photosynthesis rate (µmol CO_2 m^{-2} S^{-1}) at seedling stage in different cucumber genotypes

Relative water content (RWC) indicated significant result of increasing levels of heat stress on cucumber genotypes at $p< 0.05$. It was recorded that RWC was reduced with the increasing levels of heat stress. Cucumber Desi showed minimum RWC at 45 ˚C by reducing water contents up to 15.99%. Safoora F_1-Hybrid and Fazal F_1-Hybrid achieved the maximum water contents (90.71 and 89.67%, respectively) at 23 ˚C, which were significantly higher than all other genotypes. The photosynthesis and stomatal conductance were significantly influenced by heat treatment (Table 3). The ratio of leaf stomata opened increase after heat stress, as there was an inverse relation between heat stresses and stomata conduct as shown in Table 3. The stomata conduct was increased due to heat stress in all genotypes compared to non-stressed condition. Minimum Stomata conduct was observed in C-400 and Cucumber Desi

(10.60 mmol m^{-2} s^{-1} and 9.98 mmol m^{-2} s^{-1}, respectively) at 45 ˚C. The ratio of leaf stomata opened increase after heat stress, as there was an inverse relation between heat stresses and stomata conduct as shown in Table 3. The stomata conduct was increased due to heat stress in all genotypes compared to non-stressed condition. Minimum Stomata conduct was observed in C-400 and Cucumber Desi (10.60 mmol m^{-2} s^{-1} and 9.98 mmol m^{-2} s^{-1}, respectively) at 45 ˚C.

Effect of heat stress on: Proline contents (µmol g^{-1}), chlorophyll content (mg g^{-1}) of plants and nitrogen content in leave (ppm) at seedling stage in different cucumber genotypes

Compared to the non-stress plant, heat treatment increases the leaf proline content significantly. The minimum leaf proline content was observed in Cucumber Desi (3.21 µ moles g^{-1}) and C-400 (2.62 moles g^{-1}) at. 40 ˚C Fazal F_1-Hybrid and Bush Green at the same levels had significantly more proline contents (5.19 µ moles g^{-1} and 5.19 µ moles g^{-1}). Proline contents were increased under all heat treatment compared to non-stress treatment in all genotypes. The analysis of data showed that Fazal F_1 Hybrid and Safoora F_1 Hybrid have the highest leave proline contents (6.73 µmol g^{-1} and 6.09 µmol g^{-1}) than other genotypes at extreme heat 45˚C and showed to be heat tolerant. These genotypes are grouped as semi heat tolerant genotypes than Fazal F_1 Hybrid Cucumber and Safoora F_1 Hybrid. Although, with increasing stress levels the chlorophyll contents were decreased in ten cultivars. Chlorophyll substances were significantly reduced to (2.50 mg g^{-1}) at 45 ˚C compared with 23 ˚C (4.87 mg g^{-1}) in Safoora F_1-Hybrid. Cucumber Desi and C-200 have the least interaction with the heat stress level of 40 ˚C reducing the chlorophyll content to (0.22 mg g^{-1} and 0.31 mg g^{1}). It was determined that the Safoora F_1 Hybrid and Fazal F_1 hybrid genotypes of cucumber were exhibited top chlorophyll content than other cultivars at 45 ˚C. Nitrogen contents in the leave of the cucumber genotypes were significantly affected under various levels of heat stresses. Safoora F_1-Hybrid showed higher stress result than all other cultivars. At control 23˚C, it had (6.73 ppm) and under 40˚C by achieving (4.03 ppm) of leaf nitrogen content.

Table 2 Effect of heat stress on root fresh weight (g) and root dry weight (g) and on leaf osmotic potential (Ψ_w) at seedling stage in different cucumber genotypes

Genotypes	Root fresh weight (g)			Root dry weight (g)			Leaf osmotic potential (Ψ_w)		
	23˚C±2	40˚C±2	45˚C±2	23˚C±2	40˚C±2	45˚C±2	23˚C±2	40˚C±2	45˚C±2
Safoora F_1 Hybrid	6.58^{a}	3.44^{a}	2.84^{a}	2.69^{a}	2.82^{a}	1.63^{a}	15.14^{ab} ±0.91	16.36^{a} ±1.69	15.14^{ab} ±0.91
Fazal F_1-Hybrid	6.34^{ab}	3.22^{ab}	2.60^{ab}	2.53^{ab}	2.71^{ab}	1.40^{ab}	15.21^{b} ±1.58	15.65^{b} ±1.07	15.21^{b} ±1.58
Cucumber Ana	5.80^{abc}	3.18^{ab}	2.19^{ab}	2.41^{ab}	2.53^{abc}	1.11^{d}	15.24^{a} ±0.83	15.77^{b} ±1.43	15.24^{a} ±0.83
Green Long	5.27^{abc}	2.86^{abc}	1.88^{abc}	2.37^{ab}	2.41^{abc}	0.98^{cd}	13.97^{c} ±0.81	14.07^{c} ±0.95	13.97^{c} ±0.81
Khushab Local	$5.12a^{bc}$	2.22^{abc}	1.65^{abc}	2.25^{ab}	2.20^{abc}	0.73^{c}	13.84^{c} ±2.36	12.93^{c} ±1.25	13.84^{c} ±2.36
Bush Green	$4.78a^{bc}$	$2.38a^{bc}$	1.52^{abc}	1.68^{ab}	1.63^{abc}	0.52^{d}	12.83^{bc} ±1.49	11.92^{d} ±1.36	12.83^{bc} ±1.49
Summer Green	4.35^{abc}	1.56^{bc}	1.08^{bc}	1.49^{ab}	1.24^{bc}	0.3^{e}	13.06^{bc}± 1.69	12.62^{c}± 1.60	13.06^{bc}± 1.69
Cucumber Desi	3.93^{bc}	1.72^{bc}	0.44^{e}	1.42^{b}	1.10^{bc}	0.02^{f}	8.63^{f} ±1.04	10.09^{c} ±1.16	7.54^{f} ±1.77
C-400	3.95^{bc}	1.44^{c}	0.45^{c}	1.38^{b}	0.92^{c}	0.06^{ef}	11.92^{d} ±0.58	11.50^{d} ±0.87	11.92^{d} ±0.58
C-200	3.99^{c}	11.40^{c}	0.43^{c}	1.25^{b}	0.72^{c}	0.01^{ef}	12.94^{bcd}±1.17	9.88^{e} ±1.15	12.94^{bcd} ±1.17

