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Research Article

Effects of Heat Stress on Flowering, Fruit Set, and Seed Quality in Pepper (*Capsicum annuum* L.)

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ABSTRACT

This study investigated the impact of heat stress on reproductive development in two pepper varieties, Cayenne and Yolo Wonder, by comparing two planting dates (March 20th and May 1st) in Iraq. The experimental design was a Randomized Complete Block Design in a 2×2 factorial arrangements. The later planting date exposed plants to severe heat stress, which significantly impaired all measured reproductive parameters. A dramatic increase in flower abscission was observed, particularly in Yolo Wonder (67.85%), along with a severe reduction in fruit set percentage (54.9% for Cayenne vs. 20.8% for Yolo Wonder). Heat stress also negatively affected fruit morphology, reducing average fruit weight to 51.8g for Cayenne and 74.8g for Yolo Wonder. Seed quality was considerably compromised, as an evident reduction in the number of seeds per fruit (95.0 for Cayenne vs. 62.4 for Yolo Wonder) and final germination percentage (88.36% for Cayenne vs. 65.05% for Yolo Wonder). Statistical analysis confirmed significant main effects of variety and planting date, as well as a significant interaction for most traits, underscoring the differential response of the two varieties. The Cayenne variety consistently exhibited superior thermotolerance across all parameters, highlighting its greater genetic resilience compared to the highly sensitive Yolo Wonder. Therefore, it is recommended to cultivate the heat-sensitive Yolo Wonder variety only during spring or autumn to avoid summer heat. In contrast, the heat-tolerant Cayenne variety can be successfully cultivated during summer months, maintaining acceptable productivity under heat stress conditions. These findings emphasize the critical importance of variety selection and planting schedule optimization for sustainable pepper production in heat-prone regions like Iraq.

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Introduction

Pepper (*Capsicum annuum* L.) is a nutritionally and economically important vegetable crop globally. Its fruits are a rich source of vitamins A and C, capsaicin, and other beneficial compounds, consumed in various forms whether raw, cooked, or as a spice (Sherman and Billing, 1999; Dary and Mora, 2002).

Both the biotic and abiotic factors have a significant influence on pepper cultivation as they interact to determine plant health, yield, and the quality of fruits. Biotic stresses include mostly pathogens and pests, such as oomycete like *Phytophthora capsici* that cause crown and root rot causing severe wilting and up to 100 percent yield losses in vulnerable varieties (Parisi et al., 2020).

Anthracnose disease caused by different kind of fungus, *Colletotrichum* species, appears as sunken spots on fruits, and their incidence is worsened by high humidity and warm weather, whereas powdery mildew caused by *Leveillula taurica* is characterizes with white fungal spots that disturbs photosynthesis (Parisi et al., 2020). Bacterial spot (*Xanthomonas euvesicatoria*) causes water-soaked spots which become necrotic, and the viruses such as Pepper mild mottle virus cause a decrease in vigor due to mosaic symptoms and stunting (Parisi et al., 2020). The insects, including aphids (*Myzus persicae*), spider mites, are also direct feeders that transmit viruses to worsen the destruction (Zhang et al., 2024).

These problems are compounded by abiotic factors that modify the physiology of plants and expose peppers to biotic attacks. An elevated temperature of more than 32°C interferes with the pollen viability and fruit set resulting in the loss of leaf chlorosis and pitting, whereas a low temperature of less than 12°C leads to the loss of blossom (Zhang et al., 2024). Drought stress enhances the production of the reactive oxygen species (ROS), which damages membranes and slows down the photosynthetic rates, and salinity enhances the uptake of Na+, blocking the uptake of K+ and Ca2+, crucial for fruit development (Zhang et al., 2024). Extremes of soil pH, nutrient deficiency (e.g. boron or calcium resulting in blossom-end rot), and waterlogging due to excessive irrigation further reduce growth by causing a lack of root functionality and nutrient uptake (Zhang et al., 2024).

