

The different responses of rice genotypes to heat stress associated with morphological, chlorophyll and yield characteristics

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Abstract

Sixteen rice genotypes were planted under normal and high temperature during 2018 and 2019 seasons to understand the mechanisms that make plants tolerant or susceptible to heat stress and methods which may lead to generate new varieties with sustainable yield production. The combined analysis showed significant differences at the level of probability 0.05 and 0.01 among years for all characters except, chlorophyll content and leaf rolling which would indicate wide differences among the weather of both years. Environment's variance was significant at the level of probability 0.01 over all characteristics. The significant differences at the level of probability 0.01 were recorded among genotypes and G x E interaction variances for all characteristics. The highest values of panicles number plant⁻¹ were obtained from 'Giza 178' and 'Hybrid 2', however, the highest values of 100-grain weight were recorded with 'Giza 179' and 'Egyptian Yasmine'. The minimum values of sterility% were recorded with 'Sakha 107' and 'IET1444'. Concerning the cultivars performance across two environments, the cultivars 'Giza 178', 'Giza 179', 'Sakha 107' and 'IET 1444' gave the best desirable values over natural and heat stress so, those cultivars are considered to play a vital role in breeding program to enhance heat stresses tolerance accompanied with high yield potential.

Keywords: chlorophyll; grain yield; heat stress; G x E interaction; rice

Introduction

The increasing temperatures linked with global warming may have serious direct and indirect consequences on crop yield, especially in cereals. There are many environmental stress factors affect the crops such as heat (EL Sabagh *et al.*, 2019), salinity (Hafez *et al.*, 2020), drought (Abdelaal *et al.*, 2020), chilling (Elkelish *et al.*, 2020). Heat stress has become important factor affecting crop growth and yield. Moreover, different global circulation models predict that greenhouse gases will gradually increase the average ambient

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temperature. According to a report of the Intergovernmental Panel on Climate Change (IPCC, 2004), global mean temperature will rise 0.3 °C per decade (Jones *et al.*, 1999), reaching approximately 1 and 3 °C above the present value by years 2025 and 2100, respectively. In addition, the climate is expected to be more variable, with frequent episodes of stressful temperatures during the crop-growing season. As a result, crop production may be severely affected by an increase in mean global temperature. The yield decrease is a result of heat stress alert in the physiological, morphological, biochemical and molecular changes (Wang *et al.*, 2003). Morphologically, heat stress decreases the duration of developmental phases leading to fewer and smaller organs, lower light perception due to a reduced life cycle (Saeed *et al.*, 2017). Photosynthesis, respiration and the partitioning of assimilates to different organs within the plant are also negatively affected (Elkelish *et al.*, 2020).

Rice is a staple food for more than half the world's population, so many studies were conducted to improve rice grain yield (Abou Khadrah *et al.*, 2020). Abiotic stress such as extreme temperatures and salinity frequently limits the major crop species' growth and productivity (Elkelish *et al.*, 2020; Hafez *et al.*, 2020). Rice is a very sensitive crop to heat stress, the critical temperature for spikelet fertility in rice is a maximum of 35 °C and moreover, sustained high temperature for more than a week cause severe damage (Melo *et al.*, 2021). As an important cereal crop, rice productivity might decrease globally by about 3.2% per each degree-Celsius increase in global mean temperature (Zhao *et al.*, 2017). As the rice crop is concerned, the optimum temperature is 33 °C (Jagadish *et al.*, 2007). Depending on the specific stage of rice development, heat stress reduces number of tillers/panicles, decreases grain number/plant and lower grain weight, thus negatively impacting yield formation, improving rice crop tolerance to heat stress in terms of sustaining yield stability under high day temperature, high night temperature, or combined high day and night temperature will bolster future food security (Xu *et al.*, 2020). Therefore, high temperatures over 35 °C encourage spikelets sterility and unfilled grain (Ghazy, 2012). Moreover, high temperature affects rice plant growth, for instance, during seed germination, high temperature may slow down or totally inhibit germination, depending on plant species and the intensity of the stress (Yoshida, 1981). Also, high temperature over 35 °C may harmfully affect photosynthesis, water relations, membrane stability and primary and secondary metabolites (Yoshida, 1981; Naveed *et al.*, 2020). Moreover, throughout plant ontogeny, enhanced the expression of heat shock proteins, other stress-related proteins, and reactive oxygen species accumulation (Wahid *et al.*, 2007). Plant tolerance to heat can be divided into escape (successful reproduction before the onset of severe stress), avoidance (maintenance of a high plant water status during stress e.g. caused by stomatal closure, reduced leaf area, senescence of older leaves, etc. or by increased root growth) and tolerance (the maintenance of plant function at limited water availability and/or the recovery of plant water status and plant function after stress) may involve osmotic adjustments, but may also be the result of rigid cell walls or small cells (Kondamudi *et al.*, 2012). The heat stress response depends on plant species and genotypes (Wahid *et al.*, 2007), so very important to identify the tolerant genotypes based on their morphological and physiological characters under heat conditions. Heat stress dominantly reduced fertility when drought and heat stress co-occurred and the pollen count on the stigma reduced in Moroberekan, therefore, not all drought-tolerant cultivars are tolerant to heat stress by default at sensitive stages however, heat stress seems to be the dominant stress at reproductive stage (Costa *et al.*, 2021).

The phenotypic correlation among some heat morphological characteristics and their contribution to grain yield under heat conditions were directly or indirectly expected in rice (Barnabás *et al.*, 2008). The thousand-grain weight and spikelet fertility showed a positive and significant at probability level 0.05 association with yield plant⁻¹ under heat conditions at the genotypic level. Paddy yield had a strong correlation with grain number panicle⁻¹, days to maturity and 1000-grain weight (Gaballah and Abu El-Ezz, 2019). The effect of heat stress at the seedling and tillering stages of rice led to no panicle development in the cultivars (Azameti *et al.*, 2021). The objective of this study was to evaluate the impact of high temperatures stress on yield and related parameters of rice, to identify rice genotypes tolerance for heat stress, estimate the correlation coefficient based on agro-morphological characteristics and applicability of stress index under high heat stress conditions.

Materials and Methods

Experiment preparation

Sixteen rice genotypes (Table 1) were used in the study. The experiments were conducted under natural (mild temperatures) conditions at Sakha Agricultural Research Station and high heat stress conditions at Elkharga Agricultural Research station in 2018 and 2019 successive seasons. The average temperature and humidity percentage in 2018 and 2019 years were illustrated at Table 2 according to Sakha and El-Kharga Agro- Meteorological station. The nursery was sown on the 25th and 26th of April in the 2018 and 2019 seasons, respectively, and transplanted to the field after 30 days in both locations. Each genotypes seedling was transplanted to 5 rows along 5 m, 20 cm apart between rows, with a distance of 20 cm between the hills within a row, the plot size (1.20 m x 5 m). The design of the experiment was a randomized complete block design with three replicates.