Mean sharing similar letter in a row or in a column are statistically non-significant, while different lettering shows significant difference at $P \leq 0.05$.

Effect of heat stress on phosphorous content and potassium content (ppm) in leaves at seedling stage in different cucumber genotypes

The influence of different heat treatments on cucumber genotypes described the significant statistical differences at $p < 0.05$ in terms of phosphorous (P) contents in leave. The highest phosphorous was recorded for P contents in Safoora F_1-Hybrid (6.50 ppm) at control 23 ˚C compared to (45 ˚C) that showed (4.75 ppm). Similarly, minimum (1.33 ppm) P contents were revealed in Cucumber Desi under maximum heat stress (45 ˚C). At 40 ˚C and 23 ˚C the P contents were decreased (3.91 and 2.62 ppm) respectively in Cucumber Desi. Increasing heat stress treatments (40 ˚C and 45 ˚C) caused a significant reduction in P contents in leave in all selected genotypes. It was proved that Safoora F1-Hybrid and Fazal F1-Hybrid Cucumber were kept the maximum P contents in leave **(**4.75 and 3.62 ppm, respectively**)** than other genotypes at 45˚C. Potassium (K) contents of the cucumber genotypes were significantly affected by the increasing levels of heat stress. The greatest interaction of treatment and genotypes at p>0.05 has been shown both by Safoora F1-Hybrid and Fazal F1-Hybrid, having K contents 5.60 ppm and 5.20 ppm, respectively at 23 ˚C. A statistically significant decrease in the K contents was recorded for all the ten genotypes under high heat stress (45 ˚C). The minimum potassium contents (1.07 ppm) at 45 ˚C in Cucumber Desi as shown in Table 5.

Table 3 Effect of heat stress on relative water content (RWC), stomatal conductance (mmol m^{-2} s^{-1}) and net photosynthesis rate (µmol CO_2 m^{-2} S^{-1}) at seedling stage in different cucumber genotypes

Genotypes	Relative water content (RWC)			Stomatal conductance (mmol m^{-2} s^{-1})			Net photosynthesis rate (µmol CO_2 m^{-2} S^{-1})		
	23˚C±2	40˚C±2	45˚C±2	23˚C±2	40˚C±2	45˚C±2	23˚C±2	40˚C±2	45˚C±2
Safoora F1 Hybrid	90.71^{a} ±1.03	65.45^{a} ±1.13	43.75^{a} ±0.54	2.34^{e} ±0.32	4.78^{d} ±1.07	7.02^{d} ±0.67	5.39^{a} ±0.19	2.43^{a} ±0.71	1.470^{a} ±0.36
Fazal F1-Hybrid	89.67^{b} ±2.65	60.2^{b} ±2.18	40.00^{b} ±0.88	2.06^{ef} ±.23	4.72^{e} ±1.00	6.33^{e} ±0.50	5.15^{a} ±0.32	2.36^{a} ±0.56	1.35^{a} ±0.22
Cucumber Ana	88.13^{b} ±1.30	58.78^{c} ±0.51	34.10^{c} ±1.11	2.96^{e} ±1.34	5.18^{bc} ±0.10	7.03^{cd}±0 .13	4.70^{b} ±0.39	1.51^{b} ±0.72	1.27^{b} ±0.07
Green Long	84.99^{cd} ±2.07	54.44^{bc} ±0.98	26.09^{d} ±1.09	3.01^{cd} ±1.78	4.52^{d} ±1.12	8.60^{d} ±0.11	3.61^{c} ±0.27	1.24^{c} ±0.52	1.10^{c} ± 0.17
Khushab Local	86.10^{c} ±1.11	56.43^{bc} ±0.92	28.77^{cd} ±0.47	3.06^{cd} ±1.22	5.03^{bc} ±1.56	8.79^{d} ±0.23	3.43^{bc} ±0.05	1.22^{c} ±0.12	1.00^{bc} ±0.19
Bush Green	82.5^{d} ±0.90	48.12^{d} ±1.09	18.19^{f} ±1.37	3.87^{d} ±1.99	5.60^{c} ±1.32	7.86^{cd}±0 .12	3.82^{d} ±0.06	0.98^{d} ±0.15	0.94^{d} ±0.42
Summer Green	80.76^{e} ±0.58	45.2^{e} ±1.52	20.67^{e} ±1.45	4.99^{c} ±1.89	7.70^{ab} ±1.33	9.98^{c} ±0.43	3.71^{cd} ±0.12	0.93^{d} ±0.85	0.83^{d} ±0.05
Cucumber Desi	79.13^{f} ±0.17	34.78^{g} ±0.26	15.99^{g} ±0.47	6.56^{a} ±0.98	8.38^{a} ±0,77	10.6^{a}±0. 07	3.33^{ef} ±055	0.42^{ef} ±0.40	0.14^{g} ±0.17
C-400	81.88^{de} ±1.86	46.10^{de} ±0.62	17.13^{ef} ±0.08	2.51^{e} ±0.76	4.80^{d} ±1.90	7.87^{cd}±0 .46	3.41^{f} ±0.44	0.92^{f} ±0.68	0.42^{de} ±0.27
C-200	79.97^{f} ±0.90	38.08^{ef} ±0.56	19.87^{e} ±1.19	5.03^{b} ±0.76	7.51^{b} ±0.12	9.62^{b} ±0.34	3.45^{e} ±0.32	1.00^{e} ±0.38	0.33^{f} ±0.12