Among the abiotic factors, heat stress is a major environmental constraint that severely limits plant growth and agricultural productivity. Plants, including pepper, activate complex tolerance mechanisms involving heat shock proteins, antioxidants, and osmoprotectants to cope with high temperatures. Enhancing thermotolerance is a critical objective in modern agriculture, especially as heat stress poses an escalating threat to sustainable crop production under changing climate conditions (Hasanuzzaman et al., 2013). In Iraq, the cultivation of pepper is increasingly important, as evidenced by the growing harvested area and production (Table 1). Despite this growth, yields remain low compared to global averages, likely due to unfavorable environmental conditions, particularly high summer temperatures. The optimal temperature for pepper fruit formation and quality is between 21 and 29°C (Maas, 1990). When temperatures exceed 32°C, physiological disorders such as blossom end rot (BER) can occur on fruits, fruit set is severely reduced, and overall yields decline (Knott and Deanon, 1967). Despite the critical impact of temperature, research on the response of locally grown Iraqi pepper varieties to heat stress remains limited. Therefore, this study aimed to investigate the impact of high summer temperatures in Iraq on the flowering, fruit set, and seed formation of two distinct pepper varieties, Cayenne and Yolo Wonder.

Table 1. Area harvested, yield and production of pepper spp. in Iraq between 2016 -2023.

Year	Area harvested (ha)	Yield (kg/ha)	Production (ton)
2016	2554	6063.6	15488
2017	2373	5303.6	12584
2018	2477	8855.6	21931
2019	2919	7549.8	22034
2020	5097	9050.4	46130
2021	5391	10903	58781
2022	4871	8936.2	43528
2023	4790	10142.7	48584.57

Materials and Methods

Growing the seedling and preparing the plants

In this study, two local open pollinated pepper varieties were used viz., hot pepper (Cayenne) and sweet bell pepper (Yolo Wonder). The experiment was carried out in an open field in the farm of College of Science University of AL-Anbar during the 2024 growing season using spring crop at two planting dates (20th March and 1st May). The 1st planting date experiment and the 2nd planting date experiment were terminated on July 13th and August 5th respectively. Seedlings of the two varieties were raised in the laboratory by sowing seeds in germination trays filled with peat moss. One seed per cell was sown at a depth of 1 cm and received normal services until they became ready to be transplanted into plastic pots at 5-6 true leaves stage. Seedlings of each variety were then transferred to plastic containers (16 cm height; 14 cm diameter), which were filled with mixture of soil, compost, and sand at ratio 3:2:1. The mix was sterilized with an autoclave at 120°C for 30 min, the seedlings were hardened by direct sun exposure for 6-7 days and after three weeks were subsequently transferred to the ground. Experiments were conducted in a field measuring 2,500m^2 and each experiment occupied nearly half of the total area.

Preparation of the land for conducting experiments

The experimental field was deep-ploughed and was fertilized with the fermented cow manure to the extent of 1.5 t per area unit area. It was then fertilized with mineral fertilizer (70 kg of NPK 15–15-15). The row spacing was kept at 75 cm and an inter-plant distance of 45cm per row. Irrigation was done once or twice a day on plants according to ambient temperature. The original fertilization was administered at the transplanting stage and lasted 15 days with the 2.5 kg of high phosphate water soluble fertilizer (N4P36K4).

A balanced soluble fertilizer (N20:P20:K20) was also used at a rate of 2 kg every two weeks. Natural insect pollination was allowed throughout the experiment, as the study was conducted in an open field. No hand pollination was performed to simulate natural growing conditions. Bee activity and other pollinators were observed regularly during the flowering period. Plants were irrigated using a drip irrigation system. The irrigation schedule was adjusted based on daily evaporation rates, with plants receiving 1.5-2.0 L of water per plant daily during moderate temperatures (25-35°C) and 2.0-2.5 L during high temperatures (>35°C). Irrigation frequency was increased to twice daily during extreme heat conditions (>40°C). An integrated pest management approach was implemented. Preventive fungicide application (Beltanol @ 1 ml/L) was applied to seedlings before transplanting. Insecticide (Acetamiprid powder @ 1g/L) was applied as needed. Weekly pest scouting was conducted for whiteflies, aphids, and chewing insects. The spraying process was carried out early in the morning until the plants were completely wetted. No major disease outbreaks occurred during the experiment. Temperatures and relative humidity were recorded daily throughout the experiment for both planting dates using an HTC-1 electronic thermometer and hygrometer.