Table 1. The pedigree, salience and feature of rice genotypes used in the study

Genotype	Pedigree	Salience and feature
'Giza 177'	Giza 171/Yomji No.1//Pi No.4	Japonica type - sensitive to heat – short stature, early duration- resistance to blast
'Giza 178'	Giza175/ Milyang 49	Indica/Japonica type, medium maturing, semi-dwarf, resistant to blast, medium grain, tolerant to heat and high yield
'Giza179'	GZ6296 / GZ1368	Indica/Japonica type - moderate to heat – short stature-early duration- resistant to blast
'Giza182'	Giza181/IR39422-161-1-3-1/Giza181	Indica type -sensitive to heat- short stature-early duration- resistant to blast
'Sakha101'	Giza 176/ Milyang	Japonica type - sensitive to heat – short stature- moderate duration- sensitive to blast
'Sakha 102'	GZ4096-7-1/GZ4120-2-5-2 (Giza 177)	Japonica type - sensitive to heat – short stature, early duration- resistance to blast
'Sakha 103'	Giza177/Suweon349	Japonica type - sensitive to heat – short stature- early duration- resistance to blast
'Sakha 104'	GZ4096-8-1/GZ4100-9-1	Japonica type - sensitive to heat – long stature, moderate duration- sensitive to blast
'Sakha 105'	GZ5581-46-3/GZ4316-7-1-1	Japonica type - sensitive to heat – short stature- early duration- resistant to blast
'Sakha 106'	Giza176 / Milyang79	Japonica type - sensitive to heat – moderate stature- early duration- resistant to blast
'Sakha 107'	Giza 177 /BLI	Japonica type - tolerant to heat – short stature- early duration- resistant to blast
'Sakha 108'	Sakha 101 / HR 1315824	Japonica type- moderate to heat – short stature- moderate duration- resistant blast
'Hybrid 2'	IR6962SA/Giza179	Indica type- moderate to heat – short stature- moderate duration- resistant to blast
'E. Yasmine'	Introduction	Indica type- moderate to heat – short stature- late duration- resistant to blast
'GZ1368-S-5-4'	IR 1615-31 / BG 94-2349	Indica type- moderate to heat – short stature -moderate duration- resistant to blast
'IET1444'	TN 1 / CO 29	Indica type- tolerant to heat – short stature -moderate duration- resistant to blast

Table 2. Maximum (Max.) and minimum (Min.) air temperature and relative humidity percentage for 2018 and 2019 growing seasons

	Month	Date	2018			2019		
			Max.	Min.	RH %	Max.	Min.	RH %
Sakha- Kafr EL-Sheikh	May	10-Jan	29.07	17	63.49	29.4	17.2	64.1
		20-Nov	29.96	18	61.4	30.3	18.4	62
		20-30	31.05	20.5	58.92	31.4	20.7	59.5
	June	10-Jan	29.96	19.8	51.78	30.3	20.2	52.3
		20-Nov	31.05	21.8	66.3	31.4	22.2	66.4
		20-30	29.46	21.41	63.5	29.8	21.8	63.6
	July	10-Jan	31.05	21.41	68.2	31.4	21.8	68.3
		20-Nov	33.13	22	73.1	33.5	22	73.2
		20-30	35.61	24.6	65.1	36	24.6	65.2
	August	10-Jan	35.9	26.2	67.55	36	26.3	68.2
		20-Nov	35.6	25.5	66.07	35.7	25.6	66.7
		20-30	33.9	23.3	68.25	34	23.3	68.9
	September	10-Jan	35.1	23.3	65.57	35.2	23.3	66.2
		20-Nov	34.3	23.8	66.6	34.4	23.8	66.7
		20-30	34.5	24.3	61.3	34.6	24.3	61.4
EL-Kharga - New Valley	May	10-Jan	37	21.2	26.2	36.4	21	26.3
		20-Nov	37.3	20.5	25.1	37.4	20.5	25.2
		20-30	39.2	23.8	24.1	40.4	23.8	24.1
	June	10-Jan	39.9	25.8	22.3	40	25.9	22.3
		20-Nov	38.9	24.6	26.2	39	24.6	26.3
		20-30	38.7	23.5	27.1	38.8	23.5	27.2
	July	10-Jan	38.5	23.7	28.2	38.6	23.7	28.3
		20-Nov	40.6	23.6	27.1	40.7	23.6	27.2
		20-30	43.2	25.1	21.1	43.3	25.2	21.1
	August	10-Jan	45.2	29.1	22.2	45.3	29.2	22.2
		20-Nov	44.1	30.4	26.1	44.2	30.5	26.2
		20-30	39.9	26.7	30.9	40	26.8	31
	September	10-Jan	40.5	25.4	31.1	40.6	25.5	31.2
		20-Nov	41.1	25.8	31.2	41.2	25.9	31.3
		20-30	41.5	25.8	27.1	41.6	25.9	27.2

The soil properties in the two locations are presented in Table 3. The soil samples of 0-15 cm depth were collected from selected plots to determine the chemical and physical properties. The soil samples were air-dried, crushed and passed through 2 mm sieve to analyse organic carbon and total N content. Soil organic carbon was analysed by Walkley-Black method (Allison, 1965). One g air-dried sample (passed through 0.5 mm sieve) was taken in a 500 ml Erlenmeyer flask. Ten ml 1N $K_2Cr_2O_7$ and 20 ml conc. H_2SO_4 was added to the flask and react for 30 minutes after which 200 ml distilled water and 10 ml H_3PO_4 was added. One ml orthophenanthroline indicator was added to the flask and was titrated with about 0.5N $FeSO_4$ solution. Blank titration was run to calculate the strength of the $FeSO_4$ solution. Total nitrogen was determined by Microkjeldahl instrument, 0.5 g air-dried soil sample was digested in 3 ml conc. H_2SO_4 in the presence of 0.5 g of digestion mixture (50: 10: 1 K_2SO_4 : $CuSO_4 \cdot 5H_2O$: metallic selenium). The digested sample was distillate with 40% NaOH and the distillate was collected in 4% boric acid containing three drops of mixed indicator (bromocresol green and methyl red), which was then titrated with 0.05 N H_2SO_4 . C: N ratio was calculated by dividing the results of organic carbon by total nitrogen (Bremner, 1965). All measurements of organic carbon and total nitrogen were done in triplicate. Soil pH in water was measured from a soil: water ratio 1: 2.5 using glass electrode method (Peech, 1965). Twenty gram of air-dried soil sample was taken in 100 ml of plastic bottle and 50 ml of distilled water was added. The suspension was stirred with a glass-rod at regular interval for 30 minutes. A glass electrode pH meter calibrated with buffer pH 7.0 and 4.0 and the pH of soil suspension was measured. The measurement was done in triplicate. Olsen-P was extracted with 0.5M sodium bicarbonate (pH 8.5) (Olsen *et al.*, 1954). P content in the extract was determined using ascorbic acid as reducing agent by a spectrophotometer. Available potassium (NH_4OAc -K) was extracted with neutral 1N ammonium acetate (Hanway and Heidel, 1952) and estimated by a flam photometer.