Mean sharing similar letter in a row or in a column are statistically non-significant, while different lettering shows significant difference at P ≤ 0.05

Table 4 Effect of heat stress on: Proline contents (µmol g^{-1}), chlorophyll content (mg g^{-1}) of plants and nitrogen content in leave (ppm) at seedling stage in different cucumber genotypes

Genotypes	Proline contents (µmol g^{-1})			Chlorophyll content (mg g^{-1}) of plants			Nitrogen content in leave (ppm)		
	23˚C±2	40˚C±2	45˚C±2	23˚C±2	40˚C±2	45˚C±2	23˚C±2	40˚C±2	45˚C±2
Safoora F_1 Hybrid	5.46^{a} ± 0.07	6.01^{a} ±0.87	6.73^{a} ±0.03	4.87^{a} ±0.92	4.47^{a} ±0.76	2.50^{a} ±0.42	6.73^{a} ±0.01	5.97^{a} ±0.61	4.03^{a} ±0.71
Fazal F_1-Hybrid	4.99^{b} ±0.03	5.19^{ab} ±0.06	6.09^{b} ±0.017	4.66^{b} ±0.25	4.26^{b} ±0.31	1.09^{b} ±0.14	5.99^{b} ±1.15	4.92^{b} ±0.02	3.78^{b} ±0.48
Cucumber Ana	2.15^{d} ±0.06	3.30^{ef} ±0.19	4.52^{ef} ±0.02	4.40^{ab} ±0.32	3.10^{ab} ±0.28	0.94^{c} ±0.16	3.80^{cd} ±1.13	2.17^{ef} ±0.07	1.24^{e} ±0.54
Green Long	3.72^{ab} ±0.13	4.06^{d} ±0.07	5.04^{bc} ±0.03	3.54^{bc} ±0.67	2.26^{c} ±0.54	0.32^{d} ±1.53	5.04^{ab} ±0.03	3.53^{d} ±0.02	2.99^{ab} ±0.71
Khushab Local	2.92^{c} ±0.11	3.96^{e} ±0.02	5.03^{bc} ±0.09	3.80^{c} ±1.18	2.18^{c} ±0.22	0.41^{cd} ±0.65	6.73^{a} ±0.01	5.97^{a} ±0.61	4.03^{a} ±0.71
Bush Green	4.63^{b} ±0.13	5.83^{b} ±0.08	4.15^{d} ±0.04	3.25^{bc} ±1.14	0.82^{d} ±0.32	0.20^{e} ±0.38	4.95^{bc} ±0.01	3.03^{cd} ±0.05	2.75^{c} ±0.06
Summer Green	3.90^{ab} ±0.100	4.80^{c} ±0.04	5.53^{c} ±0.02	2.63^{d} ±1.42	0.42^{e} ±0.27	0.17^{f} ±0.068	4.95^{bc} ±0.01	3.03^{cd} ±0.05	2.75^{c} ±0.06
Cucumber Desi	2.10^{e} ±0.17	3.21^{ef} ±0.33	4.07^{f} ±0.032	2.50^{e} ±0.65	0.22^{f} ±0.34	0.06^{ef} ±0.085	3.74^{e} ±0.01	2.00^{g} ±0.08	1.08^{de} ±1.0
C-400	2.67^{bc} ±0.18	2.62^{f} ±0.78	4.98^{e} ±0.021	2.74^{de} ±0.07	0.31^{ef} ±0.13	0.09^{ef} ±0.02	3.84^{d} ±0.16	2.64^{f} ±1.32	1.97^{d} ±0.07
C-200	2.88^{c} ±0.15	3.86^{e} ±0.77	4.63^{e} ±0.067	2.83^{d} ±0.08	0.50^{e} ±0.19	0.11^{f} ±0.10	3.89^{d} ±0.12	2.94^{e} ±1.15	1.81^{d} ±0.03

Mean sharing similar letter in a row or in a column are statistically non-significant, while different lettering shows significant difference at P ≤ 0.05.

