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Foliar applications of ferulic acid and salicylic acid improve growth, bioactive compounds, and fruit quality in tomato

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Abstract: Ferulic acid (FA) and salicylic acid (SA) are phenolic compounds with recognized potential as metabolic elicitors in plants. This study evaluated the effects of exogenous application of FA and SA (0.5 mmol L⁻¹), applied individually or in combination, on the morphophysiological and biochemical responses of tomato cv. Santa Clara. The experiment was conducted in a greenhouse using plants cultivated in pots. Morphophysiological evaluations included growth and yield parameters, while biochemical analyses comprised photosynthetic pigments, soluble sugars, and phenolic compounds in the fruits. FA increased chlorophyll content and soluble sugars, whereas SA enhanced the accumulation of polyphenols and flavonoids. The combined application of FA+SA led to greater increases in leaf area, plant height, fruit number, fruit diameter, and biomass production, particularly root dry mass. In addition, the combined treatment improved the accumulation of bioactive compounds associated with fruit nutritional quality. Overall, the results indicate that FA and SA, particularly when applied together, represent a promising strategy to enhance tomato growth, productivity, and fruit quality under protected cultivation conditions.

1. Introduction

Tomato, belonging to the Solanaceae family, is one of the most widely cultivated and consumed crops worldwide. China is among the largest producers, while Brazil ranks among the top ten (CONAB, 2025). Beyond its economic importance, tomatoes stand out for their nutritional value, being rich in vitamins (A and ascorbic acid), phenolic acids (*p*-coumaric, ferulic acid, and caffeic acid), flavonoids (rutin, quercetin, naringenin, and kaempferol), and carotenoids (lycopene, α -carotene, and β -carotene) (Chaudhary *et al.*, 2018). Additionally, they provide essential minerals, sugars, amino acids, and fiber, underscoring their nutritional value (Shah *et al.*, 2021).

Despite their nutritional and economic importance, tomato crops face several challenges, particularly biotic and abiotic stresses. Climate variability, including rising temperatures, water scarcity, and extreme rainfall, can adversely affect yield and fruit quality (Ahmad *et al.*, 2023). These factors directly affect essential physiological processes, such as plant photosynthesis and cellular respiration, thereby compromising plant development and fruit quality (Bacelar *et al.*, 2024). Additionally, these climate changes can increase the incidence of fungal and bacterial diseases, exacerbating productivity losses (Singh *et al.*, 2023).

To face these challenges, new agricultural strategies have been explored. The use of elicitors has gained prominence as a sustainable tool in agriculture, enabling increased yield and improved crop quality (Gorni *et al.*, 2023; Silveira *et al.*, 2025). Compounds such as ferulic acid (FA) and salicylic acid (SA), recognized as elicitors, have been widely studied for their roles in modulating plant physiological and biochemical responses, enhancing tolerance to environmental stresses, and improving agronomic traits (Gorni *et al.*, 2021 a; Linić *et al.*, 2021).

Regarding elicitors that act on secondary metabolism, it is well established that SA is a phenolic compound abundantly present in plants and is involved in various physiological processes of the plant, functioning both as a growth regulator and as an abiotic elicitor, capable of enhancing the synthesis of secondary compounds of commercial interest (Gorni and Pacheco, 2016; Gorni *et al.*, 2020). SA acts to promote plant tolerance to high and low temperatures (Jalili *et al.*, 2023), in the induction of flowering (El-Hady *et al.*, 2021), in the stimulation of root development, in the closure of stomata, and in the reduction of transpiration (Saleem and Fariduddin, 2024) and in the regulation of the synthesis of chlorophylls, carotenoids, photosynthesis, transpiration, and stomatal conductance (Ramos Melo *et al.*, 2023; Saleem and Fariduddin, 2024). SA increases the activation of enzymes involved in the biosynthesis of certain classes of secondary metabolites (Ali, 2021; Gorni *et al.*, 2021 b).

FA is a phenolic acid commonly found in plant cell walls, providing structural support and protection against environmental stresses (Khan *et al.*, 2024) and exerts different effects on plant morphological,

physiological, and biochemical processes (He and Ding, 2020; Fiorito *et al.*, 2024), including in promoting plant growth and development (Fiorito *et al.*, 2024), increasing photosynthetic efficiency (Linić *et al.*, 2021), and in addition to being associated with defense processes (Khan *et al.*, 2024). In addition, FA is a molecule that interacts with different phytohormones such as jasmonic acid (JA) and SA, which are key regulators of plant defense responses (Niharika *et al.*, 2021), and this elicitor has been used in applications as a stimulating agent to produce several secondary metabolites (Gorni *et al.*, 2021 b; Fiorito *et al.*, 2024).