Phosphorus and potassium were applied in full dose (150 Kg P₂O₅ and 100 Kg K₂O, respectively) during land preparation in the permanent field and Nitrogen (150 Kg N₂O) fertilizer was applied in three splits as a top dressing. Insects and weeds were controlled periodically using pesticides and herbicides, respectively, as required.

Table 3. Physical and chemical properties of the soil in Sakha and Kharga research stations in 2018 and 2019 seasons

Soil physical and chemical properties	Sakha, Kafr El-Sheikh		Kharga, El-Wady El-Gaded	
	2018	2019	2018	2019
Clay%	55	55	3.56	3.56
Silt %	32.4	32.4	1.77	1.77
Sand %	12.6	12.6	94.65	94.65
Texture	Clayey	Clayey	Loamy Sandy	Loamy Sandy
Organic Matter	1.39	1.39	1.14	1.29
pH	8.1	8.2	8.8	8.7
Ec (Ds/m)	3.3	3.33	1.56	1.55
Total N (ppm)	512	518	235	252
Available P (ppm)	15.09	16.03	13.6	13.9
Co ³⁻	-	-	2	2
HCO ³⁻	5.55	5.56	1.5	1.5
Mg ⁺²	4.3	5	0.48	0.48
Na ⁺¹	1.88	1.69	0.035	0.032
K ⁺¹	16	16	0.23	0.27
Fe ⁺³	4.55	4.55	53.1	51.6
Mn ⁺²	3.1	3.5	1.3	1.4

The analyses of soil physical and chemical properties were done by laboratory of Rice Research and Training Center, Field Crop Research Institute.

Studied characteristics

The randomly selected plants (25 plants) were harvested manually at harvest, put into a Nylon mesh bag, sun-dried and threshed. Grain yield for each plot was recorded from a 25-hill sample, adjusted to a moisture content of 14% and expressed as g plant⁻¹. Days to 50% heading (DTH), were determined as the number of days from the date of sowing to the first panicle exertion date. SPAD-chlorophyll content (SPAD; CC) was determined using chlorophyll analytical apparatus (chlorophyll meter SPAD-502 Minolta camera Co. Ltd., Japan). Five flag leaves were measured from the widest part of the leaf of the main culms for each entry in all replications. Leaf rolling (LR) scores and symptoms for leaf rolling and heat resistance at the vegetative stage were done according to De Datta *et al.* (1988). Plant height (cm; PH) was measured (cm) from the soil surface to the tip of each plant's main panicle. Panicle length (cm; PL) for the main panicle was measured from panicle base up to a piculus of the panicle's uppermost spikelet. The number of panicles plant⁻¹ (NP) was determined by counting the number of panicles plant⁻¹ at harvesting. 100-grain weight (g; HGW) was recorded as the weight of 100 random chosen filled grains plant⁻¹. Sterility percentage was calculated by account number of unfilled grain panicle⁻¹ divided on number of spikelets panicle⁻¹. All results were recorded according to the standard evaluation system IRRI (IRRI, 2002).

Statistical analysis

Analysis of variance was conducted for all the characteristics using combined analysis according to Steel *et al.* (1997). Comparison among genotypes, genotypes and environments, years and environments were determined by Tukey's HSD test (P < 0.05). The correlation coefficient was carried out using the formula given

by Kown and Torrie (1964). Correlation among values of the different characteristics was performed based on Pearson's Product-Moment. The genetic analysis, phenotypic correlation and genetic variability were performed on combined data across seasons.

Results

The analysis of variance for studied characteristics was presented in Table 4. The environments, genotypes and genotypes \times environments variances were found to be statistically significant differences at the level of probability 0.01 for all characteristics. The days to heading, chlorophyll content, leaf rolling, plant height, panicle length, number of tillers and number of panicles plant⁻¹, 100-grain weight and sterility percentage as well as grain yield were exhibited not significant difference over the years.

Table 4. Analysis of variance for studied agronomic traits, years, environments and genotypes

Source of variance	Years (Y)	Environments (E)	E x Y	Blocks (EY)	Genotypes (G)	G x Y	G x E	G x E x Y	Pooled error	Total
df	1	1	1	8	15	15	15	15	120	191
Days to heading (day)	3.19 ^{ns}	19865.47 [*]	2.09 ^{ns}	2.48	278.58 ^{**}	1.587 ^{ns}	118.78 ^{**}	2.21 [*]	1.17	
Chlorophyll content (mg dS ⁻¹)	2.09 ^{ns}	4101.13 ^{**}	10.05 ^{ns}	2.57	35.67 ^{**}	0.58 ^{ns}	46.58 ^{**}	1.05 ^{ns}	1.06	
Leaf rolling	0.08 ^{ns}	963.02 [*]	0.08 ^{ns}	0.66	5.57 ^{**}	0.15 ^{ns}	5.82 ^{**}	0.150 ^{ns}	0.41	
Plant height (cm)	42.13 ^{ns}	17557.31 ^{**}	2.19 ^{ns}	0.57	183.52 ^{**}	1.39 ^{**}	157.05 ^{**}	2.29 ^{**}	0.56	
Panicle length (cm)	2.46 ^{ns}	810.76 [*]	1.07 ^{ns}	0.6	6.33 ^{**}	0.62 ^{ns}	20.03 ^{**}	0.21 ^{ns}	0.59	
Number of tillers plant ⁻¹	7.27 ^{ns}	2442.59 ^{**}	1.17 ^{ns}	1.2	11.22 ^{**}	0.28 ^{ns}	20.60 ^{**}	0.45 ^{ns}	0.87	
Number of panicles plant ⁻¹	4.15 ^{ns}	2647.74 ^{**}	1.56 ^{ns}	1.88	21.20 ^{**}	0.62 ^{ns}	16.77 ^{**}	1.19 ^{ns}	0.81	
100-grain weight (g)	0.02 ^{ns}	6.70 [*]	0.02 ^{ns}	0.001	0.57 ^{**}	0.01 ^{ns}	0.45 ^{**}	0.02 ^{ns}	0.01	
Sterility percentage (%)	1.85 ^{ns}	56123.33 ^{**}	21.16 ^{ns}	2.77	706.06 ^{**}	0.77 ^{ns}	1058.23 ^{**}	0.78 ^{ns}	0.91	
Grain yield plant ⁻¹ (g)	30.59 ^{ns}	185803.54 ^{**}	0.30 ^{ns}	2.24	227.03 ^{**}	1.05 ^{ns}	220.00 ^{**}	1.41 [*]	0.82	