Assessment of impact of heat stress on the flowering of pepper plants

The following data were recorded for each plant throughout the experimental period:

Phenological development

Days to first anthesis (DFA) was recorded as the number of days from transplanting to the opening of the first flower.

Floral dynamics

Flower production

The number of newly opened flowers was counted every 4 days. The cumulative flower count was calculated for each plant.

Flower abscission

The number of abscised flowers (fallen flowers collected from the base of the plant) was counted every 4 days, and percent flower abscission was calculated as follows:

$$\%\ \text{flower abscission} = \frac{\text{Number of abscised flowers}}{\text{Total number of flowers}} \times 100$$

Assessment of impact of heat stress on fruit setting and quality of pepper fruits

Four primary branches were selected and labeled on each plant at the start of flowering, and the numbers of newly opened flowers on these branches were regularly recorded every 4 days. The total number of flowers and the number of successfully set fruits were recorded for each branch. Percent fruit set was calculated as follows (Vijayakumar et al., 2021):

$$\%\ \text{fruit set} = \frac{\text{Number of fruits set}}{\text{Total number of flowers}} \times 100$$

The duration of fruit growth period was recorded as the number of days from successful pollination (estimated by style wilting and the onset of ovary swelling) to horticultural maturity (for non-pungent types) or full color break (for pungent types). Fruits of non-pungent pepper were harvested when reached full size of edible maturity depending on the germplasm and became firm but before turning color (Yellow, Orange or Red) (Berke et al. 1999), while the pungent pepper fruits were harvested when fully red (Kim et al., 2023). Fresh weight (g) and fruit dimensions (cm) including length and width of each fruit from the labeled branches were then measured, and the total productivity per plant was measured at the end of experiment.

Assessment of impact of heat stress on seed formation and quality in pepper plantsfter harvest, fruits were stored at 25°C and 50% relative humidity for 7-10 days after-ripening. Seeds were manually extracted and air-dried at room temperature for 48 h. For germination testing, three replicates of 40 seeds each were placed on sterilized filter paper in 9 cm Petri dishes. The dishes were incubated under alternating temperatures: 30°C for 8 h in darkness and 20°C for 16 h in light, for 14 consecutive days (Pagamas and Nawata, 2008). Germinated seeds (radicle emergence ≥2 mm) were counted daily to calculate Germination Index (GI) and Final Germination Percentage (FGP).

The GI was calculated for each treatment according to the equation of Orchard (1977):

$$GI = \frac{\sum T_i N_i}{S}$$

Where Ti is the number of days from the start of the test (seed imbibition), N_i is the number of seeds that germinated on day i, S is the total number of seeds that germinated by the end of the test.

Lower GI value indicates more rapid and synchronized germination.

The FGP was calculated according to Bohra et al. (2024) as follows:

$$\% \text{ FGP} = \frac{\text{Total seeds germinated}}{\text{Total seeds tested}} \times 100$$

A higher FGP value indicates greater germination capacity of the seed population (Scott et al., 1984).

Data analysis

The experiment was arranged as a Randomized Complete Block Design (RCBD) in a 2 × 2 factorial scheme, with the two factors being pepper variety (Cayenne and Yolo Wonder) and planting date (March 20th and May 1st). The four treatment combinations were randomly allocated to five replications (blocks). Each experimental unit within a replication contained 15 seedlings of the same treatment, resulting in a total of 20 experimental units and 300 plants.

Prior to statistical analysis, all data were tested to verify the assumptions of analysis of variance. Normality of distribution was assessed using the Shapiro-Wilk test, and homogeneity of variances was evaluated using Levene's test. To meet the assumptions of normality and homoscedasticity for percentage data (such as flower abscission, fruit set, and germination percentage), values were subjected to an angular transformation (arcsine of the square root) prior to analysis. However, for clarity of interpretation, untransformed means were presented in the results section.

For data analysis, the mean values of the measurements were calculated for each experimental unit, resulting in a total sample size of (n = 5) for each treatment group. These means were then analyzed using a two-way analysis of variance (ANOVA) to test the main effects of variety, planting date, and their interaction. Mean comparisons were performed using the Least Significant Difference (LSD) test at a 5% significance level. All statistical analyses were conducted using SPSS software (Version 25, IBM Corp., USA).