Table 5 Effect of heat stress on: phosphorous content and potassium content (ppm) in leaves at seedling stage in different cucumber genotypes

Genotypes	Phosphorous content in leaves (ppm)			Potassium contents in leaves (ppm)		
	23°C±2	40°C±2	45°C±2	23°C±2	40°C±2	45°C±2
Safoora F1 Hybrid	6.50^{a}±0.03	5.59^{a}±0.02	4.75^{a}±0.02	5.60^{a}±0.25	4.90^{a}±0.22	3.80^{a}±0.04
Fazal F1-Hybrid	5.91^{b}±0.07	4.72^{b}±0.01	3.62^{b}±0.11	5.20^{b}±0.07	4.50^{b}±0.07	3.39^{b}±0.02
Cucumber Ana	3.98^{e}±1.28	2.89^{d}±0.62	1.73^{bc}±0.16	3.70^{d}±2.19	2.63^{cd}±2.19	1.48^{j}±0.11
Green Long	5.06^{c}±0.30	3.86^{c}± 0.14	2.89^{c}±1.21	4.99^{c}±0.02	3.09^{ab}±2.18	2.99^{ab}±0.05
Khushab Local	5.79^{ab}±0.79	4.60^{ab}±1.24	3.05^{ab}±0.01	3.65^{e}±2.13	2.54^{e}±0.26	1.88^{d}±0.01
Bush Green	4.81^{cd}±0.02	3.67^{bc}±0.03	2.99^{c}±0.05	5.00^{ab}±0.01	3.04^{ab}±0.04	2.46^{c}±0.37
Summer Green	3.29^{de}±0.32	2.62^{de}±0.97	1.46^{e}± 0.53	4.71^{bc}±0.02	2.97^{c}±0.02	1.98^{ij}±0.07
Cucumber Desi	3.91^{f}±0.22	1.97^{ef}±0.10	1.33^{de}±0.96	3.45^{de}± 0.14	2.41^{f}±0.25	1.07^{de}±0.02
C-400	4.00^{cd}±0.62	2.08f±0.62	1.50^{d}±0.89	4.06^{bc}±2.11	2.75^{d}±0.13	1.63^{d}±0.05
C-200	4.56^{d}±0.42	2.76^{e}±0.45	1.61^{d}±0.96	3.83^{d}±2.10	2.69^{cd}±0.12	1.52^{e}±0.08

Mean sharing similar letter in a row or in a column are statistically non-significant, while different lettering shows significant difference at $P \leq 0.05$.

Discussion

Worldwide temperature has a universal adverse influence on plant development. The high temperature influence on crop production and the consequence of famine (Bita and Gerats, 2013). In this investigation, we evaluated the effect of temperature modification on summer vegetable production. High-temperature stress influenced on cucumber genotypes in terms of morpho-physiological and biochemical attributes. In this evaluation, we analyzed to identify the plants tolerant to heat stress. As an assumption, we impost potential practices and approaches which could lead to screen out the genotypes with economical production.

In present research, it was concluded that, heat stress significantly disturbs the morphological attributes e.g., stem length, shoot fresh biomass, shoot dry biomass, and root fresh weight and root dry weight. Based on growth imperative challenges, genotypes can be characterized into tolerant and sensitive based. Heat stress adverse relation with shoot length, Cucumber Desi showed minimum shoot length. Safoora F1-Hybrid showed highest shoot length than all other genotypes as presented in seedling, maturity and reproductive periods. Extreme temperature reduced the cell division, which disturbed plant development by influencing the shoot length. Similar results reported by Prasad et al. (2006), they determined that heat stress declined seed viability, shoot growth, plant height, generally plant development and biomass of sorghum. If plants are exposed to high temperature, it decreases the stem development consequently, reduced the plant height endure high temperature for prolong period. It disturbed the morphological attributes such as plants containing leaves, sprouts, flower-buds, plant height, and roots development (Tsukaguchi et al., 2003; Iwaya-Inoue et al., 2004). Our results coordinated with Abdelmageed et al. (2003) who described that high temperature decreases the accumulation of carbohydrates and starch in leaves that compressed the shoot length.

In present study, heat stress also influenced fresh biomass of shoot and root fresh and dry biomass of shoot and root of cucumber genotypes (Safoora F1 Hybrid and Fazal F1-Hybrid) showed maximum value of shoot and root fresh weight. Cucumber Desi showed sensitivity to the temperature at seedling, maturity and reproductive stages. As reported earlier high temperature reduced the fresh biomass of shoot, root, net photosynthetic rate and enhanced the burning of foliage and leaf senescence due to heat stress and development of crops like maize, sugarcane and pea (Wahid et al., 2007). According to Porter and Gawith (1999) also observed that heat stress reduced the roots and shoot, development. Individually, our findings bear a resemblance with another study done by Wahid et al. (2007) which delineate that lowering of such variables could be caused by reduction in the concentration of starch. Our results are in line with these results when, the temperature increased then curling and senescence of leaves finally reduction in fresh and dry weight of plants at seedling, maturity, and reproductive stage (Giaveno & Ferrero, 2003). When heat stress increases, the water vaporized, and salinity developed it reduced the morphological attributes of plants. Our result could be compared with Tantawy et al. (2009) who determined that plant height, total chlorophyll, leaf area, and ionic contents, shoot fresh biomass and root dry weight, as well as yield attributes, declined under salinity in tomato vegetable. Similar findings were observed by Khadija et al. (2022; 2024) who measured the flower, leaf attributes in different cucumber cultivars and embryogenesis from anthers. During current study it was recorded that morphological parameters decreased in all selected cultivars. Safoora F1-Hybrid had the maximum heat tolerant capacity as compared Desi Cucumber. Our investigations matched with the results of Yilmaz et al. (2004) they described that various environmental stress as heat stress, drought and salt stress influenced on the plant growth.