(^{ns}) = Not Significant, (^{*}) = Significant at 0.05 and (^{**}) = Significant at 0.01 level of probability

The interaction between genotypes and Environments

Regarding the interaction in Table 5, the results indicated that, the shortest days to heading genotypes were observed in 'Sakha 102' under heat conditions while the longest days to heading genotypes were recorded with 'Egyptian Yasmine' and 'Sakha 101' under natural conditions. The highest values of chlorophyll content were found in 'Sakha 103' under natural conditions and the minimum values were found in 'Giza 177' under heat conditions. Concerning leaf rolling, the minimum mean values were achieved from 'Giza 178', 'Giza 179' and 'Sakha 107' under heat stress conditions, nevertheless, the highest values were obtained with 'Giza 177', 'Giza 182', 'Sakha 101', 'Sakha 102' and 'Sakha 103' under heat stress conditions. The highest panicles length was recorded with 'Egyptian Yasmine' under natural conditions and the minimum values were achieved with 'Sakha 105' under heat stress conditions.

The interaction between rice genotypes and environments for grain yield and its related characters were shown in Table 6. The maximum values of panicles number plant⁻¹ were achieved with 'Hybrid 2' under natural conditions; however, the minimum values were obtained with 'Sakha 103' under heat conditions. The heaviest 100-grain weight was recorded with 'Egyptian Yasmine' and 'Sakha 104' under natural conditions and the minimum grains weight was obtained with 'Giza 178', 'Sakha 103', 'Sakha 104' and 'Sakha 106' under heat stress. The minimum values of sterility percentage were recorded with 'Sakha 101', 'Sakha 103' and 'Sakha 107' under natural conditions, therefore the highest percentage was obtained from 'Sakha 106' under heat stress. Additionally, the highest mean values of grain yield plant⁻¹ were obtained from 'Hybrid 2' under natural conditions, however, the minimum values were obtained with 'Sakha 102' and 'Giza 177' under heat conditions.

Table 5. The interaction between rice genotypes and environments for growth characters over average 2018 and 2019 growing seasons

Genotypes	Days to heading (day)		Chlorophyll content (mg dS ⁻¹)		Leaf rolling
	Natural	Heat	Natural	Heat	Natural
'Giza 177'	93.00 hij	74.70 pq	43.92 bc	29.47 o	1.33 h
'Giza 178'	105.00 c	78.10 mn	44.64 b	33.26 i-l	1.67 h
'Giza179'	92.17 j	76.73 no	42.27 cd	32.26 k-n	1.33 h
'Giza182'	94.50 h	75.45 op	36.08 fg	33.26 i-l	1.33 h
'Sakha 101'	108.50 b	85.70 k	44.62 b	35.78 fgh	1.33 h
'Sakha 102'	94.50 h	73.47 q	43.49 bcd	33.35 i-l	1.67 h
'Sakha 103'	92.50 ij	76.82 no	46.55 a	32.48 j-n	1.33 h
'Sakha 104'	98.50 f	76.50 no	41.78 d	33.36 i-l	1.33 h
'Sakha 105'	94.00 hi	79.52 m	43.50 bcd	31.02 mn	1.33 h
'Sakha 106'	96.33 g	79.53 m	44.50 b	32.62 j-m	1.33 h
'Sakha 107'	94.33 h	77.40 n	43.34 bcd	34.14 h-k	1.33 h
'Sakha 108'	99.50 ef	77.58 n	42.19 cd	31.90 lmn	1.33 h
'Hybrid 2'	100.50 e	78.03 mn	42.85 bcd	30.76 n	1.67 h
'E. Yasmine'	119.50 a	79.73 m	34.96 ghi	32.25 k-n	1.67 h
'GZ 1368'	103.67 d	86.72 k	39.03 e	34.32 hij	1.33 h
'IET1444'	81.80 l	76.17 g	37.16 f	34.60 ghi	1.33 h
Genotypes	Leaf rolling	Plant height (cm)		Panicle length (cm)	
	Heat	Natural	Heat	Natural	Heat
'Giza 177'	7.17 ab	100.31 d	77.67 m	19.68 fg	16.51 jk
'Giza 178'	4.17 fg	101.20 d	81.63 k	23.17 b	18.03 hi
'Giza179'	4.50 fg	90.32 g	77.02 mn	20.28 ef	17.37 hij
'Giza182'	7.33 ab	101.27 d	76.00 n	19.50 fg	17.93 hij
'Sakha 101'	7.83 a	100.30 d	78.80 l	21.00 de	17.90 hij
'Sakha 102'	7.17 ab	109.50 a	88.60 hi	23.27 b	16.69 ijk
'Sakha 103'	8.00 a	105.00 c	78.72 l	22.28 bc	16.98 ijk
'Sakha 104'	5.17 ef	107.50 b	88.93 hi	20.30 ef	17.85 hij
'Sakha 105'	5.67 de	104.50 c	76.30 n	22.73 bc	14.73 l
'Sakha 106'	6.67 bc	105.50 c	82.05 jk	20.27 ef	17.79 hij
'Sakha 107'	4.33 fg	107.50 b	81.95 jk	21.17 de	18.42 gh
'Sakha 108'	6.33 bcd	96.85 f	79.08 l	21.82 cd	17.34 hij
'Hybrid 2'	5.50 de	97.68 ef	89.57 gh	23.12 b	15.84 k
'E. Yasmine'	5.83 cde	101.00 d	82.95 j	25.37 a	16.57 jk
'GZ 1368'	5.00 ef	88.28 i	88.23 i	19.50 fg	18.38 gh
'IET1444'	3.67 g	98.02 e	81.38 k	19.68 fg	19.50 fg

The same letters are not significantly different at the 5% probability level according to Tukey's test.