Results and Discussion

As we had two planting dates, maximum temperature and relative humidity were recorded daily for the two experiments. The temperature through the first experiment ranged between 16 to 50°C, while relative humidity ranged between 22 to 53%. For the second experiment, the temperature ranged between 22 to 50°C and relative humidity ranged between 22 to 32%. For both experiment, temperatures reached the peak at 5th June, with extreme temperatures above 40°C occurring on 16 and 58 days in the first and second experiments, respectively (Figure 1, 2). The difference in temperature between the 1st and the 2nd planting date for the 1st flower opening was 12°C.

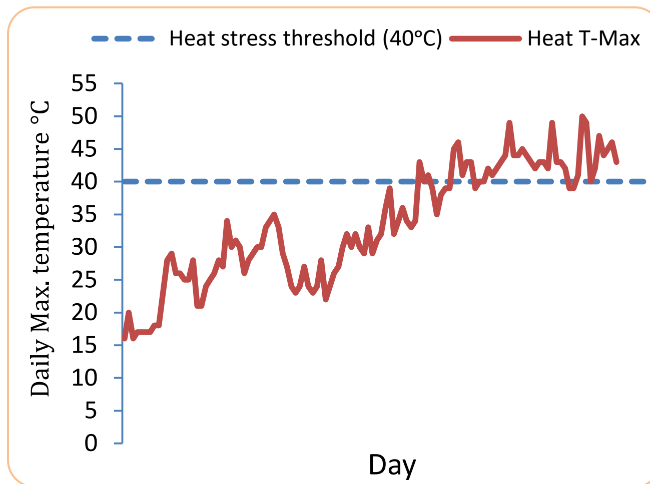

Figure 1. Daily maximum temperatures (°C) during the first planting date experiment.

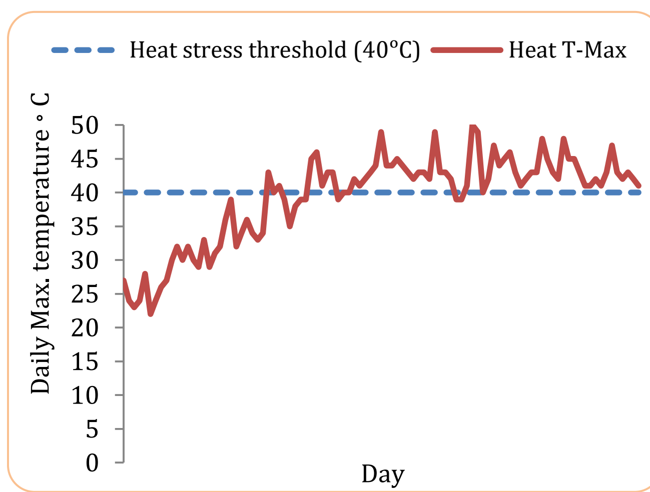

Figure 2. Daily mean temperatures (°C) during the second planting date experiment.

Effect of heat stress on flowering in pepper plants

Results in Table 2 showed that for both varieties, plants from the 2nd planting date reached first anthesis significantly earlier than those from the 1st planting date (Cayenne: 38.25 vs. 33.56 days; Yolo Wonder: 42.58 vs. 36.05 days). This was likely due to the accelerated developmental rate induced by higher temperatures, as the temperature at the time of first flowering was about 27°C and 39°C for the 1st and 2nd planting dates, respectively. Our results are consistent with those of Thuy and Kenji (2015), who studied the effect of heat stress on sweet pepper under field conditions in Japan. On the other hand, Cayenne flowered earlier than Yolo Wonder at both planting dates, indicating a genetic predisposition for earliness in Cayenne and revealing a clear pattern of phenological plasticity in response to increasing temperatures.

Table 2. Days to first anthesis, number of flowers per plant, and flower abscission percentage (mean ± S.E.) of Cayenne and Yolo Wonder pepper varieties as affected by planting date.