It was recorded from present investigation that heat stress also has a significance effect on the gas exchange characteristics such as photosynthesis rate, stomatal conductance, chlorophyll contents, and water use efficiency in all selected genotypes. As the temperature increased than leaf area condensed, a stomatal disturbance occurred, mesophyll cell damage so green pigment reduced, and leaves become yellow of all the varieties. It was recorded earlier by Shahid et al. (2011) they investigated the photosynthesis rate and stomatal conductance declined in okra (*Abelmoschus esculentus* L.) under heat and salinity stress. They also reported that germination cannot be considered a good parameter for screening under heat stress. It was reported earlier that reduction in growth was early response and a common attribute in plants when subjected to environmental stresses like heat, salinity, and drought *in vitro* or in vivo were recorded in soybean (Zhang et al., 2008) and in okra (Wu & Cosgrove 2000). Similarly, results from the *Arabidopsis* genome studies showed that many genetic mechanisms including cell and genome protection in contradiction of biotic and abiotic stress are reserved in eukaryotic cells (Joyce et al., 2003).

Heat stress has a strong link with salt stress because more water transpired, and salinity occurred will reduce the osmotic potential in leaves were stimulated. Heat stress also has significance influence on chlorophyll contents, transpiration rate, and photosynthesis as well as water use efficiency of plants of ten selected genotypes of cucumber. A particular temperature is necessary for plant growth, less or above vital temperature cause of influence the plant evolution sequence. Photosynthesis, stomata conductance, chlorophyll contents, transpiration rate, and water use efficiency are vital to process and key factors in determining the heat tolerant and heat sensitive genotypes. Thuzar et al. (2010**)** reported that temperature interrupts through an effect on photosynthesis rate, leaf area expansion, and fruit development. They also observed that the photosynthesis rate per unit area and a number of seeds have a strong correlation with crop development.

Kim et al. (2007) described that the plant's grow in heat stress influenced the biomass, enzyme activities, leaf area and photosynthesis process in plants. More reduction was recorded temperature increased gradually. Shah and Paulsen, (2003) stated that the process of photosynthesis, shoot length, fresh biomass and leaf area of wheat crops are rapidly declined under extreme temperature. Yamane et al. (2000) reported that when elevated in the temperature then stomatal and non-stomatal restraint occurred. Dekov et al. (2001) described that chlorophyll accumulation reduced structural and functional interrupting of chloroplasts in temperature stress. The outcomes of the present study also proved that whenever temperature increased gradually then, upgraded the water used efficiency at all phases of cucumber’s genotypes. water and sun light are essential for the photosynthesis process, when heat stress occurs in the plant it disturbs the growth of plants and physiological process like, photosynthesis process slow down, increase in transpiration rate and closing stomata. In recent research we concluded that heat stress effects photosynthesis process, transpiration rate, stomatal conductance chlorophyll contents, and water use efficiency in heat tolerance genotype (Safoora F1 Hybrid) and heat sensitive genotypes (Cucumber Desi). Safoora F1-Hybrid showed the maximum performance while Cucumber Desi exhibited poor performance.

Under the temperature stress, leaf area decreased, and improvement of the transpiration rate rapidly declined. It was recognized that growth achieved under limited usage of resources in osmolytes production, supports plants in attaining determined production under high stress (Serraj & Sinclair, 2002; Alves & Setter, 2004). Relative water content is one of the basic techniques to evaluate drought response in plants. Drought affects the chlorophyll contents of the plants in several ways, but the closing of stomata is the most significant reason for the reduction in the chlorophyll contents (Kawasaki et al., 2001). Under high temperature declined the translocation and assimilation of the photosynthetic process (Demiral et al., 2005). Dichio et al. (2006) described that osmotic regulation of the plants through decline osmotic potential. These osmotic potential supporting plants to survive under drought salinity and heat stress. These osmolytes are such as proline, aspartic acid, glutamic acid, glycine betaine and alanine betaine that protect the plant form abiotic stress (Sakamoto & Murata, 2002).

According to the study of Bajji et al. (2002), the rapid increase in osmotic potential may cause by electrolyte break out from cells as membrane variability is important in adjoining or surrounding osmolytes. *In vitro* study of other plant cultures demonstrated the reduced content of chlorophyll in case of increased stress rate of drought (Molasiotis et al., 2006). The estimate of the strength and steadiness in chlorophyll rate are discovered responsible for the certain and rapid method which is used to assess the tolerance capacity of the plant under certain drought stress (Perceival & Sherif, 2002). Biochemical parameter like proline contents is affected in temperature stress. Proline accumulation in roots and shoots increased in all genotypes of cucumber at vegetative and reproductive phases. Safoora F1-Hybrid showed maximum performance for the accumulation of proline (roots and shoots). Cucumber Desi showed the lowest performance. Plants under saline stress, drought and heat stress adopt diverse contraptions to regulate the osmotic adjustment. It was also concluded from the results of Ramanjulu & Sudhakar (2001). They described that osmotic modification through accumulating of compatible solutes such as Proline, Glycine-betaine, and Polyols under heat stress. Alike result also obtained from Yang et al. (2003) they proved that proline accumulation is recognized as the alteration of salinity and Glycine-betaine. These are the most abundant quaternary ammonium developments developed in higher plants due to salinity.