FA and SA applications in tomato plants have demonstrated potential to stimulate physiological responses and improve fruit quality (Shu *et al.*, 2022; Fiorito *et al.*, 2024). Previous studies at concentrations of 1 mmol L⁻¹ reported positive effects, including increases in biomass accumulation, chlorophyll content, and secondary metabolite biosynthesis (Gorni *et al.*, 2021 a; Shu *et al.*, 2022; Gorni *et al.*, 2023; Fiorito *et al.*, 2024). However, higher concentrations of phenolic elicitors may also induce oxidative stress or phytotoxic effects, potentially limiting plant performance and productivity. Despite the growing interest in the use of phenolic compounds in horticultural crops, studies evaluating the combined application of FA and SA at lower concentrations remain limited, particularly under greenhouse cultivation conditions for tomato plants.

In this context, using 0.5 mmol L⁻¹ for both FA and SA represents a more balanced and potentially safer strategy to enhance plant metabolism while minimizing adverse effects associated with excessive elicitor concentrations. Furthermore, investigating the interaction between these compounds at reduced doses may provide important insights into their combined effects on plant growth, fruit production, and biochemical quality.

We hypothesized that the combined application of FA and SA at low concentrations would improve morphophysiological traits, yield, and the accumulation of bioactive compounds in tomato plants, without causing metabolic stress. Therefore, this study aimed to evaluate the effects of exogenous application of FA and SA, individually or in combination, at 0.5 mmol L⁻¹ on growth, productivity, photosynthetic pigments, soluble sugars, and phenolic compounds in tomato plants cultivated under protected conditions.

2. Materials and Methods

Experimental conditions and plant cultivation

The experiment was conducted in 18-liter pots in a greenhouse at the Escola Superior de Agronomia de Paraguaçu Paulista (ESAPP), Paraguaçu Paulista, São Paulo, Brazil, from March 2020 to July 2020. The soil, classified as a Latosol with a sandy texture, was collected from the field and passed through a 5-mm mesh sieve prior to use. Its chemical analysis revealed the following characteristics as shown in Table 1. The soil was corrected with 15 g of dolomitic limestone per pot 30 days before sowing. Subsequently, each pot was fertilized with 0.6 g N, 6.3 g P₂O₅, 1.8 g K₂O, 22.5 mg B, 27 mg Cu, and 45 mg Zn, according to the recommendations in Bulletin 100 (Cantarella *et al.*, 2022).

Four seeds of the tomato cultivar Santa Clara were sown per pot. This cultivar was selected because it is widely cultivated in Brazil, is well adapted to greenhouse conditions, and exhibits uniform plant growth and fruit production, making it suitable for studies of plant physiology and fruit quality (Oliveira *et al.*, 2021). After germination, seedlings were thinned to ensure uniformity, leaving one plant per pot. Irrigation was provided twice daily using a sprinkler system. During this period, the average maximum and minimum temperatures were 26.2 and 16.9°C, respectively, and the mean relative humidity of 70.2%. The average solar radiation was 15.9 MJ m⁻² day⁻¹, characterizing the microclimatic conditions during the experiment.

Experimental design and treatments

The experiment was arranged in a completely randomized design with four treatments and five replicates per treatment, with each replicate consisting of one plant.

Twenty days after planting, treatments were applied. Foliar applications of FA and SA at a dose of

0.5 mmol L⁻¹, both individually and in combination, were applied to plants using a 2 L pressurized hand sprayer, with the nozzle adjusted for a fan spray, aiming to ensure uniform distribution of the solutions on the leaf surface. The spray volume was standardized to 10 mL per plant across all treatments, and applications were carried out until runoff for three consecutive days. The spray solution contained 50 µL L⁻¹ of the non-ionic surfactant Agral® as an adjuvant to improve leaf coverage and absorption. Agral® is composed of nonyl phenoxy poly(ethyleneoxy) ethanol (200 g L⁻¹) and inert ingredients (824 g L⁻¹). Control plants received only distilled water plus surfactant under the same application conditions. Each treatment consisted of five independent replications, maintaining the same conditions of application and experimental management.

Leaves and roots were collected at the final harvest from all plants within each treatment (n= 5 plants per treatment). Fruits were harvested at full maturity throughout the experimental period, and all fruits from the five plants in each treatment were collected for evaluation.

Chlorophyll and soluble sugar determination

Leaf chlorophyll extraction was performed following the method described by Lichtenthaler (1987); 0.2 g of leaf discs (5 mm) was added in 80% acetone (v/v). For quantification of total soluble sugars, 0.5 g of fresh tissue was macerated in 80% ethanol and centrifuged at 5000 rpm for 10 min. The supernatant was mixed in phenol-sulfuric solution following the Dubois *et al.* (1956) method, using glucose as the standard.

Determination of total polyphenols and flavonoids

Tomato juice was used as the sample matrix for polyphenol and flavonoid analyses. Initially, 50 g of fresh tomato fruits were washed with distilled water,

Table 1 - Chemical and granulometric characterization of the soil used in the pot experiment, including texture (sand, silt, and clay fractions) and other physical attributes

pH CaCl ₂	P (resin) mg/dm ³	O.M.S. g/kg	K ⁺ mmol/dm ³	Ca ²⁺ mmol/dm ³	Mg ²⁺ mmol/dm ³	H+Al mmol/dm ³	BS mmol/dm ³	CEC mmol/dm ³	V %
4.4	6.0	6.0	0.8	7.0	2.0	20.0	12.5	30.0	33.0
Particle size distribution (g/kg)									
			Sand 751	Silt 69	Clay 180				

weighed, and homogenized using a Philips blender operating at 3000 rpm for 2 min. The homogenized material was then filtered through a 2-mm mesh sieve to obtain a uniform sample for subsequent analyses.