Table 6. The interaction between rice genotypes and environments for grain yield and its characters over average two seasons

Genotypes	Number of panicles plant ⁻¹		100-grain weight (g)	
	Natural	Heat	Natural	Heat
'Giza 177'	21.12 cde	12.33 mn	2.84 cd	2.25 k-n
'Giza 178'	22.63 bc	17.03 i	2.25 k-n	1.93 p
'Giza179'	20.27 ef	14.18 jk	2.61 efg	2.28 j-m
'Giza182'	17.50 hi	12.72 k-n	2.55 f-i	2.33 jkl
'Sakha 101'	22.50 bc	13.97 jkl	2.75 cde	2.42 h-k
'Sakha 102'	19.50 fg	14.25 jk	2.70 def	2.52 ghi
'Sakha 103'	18.62 gh	10.75 o	2.55 f-i	1.89 p
'Sakha 104'	22.00 bcd	14.50 j	2.89 abc	1.97 p
'Sakha 105'	21.17 cde	14.08 jkl	2.84 cd	2.13 mno
'Sakha 106'	20.73 def	13.57 j-m	2.78 cde	1.94 p
'Sakha 107'	19.57 gh	14.82 j	2.76 cde	2.44 hij
'Sakha 108'	23.05 b	12.12 mn	2.66 d-g	2.22 lmn
'Hybrid 2'	24.50 a	12.47 lmn	2.58 fgh	2.29 j-m
'E. Yasmine'	22.50 bc	11.35 no	3.00 a	2.40 ijk
'GZ 1368'	17.50 hi	12.80 k-n	2.11 no	2.05 k-n
'IET1444'	19.80 efg	13.63 j-m	2.34 jkl	2.01 op
Genotypes	Sterility percentage (%)		Grain yield plant ⁻¹ (g)	
	Natural	Heat	Natural	Heat
'Giza 177'	4.50 pq	63.73 c	40.50 e	13.63 r
'Giza 178'	7.50 n	29.05 h	45.55 b	30.58 ij
'Giza179'	5.50 op	34.67 g	44.50 b	26.21 k
'Giza182'	6.50 no	34.06 g	34.15 g	24.43 l
'Sakha 101'	3.50 q	33.74 g	45.52 b	17.55 p
'Sakha 102'	4.50 pq	43.34 e	42.00 cd	12.90 r
'Sakha 103'	3.50 q	72.07 b	37.18 f	19.74 o
'Sakha 104'	6.80 no	38.22 f	42.54 c	15.15 q
'Sakha 105'	5.50 op	36.99 f	41.00 fe	21.12 n
'Sakha 106'	4.50 pq	80.71 a	42.52 c	20.01 o
'Sakha 107'	3.50 q	24.93 i	42.53 c	31.89 h
'Sakha 108'	6.40 no	45.96 d	45.50 b	16.15 q
'Hybrid 2'	9.43 m	34.81 g	56.50 a	24.39 l
'E. Yasmine'	12.25 l	37.23 f	34.00 g	19.40 o
'GZ 1368'	11.43 l	28.69 h	31.50 hi	22.21 m
'IET1444'	16.15 k	22.15 j	30.17 j	25.71 k

The same letters are not significantly different at the 5% probability level according to Tukey's test

The reduction percentage caused by heat stress

Regarding days to heading as affected by heat stress (Figure 1), the 'E. Yasmine' was the most affected by high temperature conditions (33.28%) followed by 'Giza 178' (25.62%). Nonetheless, the minimum genotype which affected by high temperature was 'IET1444' (6.88%). 'Giza 177', 'Sakha 103' and 'Sakha 105' had the most decrease percentage by heat stress in chlorophyll content and most standing cultivars were 'IET1444' and 'E. Yasmine'.

'Sakha 103', 'Sakha 101' and 'Giza 182' had high score reduction in leaf rolling resulting by heat stress, while 'Giza 178' and 'IET1444' had minimum leaf rolling. The shortest stature cultivars as affected by heat stress 'Sakha 103', 'Sakha 105' and 'Giza 182' otherwise minimum genotypes 'GZ1368-s-5-1' and 'Hybrid 2'.

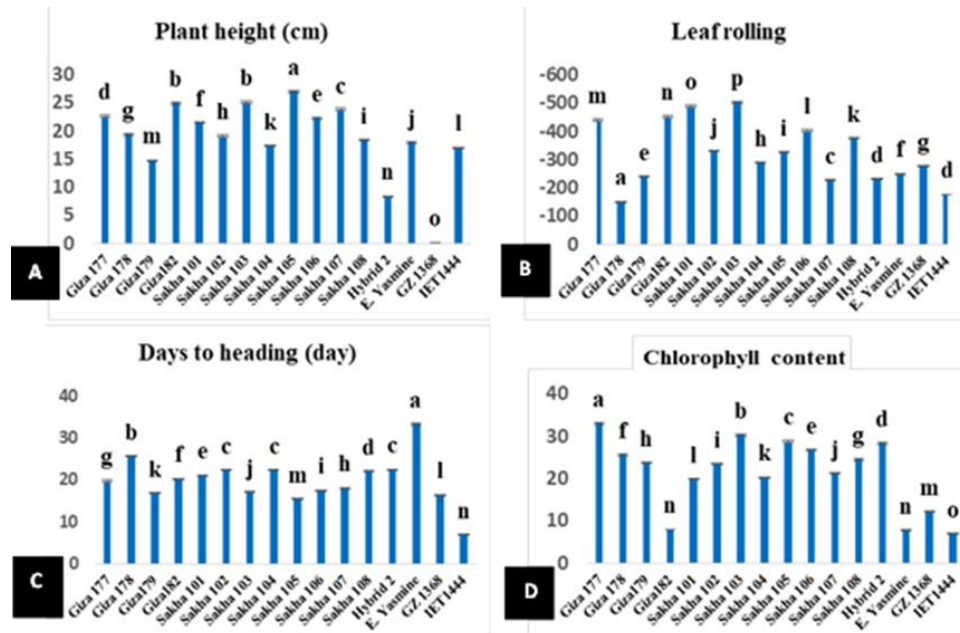


Figure 1. Effect of heat stress on reduction percentage in plant height (A), leaf rolling (B), days to heading (C) and chlorophyll content (D) in rice cultivars

The reduction percentage caused by the high temperature effect on grain yield and related traits were presented in Figure 2. Concerning panicle length, the genotypes with minimum reduction by heat stress were 'IET1444', 'GZ1368' and 'Giza 182', unlike 'Giza 182' and 'Sakha 103', which had minimum effect according to tillers number plant⁻¹. 'Sakha 107' and 'Giza 178' reduced the number of panicles plant⁻¹, while 'GZ1368' and 'Sakha 102' reduced 100-grain weight. The minimum reduction values concerning sterility percentage were achieved in 'IET1444', 'GZ1368' and 'E. Yasmine'. For grain yield plant⁻¹, the most standing genotypes under heat conditions with minimum reduction yield were 'IET1444' and 'Sakha 107' while 'Sakha 102', 'Giza 177', 'Sakha 108' and 'Sakha 104' the most cultivars sensitive to heat stress.