It was screen out from present study that heat tolerant genotype (Safoora F1 Hybrid) protection against heat stress was more than heat sensitive genotypes (Cucumber Desi). Some compounds protect the heat tolerant plants from different stresses by osmotic adjustment, detoxification of ROS, support of tissue veracity, the stability of amino acid and enzymes (Bohnert & Jensen, 1996). Rhodes and Hanson, (1993) informed that proline, polyols, trehalose, sucrose, glycinebetaine, alaninebetaine, proline betaine, choline O-sulphate, hydroxyl proline betaine, and pipecolate betaine are low molecular weight, highly soluble that is non-toxic in higher ratio at the cellular level under high temperature stress. Ramanjulu and Sudhakar, (2001) specified that under salinity, pea plants regress multifarious mechanisms consenting for adjustment to osmotic and ionic stress. Such scheme makeup osmotic adaptation accumulation of well-suited solutes, for example polyol, proline and glycinebetaine,

According to Verbrugen and Hermans (2008), plant yield, growth and buildup of proline are negatively affected by drought stress among other environmental stresses. Practically this accumulation happens during stress condition in all plants and there are also records of high buildup of proline in tissues. Surely their water content found low, for example inflorescence and seed (Khan et al., 2020). When plant is exposed to conditions full of stress, their primary response is usually buildup of proline to avoid cell injury (Ashraf & Foolad, 2007). Between such functions, relative capability as an osmolytes, proteins (stabilizing) regulation of cystolic pH and free radical scavenging of hydroxyl radicals was also recorded (Hong et al., 2000). Process of Osmoregulation via proline collection of proline and inorganic ions has been proposed as possible profit for protection from stress caused by water in *in vitro* plant (woody) cultures (Brito et al., 2003). More buildup of proline was found in tolerant plants towards drought stress as compared to those which were sensitive (Verbruggen & Herman, 2008). The study of Bor et al. (2003) demonstrated that wild plant species which were tolerant to drought contained more proline than sensitive ones. Moreover, drought tolerance was improved through increased proline contents by the role of transgenic methods (Hong et al., 2000; Roosens et al., 2002).

Rafiq et al. (2013) showed that raised level of temperature has potential to reduce the biological, physical and morphological characteristics and functioning including development of plant and its enlargement that ultimately lessens the yield. Biogenetic actions, molecular settlement and exogenic use of osmo-protectants such as proline are used to raise tolerance levels in plants against heat stress. They determined that the result of high temperature influenced the flower wilting, dryness of anthers, flower and fruit development. Ahmad et al. (2013) informed that stress tolerance ability of could be recognized through the growing of plants under environmental stresses. It was concluded that Safoora F_1-Hybrid and Fazal F_1-Hybrid can be regarded as heat tolerant among ten genotypes. These genotypes revealed as heat tolerant, while Local Khushab Green, Summer Green, Bush Green, C-200, C-400 and Cucumber Desi were considered as heat sensitive cucumber genotypes.

Conclusion

In current study, it was concluded that Safoora F_1 Hybrid and Fazal F_1-Hybrid Cucumber can be categories as heat tolerant, while Khushab Local, Summer Green, Bush Green, C-200, C-400 and Cucumber Desi were regarded as heat sensitive cucumber genotypes at seedling stage.

Conflict of Interest: The authors declare that they have no conflict of interest.

References

Abdelmageeda, A. H., Nazim, G., & Bernd, G. (2003). Effect of High Temperature and Heat Shock on Tomato (*Lycopersicon esculentum* Mill.) Genotypes under Controlled Conditions. *Journal of the American Society for Horticultural Science, 116*(6), 1113-1116.

Ahmad, M., Hassan, F., Aslam, M., & Aslam, M. (2013). Physiological attributes based resilience of wheat to climate change. *International Journal of Agriculture and Biology, 14*(3), 407-412.

Ali, A. H., Abdelrahman, M., Radwan, U., El-Zayat, S., & El-Sayed, M. A. (2018). Effect of Thermomyces fungal endophyte isolated from extreme hot desert- adapted plant on heat stress tolerance of cucumber. *Applied Soil Ecology*, *124*, 155–162.

Ali, S., Shah, A. H., Gul, R., Ahmad, H., Nangyal, H., & Sherwani, S. K. (2021). Morpho-agronomic characterization of okra (*Abelmuscus esculentus* L.). *World Applied Science Journal, 31*(3), 2764-2769.

Alves, A. A., & Setter, T. L. (2004). Response of cassava leaf area expansion to water deficit: Cell proliferation, cell expansion and delayed development. *Annals of Botany*, *94*(4), 605-613.

Ammar, A. T., & Arif, A. M. (2019). Phytochemical screening and in vitro antibacterial and anticancer activities of the aqueous extract of Cucumis sativus. *Biological Sciences*, *26*(3), 600-604.

Ajayeoba, T. A., Atanda, O. O., Obadina, A. O., Bankole, M. O., & Adelowo, O. O. (2015). The incidence and distribution of Listeria monocytogenes in ready- to- eat vegetables in South- Western Nigeria. *Food Science & Nutrition*, *4*(1), 59–66. https://doi.org/10.1002/fsn3.263.

Ashraf, M., & Foolad, M. (2007). Roles of glycine betaine and proline in improving plant abiotic stress resistance. *Environmental and Experimental Botany*, *59*(2), 206-216.

Bajji, M., Kinet, J. M., & Lutts, S. (2002). The use of the electrolyte leakage method for assessing cell membrane stability as a water stress tolerance test in durum wheat. *Plant Growth Regulation, 36*(1), 61-70.

Bita, C., & Gerats, T. (2013). Plant tolerance to high temperature in a changing environment: scientific fundamentals and production of heat stress-tolerant crops. *Frontiers in Plant Science*, *4*, 273.

Bates, L. S., Waldorna, R. P., & Teare, I. D. (1973). Rapid determination of free proline for water stress studies. *Plant Soil*, *39*, 205–208.