Planting date	Pepper variety	First anthesis (day)	No. of flowers /plant	Flower abscission (%)
20th March	Cayenne	38.25 ± 0.99^{aA}	32.86 ± 1.82^{aA}	33.52 ± 3.71^{aA}
	Yolo Wonder	42.58 ± 1.32^{bA}	25.53 ± 2.14^{bA}	48.83 ± 2.67^{bA}
1st May	Cayenne	33.56 ± 1.21^{aB}	28.47 ± 2.35^{aB}	40.36 ± 1.65^{aB}
	Yolo Wonder	36.05 ± 1.54^{bB}	18.26 ± 1.94^{bB}	67.85 ± 2.34^{bB}

$^{a-b}$ Comparison of the two pepper varieties within the same planting date, means within the same row followed by different letters are significantly different at $p \leq 0.05$.

$^{A-B}$ Comparison of the two planting dates within the same pepper variety, means within the same column followed by different letters are significantly different at $p \leq 0.05$.

The number of flowers produced per plant showed a significant decline for both varieties in the later planting date, which experienced higher temperatures. Cayenne consistently produced more flowers than Yolo Wonder under both conditions. Its ability to maintain a relatively high flower count (28.47 in Cayenne vs. 18.26 in Yolo Wonder) under the stressful May planting conditions is a strong indicator of its superior thermotolerance. Yolo Wonder appeared more sensitive, showing a more drastic reduction in its reproductive potential.

The flower abscission rate for Yolo Wonder reached 67.85% under the hotter May conditions, compared to an already high 48.83% in March. This means that under severe heat, over two-thirds of its flowers abscised and failed to develop into fruit. This high rate of flower abscission is a primary mechanism underlying yield loss in heat-sensitive varieties. In contrast, Cayenne not only showed a lower abscission rate overall but also a much smaller increase in abscission under stress (from 33.52% to 40.36%). This robustness suggests that Cayenne possesses more effective mechanisms to maintain pollen fertility and hormonal balance under heat stress. These mechanisms could include better antioxidant systems, higher concentrations of heat-shock proteins, or more stable pollen tube growth.

Our findings suggest that Cayenne is genetically better equipped to handle heat stress through a combination of earliness, sustained flower production, and, most critically, physiological mechanisms that drastically reduce flower abscission. For cultivation in regions experiencing terminal heat stress or for early summer plantings, Cayenne presents a far more reliable and productive choice compared to Yolo Wonder. According to FAOSTAT (2025), global crop production data indicate that temperature extremes significantly affect yield stability and productivity in major crops, including pepper.

Effect of heat stress on fruit setting in pepper plants

According to our findings, the fruit set percentage for both varieties was higher in the 1st planting date (63.87% and 32.63% for Cayenne and Yolo Wonder, respectively). In contrast, the 2nd planting date led to a significant decrease in the fruit set rate for both varieties (54.9% and 20.8% for Cayenne and Yolo Wonder, respectively). In both cases, the Cayenne variety showed the highest fruit set rate (Table 3). Our results agree with those of Saha et al. (2010), who studied the effect of high-temperature stress on the performance of twelve sweet pepper genotypes. This decrease in fruit set percentage can be explained by the fact that high temperature reduces indole-3-acetic acid levels, particularly the auxin transport capacity in the reproductive organs, which ultimately induces reproductive organ abscission and consequently reduces the percentage of fruit set (Huberman et al., 1997). Furthermore, exposure to high temperature during the critical developmental window from microspore mother cell meiosis to tetrad dissolution significantly reduces pollen viability and prevents anthers from dehiscing (Erickson and Markhart, 2002). Specifically, heat stress inhibits pollen grain development between final tetrad formation and tetrad dissolution. This reduction in pollen viability subsequently leads to decreased fruit set and results in smaller fruit size (Erickson and Markhart, 2001).

The number of fruits produced per pepper plant decreased for both varieties under high-temperature conditions (Table 3). At both planting dates, Yolo Wonder produced the least number of fruits, with a significant difference between the two dates (8.76 and 3.72 fruits per plant for the 1st and 2nd planting dates, respectively). In contrast, the difference for Cayenne between the two planting dates was not statistically significant (15.21 and 10.45 fruits per plant for the 1st and 2nd planting dates, respectively). This trend was consistent with the findings of Saha et al. (2010). Furthermore, exposure to high temperatures significantly reduced the fresh weight of fruits in both varieties when planted in May.

