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Effect of exogenous salicylic acid on photosynthetic efficiency and yield of wheat under heat stress

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Abstract

This research examined exogenous salicylic acid (SA) effects on photosynthetic efficiency and grain yield of spring wheat (*Triticum aestivum* L. cv. Bezostaya) under heat stress. A factorial field experiment at Ankara University, Turkey (2021-2023) combined two temperature regimes (ambient 25 ± 2 °C; heat stress 38 ± 2 °C via open-top chambers) with four SA concentrations (0, 0.5, 1.0, 1.5 mM) applied as foliar spray at booting. Heat stress reduced net photosynthetic rate by 39.1% and Fv/Fm by 41.9%. Application of 1.0 mM SA under heat restored Fv/Fm to 0.63 from 0.47 (34.0% recovery). Grain yield increased by 23.7% with 1.0 mM SA under heat. The 1.5 mM dose was less effective, indicating a dose-dependent optimum. SA-mediated photosynthetic protection appears a practical heat-stress strategy for wheat.

Keywords: Salicylic acid, wheat, heat stress, photosynthetic efficiency, Fv/Fm, chlorophyll fluorescence, grain yield, stomatal conductance

Introduction

What happens to wheat productivity when temperatures climb beyond the crop's tolerance window? Wheat feeds roughly 2.5 billion people, yet it remains sensitive to temperatures exceeding 30 °C during anthesis^[1]. Heat events of three to five days above 35 °C can reduce grain number per spike by 30-40%^[2]. PSII reaction centres are particularly vulnerable, and Fv/Fm drops sharply above 35 °C^[3]. Rubisco activase denaturation compounds carbon fixation losses^[4, 5].

Salicylic acid (SA) is a phenolic signalling molecule that improves thermotolerance through membrane stabilisation and antioxidant upregulation^[6, 7, 8]. In wheat, SA pre-treatment reduced malondialdehyde under high temperatures^[9], but field evidence linking SA to photosynthetic gains and grain yield is limited. Open-top chambers allow controlled field warming while retaining natural conditions^[10, 11].

This research tested whether foliar SA at booting would protect PSII function and sustain grain yield in wheat exposed to a 13 °C temperature increase above ambient.

Materials and Methods

Research area and Environmental conditions

The experiment was conducted at the Dışkapı Agricultural Research Station (39°49'N, 32°52'E, 891 m a.s.l.) of Ankara University. The soil is a Typic Haploxerept (clay-loam, pH 7.8, organic matter 1.46%). Mean annual rainfall is 387 mm. During the 2021-2022 season, April-June mean temperature was 16.3 °C (max 34.8 °C); the 2022-2023 season was warmer (17.1 °C mean, 36.2 °C max). Humidity during heat-stress imposition ranged 28-42%.

Field experimental design

A split-plot design in randomised complete blocks (three replications) was used. Main plots had two temperature regimes: ambient and heat stress via polycarbonate open-top chambers (2.5 m diameter, 2.0 m height). Sub-plots received SA at 0, 0.5, 1.0, or 1.5 mM with 0.05% Tween-20 in 500 L ha⁻¹, applied at booting (Zadoks GS 45) and five days later. Each sub-plot was 3 m × 2 m. Chambers were maintained 15 days from booting.

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Materials

Spring wheat cv. Bezostaya was sown at 450 seeds m^{-2} on 15 October 2021 and 12 October 2022. Basal fertiliser: 60 kg N ha^{-1} + 40 kg P_2O_5 ha^{-1} at sowing, 40 kg N ha^{-1} top-dress at tillering. SA ($\geq 99\%$ purity, Sigma-Aldrich) was prepared fresh; controls received water plus surfactant.

Methods

Gas exchange (Pn, gs, E, Ci) was measured with a LI-6400XT

(LI-COR) on flag leaves at 0, 3, 6, 9, 12, and 15 days after SA application. Chlorophyll fluorescence (Fv/Fm) was recorded using a PAM-2500 (Walz); chlorophyll content was estimated with a SPAD-502 meter. At maturity, grain yield, 1000-grain weight, and harvest index were determined from 1 m^2 harvests. Split-plot ANOVA was performed in SAS 9.4 with Tukey's HSD at $p \leq 0.05$.