Bohnert, H. J., & Jensen, R. G. (1996). Strategies for engineering water-stress tolerance in plants. *Trends in Biotechnology, 14*(3), 89-97.

Bor, M., Özdemir, F., & Türkan, I. (2003). The effect of salt stress on lipid peroxidation and antioxidants in leaves of sugar beet *Beta vulgaris* L. and wild beet *Beta maritima* L. *Plant Science*, *164*(1), 77-84.

Brito, G., Costa, A., Fonseca, H. M., & Santos, C. V. (2003). Response of Olea europaea ssp. maderensis *in vitro* shoots exposed to osmotic stress. *Scientia Horticulturae*, *97*(3), 411-417.

Dekov, I., Tsonev, T., & Yordanov, I. (2001). Effects of water stress and high-temperature stress on the structure and activity of photosynthetic apparatus of *Zea mays* and *Helianthus annuus*. *Photosynthetica, 38*(3), 361-366.

Demiral, M. A., Aydin, M., & Yorulmaz, A. (2005). Effect of salinity on growth chemical composition and antioxidative enzyme activity of two malting barley (*Hordeum vulgare* L.) cultivars. *Turkish Journal of Biology, 29*(2), 117-123.

Dichio, B., Xiloyannis, C., Sofo, A., & Montanaro, G. (2006). Osmotic regulation in leaves and roots of olive trees during a water deficit and re-watering. *Tree Physiology*, *26*(2), 179-85.

Giaveno, C., & Ferrero, J. (2003). Introduction of tropical maize genotypes to increase silage production in the central area of Santa Fe, Argentina. *Crop Breed Application Biotechnology, 3*(2), 89-94.

GOP (Government of Pakistan). 2018-19. Agricultural Statistics Year Book 2018-19. Ministry of National Food Security and Research, Islamabad, Pakistan.

Grubben, G. J. H., & Denton, O. A. (2004). Plant Resources of tropical Africa II: Vegetables, Leiden, Wageningen: Backhuys Publishers.

Hameed, R., Ali, M., & Murtaza, A. (2018). Holistic benefits of cucumber. Agrinfo Bank Pakistan. Retrieved from http://agrinfobank.com.pk/holistic-benefits-of-cucumber/.

Hong, Z., Lakkineni, K., Zhang, Z., & Verma, D. P. S. (2000). Removal of feedback inhibition of Δ1-pyrroline-5-carboxylate synthetase results in increased proline accumulation and protection of plants from osmotic stress. *Plant Physiology, 122*(4), 1129-1136.

Iwaya Inoue, M., Matsui, R., & Fukuyama, M. (2004). Cold- or heat-tolerance of leaves and roots in perennial ryegrass [*Lolium perenne*] determined by ^{1}H-NMR. *Plant Production Science, 7*(2), 118-128.

Joyce, S. M., Cassells, A. C., & Jain, S. M. (2003). Stress and aberrant phenotypes in vitro culture. *Plant Cell, Tissue and Organ Culture, 74*(2), 103-121.

Khadija, F., Fatima, B., Usman, M. & Khan M. S. (2024). Floral bud size and culture conditions' effect on embryogenesis in anther-derived calli of cucumber. *SABRAO Journal of Breeding and Genetics*, *56*(5), 2079-2089. http://doi.org/10.54910/sabrao2024.56.5.30.

Khadija, F., Fatima, B., & Usman, M. (2022). Evaluation of cucumber germplasm for yield and its contributing attributes. *Journal of Agriculture and Food, 3*(1), 1-9.

Kawasaki, S., Borchert, C., Deyholos, M., Wang, H., Brazille, S., Kawai, K., & Bohnert, H. J. (2001). Gene expression profiles during the initial phase of salt stress in rice. *The Plant Cell*, *13*(4), 889-905.

Khater, E.S.G. (2017). Effect of acclimatization temperature and light intensity on the graft-take of cucumber seedlings. *Journal of Environmental and Analytical Toxicology*, *7*, 1-14.

Kim, J. R., Jung, S. H., Regan, J. M. & Logan, B. E. (2007). Electricity generation and microbial community analysis of alcohol powered microbial fuel cells. *Bio-resource Technology*, *98*(13), 2568-2577.

Khan, M. A., Ungar, I. A., & Showalters, A. M. (2000). Effects of salinity on growth, water relations and ion accumulation of the subtropical perennial halophyte, Atriplex griffithii var. stocksii. *Annals of Botany*, *85*, 225–232.

Marschner, H. (1995). Adaptation of plants to adverse chemical soil conditions. *Mineral Nutrition of Higher Plants*, *2*, 596-680.

Molassiotis, A., Sotiropoulos, T., Tanou, G., Kofidis, G., Diamantidis, G., & Therios, E. (2006). Antioxidant and anatomical responses in shoot culture of the apple rootstock MM 106 treated with NaCl, KCl, mannitol or sorbitol. *Biologia Plantarum, 50*(1), 61-68.

Percival, G. C., & Sheriffs, C. N. (2002). Identification of drought-tolerant woody perennials using chlorophyll fluorescence. *Journal of Arboriculture*, *28*(5), 215-223

Porter, J. R. & Gawith, M. (1999). Temperatures and the growth and development of wheat: a review. *European Journal of Agronomy*, *10*(1), 23-36.

Prasad, P. V., Boote, K. J. & Allen, L. H. (2006). Adverse high temperature effects on pollen viability, seed-set, seed yield and harvest index of grain-sorghum [*Sorghum bicolor* (L.) Moench] are more severe at elevated carbon dioxide due to higher tissue temperatures. *Agricultural and Forest Meteorology*, *139*(3), 237-251.