The interaction among genotypes, years and environments

The interaction among genotypes for days to heading and grain yield plant⁻¹ characteristics was presented in Table 7. For days to heading the genotypes 'Sakha 102' gave the shortest days to heading under heat conditions in 2019 growing season. Otherwise, the latest genotype was 'Egyptian Yasmine' at 2018 season under natural conditions. Regarding grain yield plant⁻¹, the highest values were recorded with 'Hybrid 2' under natural conditions at 2018 growing season, while the minimum values were recorded with 'Sakha 102' under stress at 2019 growing season.

Correlation coefficient

The correlation coefficient had different associated between pairs traits under natural and heat stress conditions. The days to heading had positive and significant at probability 0.01 correlation with leaf rolling panicle length, tillers number plant⁻¹ and panicles number plant⁻¹. Nonetheless, the days to heading under stress conditions had positive and significant (≥ 0.05) correlation with chlorophyll content while negative correlation was found with tillers number plant⁻¹ (Table 8).

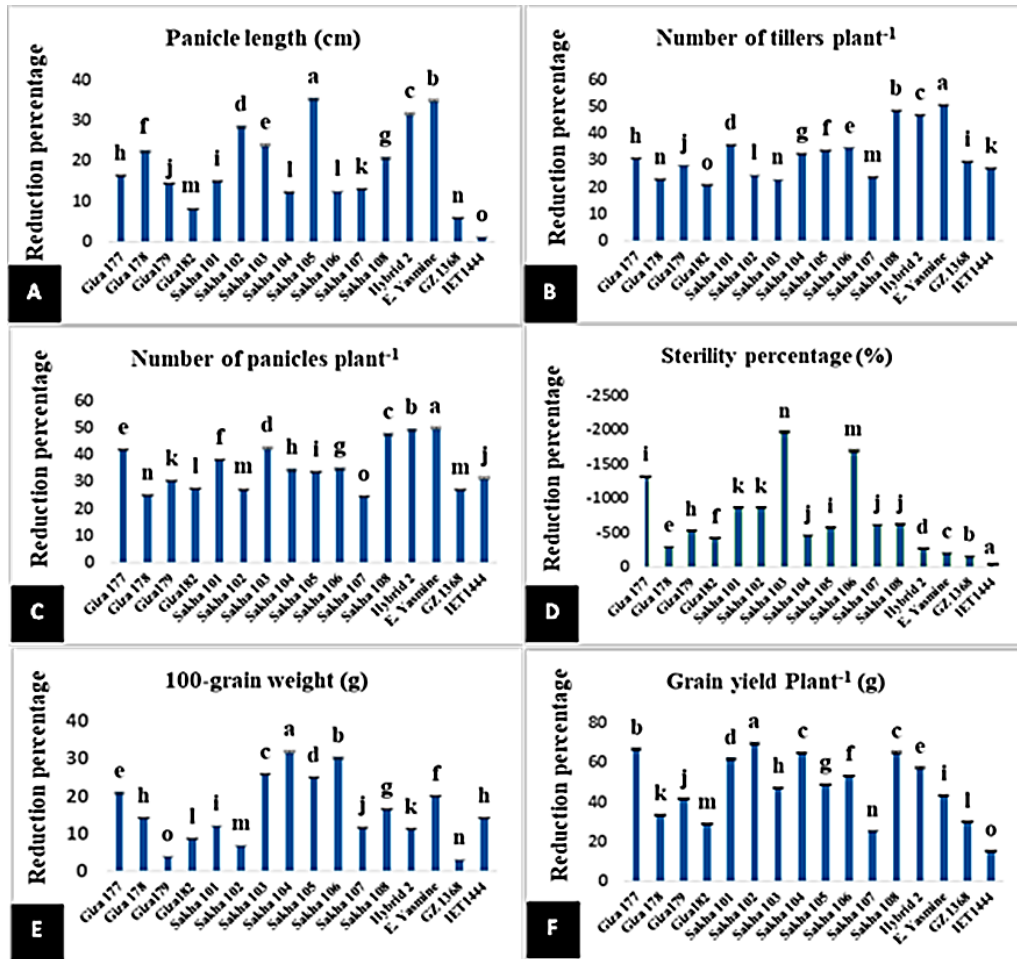


Figure 2. Effect of heat stress on reduction percentage in panicle length (A), number of tillers (B), number to panicles (C), sterility percentage (D), 100-grain weight (E) and grain yield plant⁻¹ (F) in rice cultivars

Table 7. The interaction between genotypes, years and environments on days to heading and grain yield characteristics

Genotypes	Days to heading (day)				Grain yield plant ⁻¹ (g)			
	2018		2019		2018		2019	
	Natural	Heat	Natural	Heat	Natural	Heat	Natural	Heat
'Giza 177'	94.00 g-j	75.13 t-w	92.00 ij	74.27 vw	41.00 ef	14.76 tu	40.00 f	12.51 v
'Giza 178'	105.67 c	76.97 qv	104.33 c	79.23 n-r	45.55 b	31.24 ijk	45.55 b	29.92 jk
'Giza 179'	93.00 hij	76.23 rv	91.33 j	77.23 p-u	45.00 bc	27.24 l	44.00 bcd	25.18 lm
'Giza 182'	95.00 f-i	74.40 uvw	94.00 g-j	76.50 r-v	34.15 h	25.77 lm	34.14 h	23.08 no
'Sakha 101'	109.00 b	84.30 l	108.00 b	87.10 k	46.02 b	18.66 r	45.01 bc	16.43 st
'Sakha 102'	95.00 f-i	73.53 w	94.00 g-j	73.40 w	43.00 cde	14.38 tu	41.00 ef	11.42 v
'Sakha 103'	93.00 hij	77.20 p-u	92.00 ij	76.43 r-v	37.19 g	20.21 pqr	37.18 g	19.28 qr
'Sakha 104'	99.33 de	77.50 p-t	97.67 ef	75.50 s-w	43.05 cde	16.11 stu	42.03 def	14.19 u
'Sakha 105'	94.00 g-j	80.57 mno	94.00 g-j	78.47 o-s	42.00 def	21.14 opq	40.00 f	21.11 opq
'Sakha 106'	96.67 fg	79.90 m-q	96.00 fgh	79.17 n-r	43.02 cde	20.02 pqr	42.02 def	20.01 pqr
'Sakha 107'	94.67 ghi	78.33 o-s	94.00 g-j	76.47 r-v	43.03 cde	31.70 ij	42.03 def	32.07 ij
'Sakha 108'	100.00 de	77.60 p-t	99.00 de	77.57 p-t	46.00 b	17.11 s	45.00 bc	15.20 stu
'Hybrid 2'	101.00 d	78.40 o-s	100.00 de	77.67 p-t	57.00 a	24.42 mn	56.00 a	24.36 mn
'E. Yasmine'	120.00 a	80.13 m-p	119.00 a	79.33 n-r	35.00 h	19.57 qr	33.00 hi	19.24 qr
'GZ 1368-S-5-4'	104.00 c	78.17 k	103.33 c	86.27 k	32.00 ij	22.21 op	31.00 ijk	22.22 op
'IET1444'	96.33 fg	82.27 m	96.00 fgh	81.33 mn	31.00 ijk	26.18 lm	29.33 k	25.24 lm