Results

Table 1: Gas exchange and chlorophyll parameters of wheat under heat stress and salicylic acid treatments (mean $\pm$ SE, n = 6, pooled across two seasons)

Treatment	Pn ($\mu\text{mol m}^{-2} \text{s}^{-1}$)	gs ($\text{mol m}^{-2} \text{s}^{-1}$)	E ($\text{mmol m}^{-2} \text{s}^{-1}$)	Ci ($\mu\text{mol mol}^{-1}$)	SPAD	Fv/Fm
Control + 0 mM SA	18.4 $\pm$ 0.9a	0.31 $\pm$ 0.02a	4.8 $\pm$ 0.3a	268 $\pm$ 11a	47.3 $\pm$ 1.8a	0.75 $\pm$ 0.02a
Control + 1.0 mM SA	20.3 $\pm$ 0.8a	0.34 $\pm$ 0.02a	5.3 $\pm$ 0.3a	274 $\pm$ 12a	49.2 $\pm$ 1.6a	0.77 $\pm$ 0.02a
Heat + 0 mM SA	11.2 $\pm$ 0.8d	0.18 $\pm$ 0.02d	2.9 $\pm$ 0.3d	221 $\pm$ 14c	34.4 $\pm$ 2.0c	0.47 $\pm$ 0.03d
Heat + 0.5 mM SA	14.8 $\pm$ 1.1c	0.24 $\pm$ 0.02c	3.7 $\pm$ 0.4c	243 $\pm$ 11b	39.1 $\pm$ 1.7b	0.55 $\pm$ 0.03c
Heat + 1.0 mM SA	16.9 $\pm$ 0.9b	0.28 $\pm$ 0.03b	4.4 $\pm$ 0.3b	258 $\pm$ 13ab	41.7 $\pm$ 1.9b	0.63 $\pm$ 0.02b
Heat + 1.5 mM SA	15.3 $\pm$ 1.2bc	0.25 $\pm$ 0.02bc	3.9 $\pm$ 0.4bc	248 $\pm$ 12b	38.6 $\pm$ 2.2b	0.57 $\pm$ 0.03c

Heat stress raised canopy air temperature by 12.7 $^{\circ}\text{C}$ above ambient during the 15-day treatment. Net photosynthetic rate declined from 18.4 to 11.2 $\mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$ in heat-stressed wheat without SA (Table 1). Application of 1.0 mM SA

maintained Pn at 16.9 $\mu\text{mol CO}_2 \text{m}^{-2} \text{s}^{-1}$, not statistically different from the unstressed control. The 0.5 mM dose gave intermediate protection while 1.5 mM was less effective than 1.0 mM.

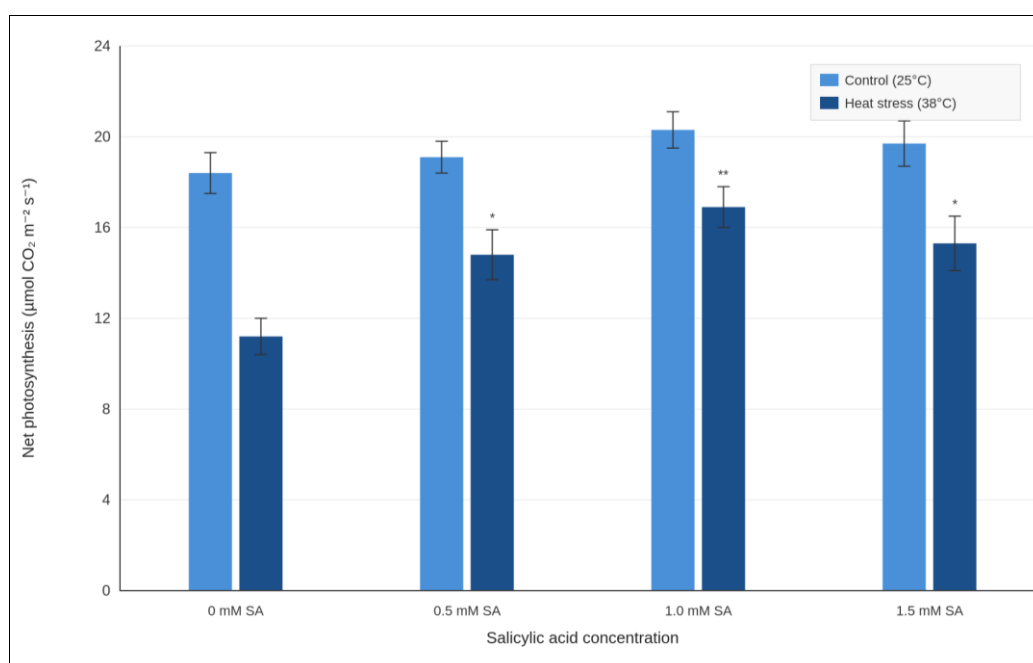


Fig 1: Net photosynthetic rate of wheat flag leaves under ambient (25 $^{\circ}\text{C}$) and heat stress (38 $^{\circ}\text{C}$) at four SA concentrations, day 15. Error bars = $\pm$ SE (n = 6). * $p < 0.05$, ** $p < 0.01$ vs heat + 0 mM SA

Fv/Fm in untreated heat-stressed plants dropped from 0.81 to 0.47 over 15 days (Figure 2). SA at 1.0 mM slowed this decline, with Fv/Fm stabilising around 0.63 by day 12. SPAD readings

mirrored this pattern; heat reduced SPAD by 27.3% but 1.0 mM SA limited the loss to 11.8%.