This finding agrees with Pagamas and Nawata (2008) and Thuy and Kenji (2015), who stated that fruit weight in pepper is significantly reduced under high temperature. Furthermore, high temperatures led to a significant decrease in both fruit length and width for the two varieties (Table 3). Thuy and Kenji (2015) noted that morphological characteristics, including fruit length and width of sweet pepper fruits, are strongly affected by high temperature. This poor growth and development of chili pepper fruits and seeds under a high-temperature regime can be attributed to the reduction in the accumulation of assimilates in these organs (Pagamas and Nawata, 2007). Additionally, high temperature causes a loss of cell water content, which reduces cell size and ultimately limits growth (Ashraf et al., 2004; Rodríguez et al., 2005). Furthermore, Pagamas and Nawata (2008) mentioned that heat stress may disrupt both early-stage cell division and later-stage cell enlargement during fruit growth and development, ultimately negatively impacting fruit characteristics, this failure in cell division could be attributed to oxidative damage caused by reactive oxygen species (ROS) under heat stress, which disrupts cellular structures and mitotic processes.

The results showed that heat stress had a significant negative effect on the fruit growth period. The fruit growth period decreased for both studied varieties in line with the increase in temperature. However, Yolo Wonder showed a shorter fruit growth period at both planting dates, with the shortest period occurring under heat stress (declining from 68.74 to 51.86 days for the 1st and 2nd planting dates, respectively) (Table 3). The acceleration of fruit maturation and ripening under high temperatures is an adaptive mechanism, allowing plants to disperse seeds faster under stress conditions (Mizrahi, 1982), which may be attributed to an escape mechanism (Rodríguez, 2005).

When comparing plant productivity for both varieties, exposure to heat stress caused a significant decrease in yield for both (Table 3), which agrees with Erickson and Markhart (2001), who stated that fruit set and productivity of pepper are reduced during periods of high temperature. The Yolo Wonder variety showed a clear decline in production compared to the Cayenne variety, which exhibited a more relative decrease. The deterioration in Yolo Wonder was particularly pronounced, indicating its high sensitivity to heat stress. High temperatures can accelerate the accumulation of growing degree days, or heat units, resulting in a shorter growing season. A shorter growing season would lead to reduced light interception, resulting in decreased crop production. Additionally, a shorter growing season could cause inefficient use of water and nutrients, which also plays a critical role in yield reduction under heat stress conditions (Harrison et al., 2011). The loss of productivity under heat stress is primarily attributed to a decreased assimilatory capacity (Hay and Porter, 2006), resulting from reduced photosynthesis caused by altered membrane stability (Zhang et al., 2006) and enhanced maintenance respiration costs (Reynolds et al., 2007), which collectively reduce radiation use efficiency (RUE; the biomass produced per unit of light intercepted by the canopy).

Table 3. Effect of planting date and heat stress on fruiting traits (mean ± S.E.) of Cayenne and Yolo Wonder pepper varieties.

Planting date	Pepper variety	Fruit set %	Number of fruits /plant	Fruit fresh weight (g)	Fruit length (cm)	Fruit width (cm)	Fruit growth period (day)	plant Productivity (g)
20th March	Cayenne	63.87±2.53aA	15.21 ± 1.16aA	60.46± 4.37aA	12.33±2.16aA	2.18±1.53aA	73.44±1.88aA	932.32 ± 37.3aA
	Yolo Wonder	32.63±3.41bA	8.76 ± 1.88bA	91.63±3.71bA	8.49±1.73bA	6.97±1.39bA	68.74±1.13bA	725.45 ± 25.55bA
1st May	Cayenne	54.9±3.54aB	10.45 ± 0.89aB	51.8±3.54aB	10.42±0.95aB	2.01±0.92aB	63.64±2.63aB	840.52 ± 23.12aB
	Yolo Wonder	20.8±3.12bB	3.72 ± 0.98bB	74.8±2.94bB	7.1±1.21bB	5.92±1.53bB	51.86±1.95bB	291.0± 20.71bB

a-b Comparison of the two pepper varieties within the same planting date, means within the same row followed by different letters are significantly different at $p \leq 0.05$.