Table 8. Correlation coefficient among the studied characteristics

Natural	Days to heading (day)	Chlorophyll content	Leaf rolling	Plant height (cm)	Panicle length (cm)	Number of tillers plant ⁻¹	Number of panicles plant ⁻¹	100-grain weight (g)	Sterility percentage (%)
Chlorophyll content	-0.18								
Leaf rolling	0.48**	-0.08							
Plant height (cm)	-0.10	0.32*	0.14						
Panicle length (cm)	0.56**	0.05	0.79**	0.30					
Number of tillers plant ⁻¹	0.47**	0.15	0.44**	-0.04	0.47**				
Number of panicles plant ⁻¹	0.43**	0.22	0.44**	0.05	0.50**	0.97**			
100-grain weight (g)	0.21	0.09	-0.01	0.55**	0.29	0.36*	0.37*		
Sterility percentage (%)	0.08	-0.74**	0.24	-0.45**	0.04	0.04	0.04	-0.41*	
Grain yield plant ⁻¹ (g)	0.14	0.60*	0.32*	0.11	0.28	0.72**	0.72**	0.22	-0.44**
Heat	Days to heading (day)	Chlorophyll content	Leaf rolling	Plant height (cm)	Panicle length (cm)	Number of tillers plant ⁻¹	Number of panicles plant ⁻¹	100-grain weight (g)	Sterility percentage (%)
Chlorophyll content	0.43**	1.00							
Leaf rolling	-0.01	-0.15	1.00						
Plant height (cm)	0.10	0.17	-0.24	1.00					
Panicle length (cm)	0.13	0.74**	-0.35*	0.08	1.00				
Number of tillers plant ⁻¹	-0.35*	0.15	-0.16	-0.16	0.26	1.00			
Number of panicles plant ⁻¹	-0.02	0.35*	-0.52**	0.10	0.27	0.70	1.00		
100-grain weight (g)	-0.08	0.03	0.21	-0.01	-0.22	-0.25	-0.07	1.00	
Sterility percentage (%)	-0.19	-0.45**	0.64**	-0.18	-0.28	-0.06	-0.43**	-0.32*	1.00
Grain yield plant ⁻¹ (g)	0.07	0.25	-0.66**	-0.14	0.35*	0.36*	0.43**	-0.11	-0.50**

*,** is statistically significant differences at the level of probability 0.05 and 0.01, respectively

Chlorophyll content was correlated negative and significant ≥ 0.01 with sterility percentage but positive with plant height and grain yield plant under natural conditions whereas, it was correlated had a positive and significant correlation (≥ 0.05) with panicle length, number of panicles plant⁻¹ and negative with sterility percentage under heat conditions. Under natural conditions the leaf rolling had positive correlation and significant (≥ 0.01) with panicle length, tillers number, panicles number plant⁻¹ and grain yield plant⁻¹ while under heat conditions had negative correlated with panicle length, panicles number plant⁻¹ and grain yield plant⁻¹ as well as positive correlation with sterility percentage. Plant height was correlated positive and significant (≥ 0.01) with 100-grain weight and negative with sterility percentage under natural conditions. Panicle length showed positive and highly significant correlation with number of tillers plant⁻¹ and number of panicles plant⁻¹ under natural conditions thus positive correlation and significant (≥ 0.05) with grain yield plant⁻¹ under stress. The highly correlation were found between the number of tillers plant⁻¹ and number of panicles plant⁻¹, 100-grain weight and grain yield plant⁻¹ under natural conditions. However, under stress conditions, the tillers number plant⁻¹ has correlated positively and significantly (≥ 0.05) with grain yield. The 100-grain weight was negative correlated and significant (≥ 0.05) with sterility percentage under both nature and heat stress conditions. The negative correlation and significant (≥ 0.05) was found between sterility percentage and grain yield plant⁻¹ under both nature and heat stress condition. Under natural conditions the grain yield plant⁻¹ had highly correlation and positive with chlorophyll content, leaf rolling, tillers number plant⁻¹ and panicles number plant⁻¹, while the negative and significant (≥ 0.01) correlation was confirmed with sterility percentage under natural conditions. Therefore, under stress conditions the positive correlations were found among grain yield and panicle length, tillers number and panicles number plant⁻¹ however, the correlation was significant (≥ 0.01) and negative with leaf rolling and sterility percentage.

Discussion

Climate change has been an increasingly significant factor behind fluctuations in the yield and quality of rice (*Oryza sativa* L.). The development and use of heat tolerant varieties is an effective way to reduce each type of grain damage based on the existence of each varietal difference (Kown and Torrie, 1964). Mean squares due to environments were found to be significant at probability level 0.01 for all studied characteristics indicating that; all environments showed significant (≥ 0.05) effects on all characteristics. The significant differences among rice genotypes in Table 4 showed the presence of genetic variability in the used material and provided a good opportunity for yield improvement. This specified that, the studied genotypes were different from environment to the other and ranked differently from natural to heat stresses conditions. This could show that further improvement through selection for all characters studied may be effective. Gaballah and Abu El-Ezz (2019) mentioned that the analysis of variance exposed significant differences due to environments, genotypes and their interaction for all studied characteristics in both seasons.