Rafiq, A., Muhammad, S. I., Danish, I., Talat, M. M., Saqib, N., & Muhammad, K. A. (2013). Review on heat stress response in different genotypes of tomato crop (*Lycopersicon solanum*). *International Journal of Modern Agriculture*, *2*(2), 203–210.

Ramakrishna, A., & Gokare, A. R. (2011). Influence of abiotic stress signals on secondary metabolites in plants. *Plant Signal Behavior, 6*(11), 1720-1731.

Ramanjulu, S., & Sudhakar, C. (2001). Alleviation of NaCl salinity stress by calcium is partly related to the increased proline accumulation in mulberry (*Morus alba*L.) callus. *Journal of Plant Biology-New Delhi, 28*(2), 203-206.

Rhodes, D., & Hanson, A. (1993). Quaternary ammonium and tertiary sulfonium compounds in higher plants. *Annual Review of Plant Biology, 44*(1), 357-384.

Roosens, N. H., Al Bitar, F., Loenders, K., Angenon, G., & Jacobs, M. (2002). Overexpression of ornithine-δ-aminotransferase increases proline biosynthesis and confers osmotolerance in transgenic plants. *Molecular Breeding*, *9*(2), 73-80.

Sakamoto, A., & Murata, N. (2002). The role of glycine betaine in the protection of plants from stress: clues from transgenic plants. *Plant, Cell & Environment*, *25*(2), 163171.

Serraj, R. & Sinclair, T. (2002). Osmolytes accumulation: can it really help increase crop yield under drought conditions? *Plant, Cell & Environment, 25*(2), 333-341.

Shah, N., & Paulsen, G. (2003). Interaction of drought and high temperature on photosynthesis and grain-filling of wheat. *Plant,* 257, 219–226.

Shahid, M. A., Pervez, M. A., Balal, R. M., Ahmad, R., Ayyub, C. M., Abbas, T., & Akhtar, N. (2011). Salt stress effects on some morphological and physiological characteristics of okra (*Abelmoschus esculentus* L.). *Soil and Environment, 30*(1), 66-73.

Siddique, S., Ayub, G., Nawaz, Z., Zeb, S., Khan, F. S., Ahmad, N., & Rauf, K. (2017). Enhancement of growth and productivity of cucumber (*Cucumis sativus*) through foliar application of calcium and magnesium. *Pure and Applied Biology, 6*(2), 402–411.

Steel, R. G. D., Torrie, J. H. & Dickey, D. A. (1997). Principle and Procedures of Statistics. A Biometrical Approach. 3rd ed. *McGraw Hill Book Co. NY, US*: 352358.

Tantawy, A. S., A-Mawgoud, A. M. R., El-Nemr, M. A., & Chamoun, Y. G. (2009). Alleviation of salinity effects on tomato plants by application of amino acids and growth regulators. *European Journal of Science Research, 30*, 848-894.

Thuzar, M., Puteh, A., Abdullah, N. A. P., Lassim, M. M., & Jusoff, K. (2010). The effects of temperature stress on the quality and yield of soya bean [(*Glycine max* L.) Merrill.]. *Journal of Agricultural Science, 2*(1), 172-179.

Tsukaguchi, T., Kawamitsu, Y., Takeda, H., Suzuki, K., & Egawa, Y. (2003). Water status of flower buds and leaves as affected by high temperature in heat-tolerant and heat sensitive cultivars of snap bean (*Phaseolus vulgaris* L.). *Plant Production Science, 6*(1), 24-27.

Verbruggen, N., & Hermans, C. (2008). Proline accumulation in plants: a review. Amino acids, 35(4), 753-759.

Wahid, A., & Close, T. (2007). Expression of dehydrins under heat stress and their relationship with water relations of sugarcane leaves. *Biologia Plantarum, 51*(1), 104-109.

Wolf, B. (1982). The comprehensive system of leaf analysis and its use for diagnosing crop nutrient status. *Communications in Soil Science and Plant Analysis, 3*, 1035-1059.

Wu, Y., & Cosgrove, D. J. (2000). Adaptation of roots to low water potentials by changes in cell wall extensibility and cell wall proteins. *Journal of Experimental Botany ,51*, 1543-1553.

Yamane, H., Tao, R., Murayama, H., & Sugiura, A. (2000). Determining the S-genotypes of several sweet cherry cultivars based on PCR-RFLP analysis. *The Journal of Horticultural Science and Biotechnology, 75*(5), 562-567.

Yang, W.J., Rich, P. J., Axtell, J. D., Wood, K. V., Bonham, C. C., Ejeta, G., & Rhodes, D. (2003). Genotypic variation for glycine betaine in sorghum. *Crop Science, 43*(1), 162-169.

Yilmaz, S. A., Shahatha, H. A., Hagop, P. G. & Al-Rawi, F. I. (2004). Effect of salinity on the viability of rice seeds. *Plant Growth, Drought and Salinity in the Arab Region*, 235- 244.

Zhang, L. P., Mehta, S. K., Liu, Z. P., & Yang, Z. M. (2008). Copper induced proline synthesis is associated with nitric oxide generation in Chlamydomonas reinhardtii. *Plant and Cell Physiology, 49,* 411-419.

Zhang, J., Li, D. M., Gao, Y., Yu, B., Xia, C. X., & Bai, J. G. (2012). Pretreatment with 5- aminolevulinic acid mitigates heat stress of cucumber leaves. *Biologia Plantarum. 56*, 780-784.