A-B Comparison of the two planting dates within the same pepper variety, means within the same column followed by different letters are significantly different at $p \leq 0.05$.

Effect of heat stress on seed formation and quality in pepper plants

When comparing the effect of planting date and exposure to heat stress on the number of seeds per fruit, it was noted that high temperature had a negative effect (Table 4). The number of seeds decreased significantly in both studied varieties (from 112.3 to 95.0 seeds per fruit for Cayenne, and from 85.5 to 62.4 seeds per fruit for Yolo Wonder). This finding is consistent with Pagamas and Nawata (2008), who stated that the number of seeds per fruit is significantly affected by heat stress treatment. The reduction in seeds per fruit may have contributed to the smaller fruit size (Erickson and Markhart, 2002), as a positive correlation which has been observed between fruit weight and the number of seeds per fruit in sweet pepper (Heuvelink and Korner, 2001). Furthermore, high-temperature stress reduces the number of viable pollen grains, impairing fertilization and consequently leading to fewer seeds per fruit (Erickson and Markhart, 2002). High temperature also significantly increased the percentage of abnormal seeds, which agrees with Pagamas and Nawata (2008). However, Cayenne exhibited the lowest percentage of seed deformity under high-temperature stress (6.8%), in contrast to Yolo Wonder, which showed a relatively high percentage (22.7%). This indicates the greater sensitivity of Yolo Wonder in terms of seed development when exposed to heat stress. Pagamas and Nawata (2008) explained that following fertilization, seed abortion during the initial stages is likely caused by a failure in proper cell division, which may be induced by high-temperature stress; this explains the decrease in seed number in the two studied varieties under heat stress conditions.

The germination index (GI) increased significantly in both varieties under heat stress (Table 4). Contrary to common expectations where a lower GI indicates slower germination, in this specific index calculation, a higher GI value signifies a slower germination rate. In all cases, Yolo Wonder showed the highest GI values for both planting dates (2.30 and 4.27 for the 1st and 2nd planting dates, respectively), confirming its slower and more impaired germination rate compared to Cayenne.

The study revealed the negative effect of heat stress on the FGP, which decreased significantly in both varieties. This effect was more pronounced in Yolo Wonder (decreasing from 82.53% to 65.05%) than in Cayenne, further confirming this variety's greater sensitivity to heat stress.

We can conclude that the reduction in seed yield and quality under high-temperature conditions is related to the disturbance of assimilate supply from source organs and the suppression of sink strength in pepper. Furthermore, heat stress after anthesis is detrimental to subsequent seed growth and development due to disturbances in carbohydrate and lipid accumulation, resulting in a reduction in seed weight, germination rate, and vigor (Pagamas and Nawata, 2007).

Table 4. Effect of heat stress on seed traits (meanv± S.E.) of Cayenne and Yolo Wonder pepper varieties.

Planting date	Pepper variety	No. of seeds/fruit	% abnormal seeds	germination index	Final Germination Percentage
20th March	Cayenne	112.13 ± 6.61[aA]	3.24 ± 0.87[aA]	1.92 ± 0.08[a]A	92.18 ± 2.16[aA]
	Yolo Wonder	85.75 ± 5.28[bA]	8.55 ± 1.32[bA]	2.30 ± 0.11[bA]	82.53 ± 3.55[bA]
1st May	Cayenne	95.01 ± 5.54[aB]	6.68 ± 1.57[aB]	2.28 ± 0.20[aB]	88.36 ± 3.01[aB]
	Yolo Wonder	62.44 ± 7.18[bB]	22.37 ± 3.56[bB]	4.27 ± 0.35[bB]	65.05 ± 5.5[bB]

[a-b] Comparison of the two pepper varieties within the same planting date, means within the same row followed by different letters are significantly different at $p \leq 0.05$.

[A-B] Comparison of the two planting dates within the same pepper variety, means within the same column followed by different letters are significantly different at $p \leq 0.05$.