Concerning the genotypes mean performance over the two years and two environments the 'Hybrid 2', 'Sakha 107' and 'Giza 178' recorded the superior values for grain yield and related characteristics, refer to those three cultivars have good tolerance in both natural and heat stress conditions and could use in breeding programs to enhance the tolerance of cultivars against heat stress. High night-temperature stress during the reproductive stage decreases fertility of spikelet and yield by inhibiting the physiological processes. Abdallah *et al.* (2011) reported that high temperature prompts sterility, and reduced average grain yield by 22% as well as grain yield was adversely correlated with spikelet sterility under heat stress therefore, potential donors identified based on at least one trait included 'ARC 11094', 'Pinulupot 1', 'Tupa 729', 'Shinriki', 'Kameji' and 'Giza 178'. Heat and drought stress may cause harmful effects on growth characters and plant development, the main phase being injured is the reproductive phase, particularly, anthesis or grain filling phase (Faseela *et al.*, 2020). All genotypes showed spikelet sterility above 90% and indicated that increased temperature may limit grain yield by affecting fertility of spikelet and grain filling as well as net reduction in grain yield reach to 30.4% (Kumar *et al.*, 2015). Salt stress resulted significant decrease of growth and physiological characteristics in all rice genotypes; therefore, the lowest reduction was observed in some salt-tolerant genotypes namely Ghunsi, Nonabokra, Hogla, Holdegotal, Vusieri, and Kanchon (Rasel *et al.*, 2020). Maruyama *et al.* (2015) stated the increasing temperatures both accelerated and delayed the timing of heading. Extreme temperatures ($>4^{\circ}\text{C}$ daily maximum) increased the days to heading.

The most standing genotypes under stress conditions, which gave the minimum reduction were 'IET1444' and 'Sakha 107' while 'Sakha 102', 'Giza 177', 'Sakha 108' and 'Sakha 104' for most studied characteristics. 'NH219' is more tolerant to heat stress than wild type 'N22' as its percentage yield reduction is less than 'N22' (Poli *et al.*, 2013). Brestic *et al.* (2018) found that the selection for high yields was accompanied by the increase of photosynthetic productivity through unintentional improvement of leaf anatomical and biochemical characteristics including tolerance to non-optimal temperature conditions. Leaf area determination at earlier date was highly correlated with the total shoot biomass at the higher temperature (Pasuquin *et al.*, 2013). Hammer *et al.* (2020) revealed that the high temperature tolerance genotypes are more important and it is approach to genotype \times management for effective use of water through the crop cycle to maintain the productivity. At maturity, post-anthesis drought and heat stress significantly decreased the grain yield by 21.3%-65.2%, which was due to the changes of grain number per spike and thousand grain weight (Ahmad *et al.*, 2019). Improvement the efficacy of transpiration offered the best potential for adaptation relevant to climate changes. Manigbas *et al.* (2014) reported that the genetic difference in sterility % among the selected backcross populations grown in high temperature showed that great numbers of plants may be identified and selected with lower sterility%. Wang *et al.* (2020) cleared that OsNAC006 mutant expression profiles showed that growth was inhibited following drought and high temperature stress. Also, Paul *et al.* (2020) reported the two indica and one of the tropical japonica accessions exhibited severe heat sensitivity with

increased seed abortion; three tropical japonicas showed moderate heat tolerance, while temperate japonicas exhibited strong heat tolerance.

From correlation coefficient in Table 9, the results indicated that the leaf rolling under high temperature is remarkable to identify the genotypes as good tolerance were highly correlated and negative with grain yield, followed by sterility percentage and panicles number plant⁻¹. Gaballah and Abu El-Ezz (2019) recorded highly significant and positive phenotypic correlations between yield index and leaf rolling, sterility percentage, geometric mean productivity and stress susceptibility index, otherwise tillers number, panicles number plant⁻¹ and panicle weight, as yield attributes had a significant (≥ 0.01) negative correlation with the yield index. From our results we can conclude that the genotypes can be scored as heat tolerant, based on days to heading, leaf rolling, tillering productivity and spikelets sterility percentage and might be used as selection criteria for identifying heat tolerant genotypes. We could summarize the impact of heat stress on rice genotypes compared to natural conditions; heat stressed to induced earliness by 20.16%, reduction percentage by 21.77, 18.94, 19.03, 32.25, 35.56, 16.92 and 47.98 for chlorophyll content, plant height, panicle length, tillers number, panicles number plant⁻¹, 100-grain weight and grain yield plant⁻¹, respectively otherwise increasing in leaf rolling 316.70% and sterility percentage 492.45%. The significant correlation of grain yield with days to heading, days to anthesis and days to maturity was recorded with Sharma *et al.* (2013).

Conclusions

Breeding heat tolerant rice cultivars is one of the most important approaches used to alleviate the effect of heat stress, particularly in high temperature regions. The screening and selection approaches for heat conditions could differentiate the genotypes according to heat tolerance. The desirable characteristics mean values for panicles number plant⁻¹, 100-grain weight, sterility percentage and grain yield plant⁻¹ were recorded with 'Giza 178', 'Hybrid 2', 'Giza 179' and 'Sakha 107'. However, the grain yield plant⁻¹ had highly positive correlation with chlorophyll content, leaf rolling, tillers number and panicles number plant⁻¹, however, negative and significant (≥ 0.01) was confirmed with sterility percentage under natural conditions. Moreover, under heat stress conditions the positive correlations were found among grain yield, panicle length, tillers number and panicles number plant⁻¹ and had significant (≥ 0.05) and negative with leaf rolling and sterility percentage. Heat stress led to induce earliness by 20.16%, decrease ratio for panicles number plant⁻¹ (35.56%), 100-grain weight (16.92%) and grain yield plant⁻¹ (47.98%) in rice genotype.

Authors' Contributions

Conceptualization, Kh.A.A, Y.M., A.M., M.B., M.Gz., M.Gb., and Y.H. ;methodology, A.A., M.B., M.Gz., M.Gb., and Y.H. software, Kh.A.A, Y.M., A.M., M.B., M.Gz., M.Gb., and Y.H. validation, Kh.A.A, Y.M., A.M., M.B., M.Gz., M.Gb., and Y.H. investigation, A.M., M.B., M.Gz., M.Gb. resources, Kh.A.A, A.M., M.B., M.Gz., M.Gb. data curation, Kh.A.A, A.A., M.B., M.Gz., M.Gb., and Y.H. writing—original draft preparation, A.M., M.B., M.Gz., M.Gb., and Y.H. writing—review and editing, Kh.A.A, A.M., M.B., M.Gz., M.Gb., and Y.H. supervision Kh.A.A, A.M., M.G., M.B., M.G., N.K., and Y.H. funding acquisition, Kh.A.A, Y.M., A.M., M.B., M.Gz., M.Gb., and Y.H. All authors have read and agreed to the published version of the manuscript.

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Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

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