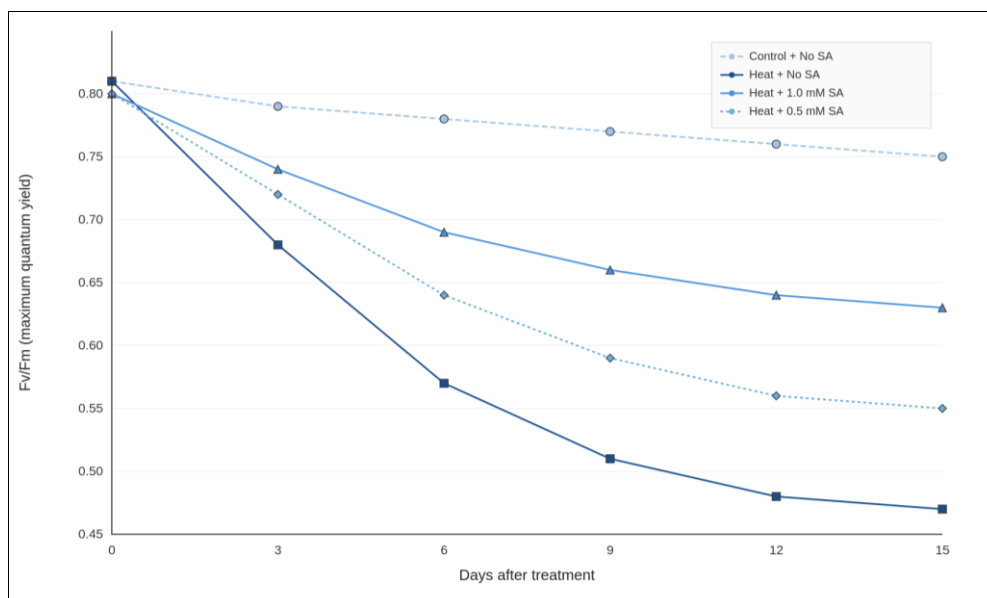


Fig 2: Temporal changes in Fv/Fm in wheat flag leaves over 15 days of heat stress under different SA treatments. Means of six replicates, two seasons

Table 2: Grain yield and yield components as affected by heat stress and SA application (mean±SE, two seasons pooled)

Treatment	Grain yield (kg ha ⁻¹)	1000-grain wt (g)	Biomass (kg ha ⁻¹)	Harvest index
Control + 0 mM SA	4870±186a	42.1±1.3a	11878±420a	0.41±0.02a
Heat + 0 mM SA	3210±174c	31.4±1.5c	8917±380c	0.36±0.02b
Heat + 0.5 mM SA	3580±195bc	35.1±1.4b	9722±410bc	0.37±0.02b
Heat + 1.0 mM SA	3970±168b	37.8±1.2b	10750±370b	0.37±0.01b
Heat + 1.5 mM SA	3640±210bc	35.9±1.6b	9944±420b	0.37±0.02b

Grain yield averaged 4,870 kg ha⁻¹ under ambient conditions versus 3,210 kg ha⁻¹ under heat alone (Table 2). The 1.0 mM SA treatment produced 3,970 kg ha⁻¹ under heat, recovering 57% of the yield gap. The 1000-grain weight was most responsive to SA, increasing from 31.4 g (heat, no SA) to 37.8 g (heat, 1.0 mM SA). Harvest index dropped from 0.41 to 0.36 under heat regardless of SA dose.

Discussion

The SA-mediated Fv/Fm recovery of 34.0% exceeds the 18-22% reported for controlled-environment wheat [9], likely because field chambers allowed gradual warming rather than abrupt transitions, giving SA-primed defence pathways more response time. Similar protective effects have been reported for maize [12] and rice [13]. The bell-shaped dose-response where 1.5 mM was less effective than 1.0 mM matches observations across multiple stress contexts [6], attributed to excessive H₂O₂ accumulation at higher SA concentrations [14].

Stomatal conductance was partially maintained by SA under heat, consistent with evidence that SA modulates aquaporin expression in wheat [15]. The harvest index showed limited response, indicating SA protected source activity without altering assimilate partitioning. On-farm validation under natural heatwaves is a logical next step [16].

Conclusion

Foliar SA at 1.0 mM during booting protected field-grown wheat photosynthesis under 38 °C heat stress, recovering Fv/Fm by 34.0% and maintaining Pn near unstressed levels. Grain yield improved by 23.7%, driven by higher 1000-grain weight. The effective dose (69 g SA ha⁻¹) is inexpensive and field-applicable. These two-season results support SA sprays as a heat-stress management tool for wheat in semi-arid regions.

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