The analysis of variance for flowering, fruiting, and seed traits provided robust statistical support for the observed differential impact of heat stress (Tables 5, 6, 7). The main effects of variety (V) and planting date (D) were highly significant ($p \leq 0.01$) for all parameters measured, confirming that both genetic identity and heat stress independently had a substantial impact on plant performance. Crucially, the interaction between variety and planting date (V × D) was also statistically significant for the vast majority of traits. This consistent and significant interaction effect verifies that the two varieties responded differently to the heat stress imposed by the later planting date. The negative impact of high temperatures was consistently and disproportionately more severe for the Yolo Wonder variety than for Cayenne across all stages of reproduction from flower abscission and fruit set to seed quality and germination. Therefore, the ANOVA results unequivocally validate the conclusion that Cayenne possesses a genetically superior thermotolerance mechanism, while Yolo Wonder is highly vulnerable to heat stress.

Conclusion

This study demonstrates a clear differential response to heat stress between the two pepper varieties, Yolo Wonder and Cayenne, under the specific environmental conditions of this single-location experiment. The findings consistently indicate that the Cayenne variety possesses a superior degree of thermotolerance under these experimental conditions, evidenced by its earlier flowering, sustained flower and fruit production, lower flower abscission rates, and better maintenance of seed quality under high-temperature conditions. In contrast, the Yolo Wonder variety exhibited greater sensitivity in this study, with its reproductive performance and seed viability being significantly more compromised under heat stress.

Table 5. Analysis of variance (F-values) for the effect of variety (V), planting date (D) and their interaction for flowering traits of pepper plants.

Source of Variation	df	First Anthesis	No. of Flowers/plant	Flower abscission
Variety (V)	1	25.4**	38.9**	105.3**
Planting Date (D)	1	18.7**	45.2**	215.8**
Interaction (V × D)	1	5.8*	12.5**	45.6**
Error	16			

*, ** Significant at $p \leq 0.05$ and $p \leq 0.01$ probability levels, respectively.

Table 6. Analysis of variance (F-values) for the effect of variety (V), planting date (D) and their interaction for fruiting traits of pepper plants.

Source of Variation	df	Fruit Set %	Number of fruits /plant	Fruit fresh weight	Fruit length	Fruit width	Fruit growth period*	plant Productivity
Variety (V)	1	125.4*	98.5**	58.9**	120.7**	895.4**	185.2**	285.4**
Planting Date (D)	1	89.6**	150.2**	45.2**	65.8**	55.1**	320.5**	410.7**
Interaction (V × D)	1	25.8**	40.3**	12.5**	15.2**	25.4**	45.8**	95.3**
Error	16							

*, ** Significant at $p \leq 0.05$ and $p \leq 0.01$ probability levels, respectively.

Table 7. Analysis of variance (F-values) for the effect of variety (V), planting date (D) and their interaction for seed traits of pepper plants.

Source of Variation	df	No. of Seeds/fruit	abnormal seeds %	germination index**	% Final Germination
Variety (V)	1	102.1**	205.4**	150.8**	88.5**
Planting Date (D)	1	77.3**	310.2**	280.5*	95.2**
Interaction (V × D)	1	18.9**	65.8**	65.4*	22.4**
Error	16				

*, ** Significant at p ≤ 0.05 and p ≤ 0.01 probability levels, respectively.

Although these results provide valuable insights into varietal differences in heat tolerance, it is important to note that this study was conducted at a single location over one growing season. Therefore, the recommendations should be interpreted with caution pending validation across multiple locations and seasons.

For the specific environmental conditions similar to this study site, the Cayenne variety presents a more resilient and reliable choice for plantings that may encounter terminal heat stress, whereas the cultivation of more sensitive varieties like Yolo Wonder may perform better when confined to cooler periods. Furthermore, the identified traits of Cayenne suggest its potential as a valuable genetic resource for future breeding programs aimed at enhancing heat tolerance in pepper. Further multi-location and multi-season trials are recommended to confirm these findings and develop more robust cultivation recommendations.

Authors' Contributions

The author conceptualized and designed the study, conducted the data analysis, and drafted the manuscript. The author reviewed and approved the final version and agrees to be accountable for all aspects of the work, ensuring accuracy and integrity in reporting.

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Conflict of Interest

The authors declare no conflict of interest.

Sustainable Development Goals Targeted

SDG 2: Zero Hunger

SDG 12: Responsible Consumption and Production

SDG 13: Climate Action

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