The concentration of total polyphenols was determined using the Folin-Ciocalteu colorimetric method, as described by Stagos *et al.* (2012). Gallic acid served as the calibration standard, and results were expressed as micrograms of gallic acid equivalents per milliliter ($\mu\text{g mL}^{-1}$).

Flavonoid content was estimated following the procedure described by Yao *et al.* (2013). Rutin was used as the reference compound, and the results were expressed in micrograms of rutin equivalents per milliliter ($\mu\text{g mL}^{-1}$).

Carotenoid analysis

Lycopene, β -carotene, and lutein were extracted from fresh tomato samples using an extractor solution containing hexane, acetone, and ethanol (2:1:1, v/v/v) (Sadler *et al.*, 1990). The quantification was based on the equations proposed by Craft and Soares (1992), using molar extinction coefficients of $172,000 \text{ L mol}^{-1} \text{ cm}^{-1}$, $135,300 \text{ L mol}^{-1} \text{ cm}^{-1}$, and $145,100 \text{ L mol}^{-1} \text{ cm}^{-1}$, respectively.

Growth and allometric measurements

To evaluate the effects of FA and SA on plant development, several growth parameters were measured: leaf area, number of leaves and fruits per plant, plant height, shoot and root dry weight, and average fruit weight. Leaf area was analyzed using ImageJ® software (Powerful Image Analysis). The number of leaves and fruits, plant height, shoots and roots dry weight were recorded after drying the plant material in a forced-air oven at 70°C until reaching constant weight.

Allometric traits were calculated using the following equations: root-shoot ratio (RDW/SDW), specific leaf area (LA/SDW), leaf area ratio (LA/TDW), and leaf mass ratio (SDW/TDW). In these formulas, RDW represents root dry weight, SDW is shoot dry weight, LA corresponds to leaf area, and TDW refers to total dry weight.

Statistical analysis

The experiment was carried out in a completely randomized design with four treatments and five replicates per treatment. Data were analyzed using two-way ANOVA to evaluate the individual effects of FA and SA, as well as the interaction between both factors (FA $\times$ SA). Before the analyses, the assumptions of normality and homogeneity of variances were verified using the Shapiro-Wilk and Levene tests, respectively. When significant differences were observed, means were compared using Tukey's test at the 5% significance level ($p < 0.05$). Statistical analyses were performed using SISVAR (Ferreira, 2019), and the results are presented as means $\pm$ standard error. For the multivariate analyses, data were normalized and auto-scaled prior to principal component analysis (PCA) and Pearson correlation analysis, which were conducted using MetaboAnalyst (Pang *et al.*, 2024). These analyses were used to better understand the relationships among variables and the overall response patterns of the treatments.

3. Results

Ferulic acid (FA) application increased the concentrations of chlorophyll *a* (153.2%) and total chlorophyll (29.3%) compared to the control (Fig. 1 a and 1 c). The FA+SA combination increased

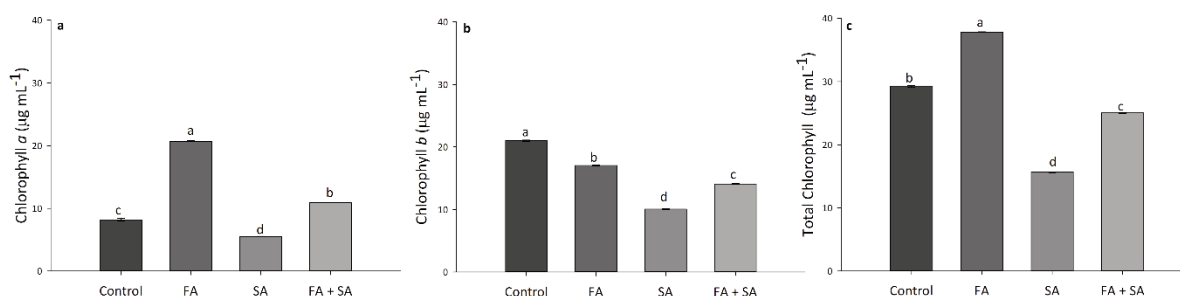


Fig. 1 - Chlorophyll *a* (a), chlorophyll *b* (b), and total chlorophyll (c) concentrations in *Solanum lycopersicum* plants treated with ferulic acid (FA, 0.5 mmol L^{-1}), salicylic acid (SA, 0.5 mmol L^{-1}), and the combination of FA and SA (0.5 mmol L^{-1}). Different letters indicate significant differences by Tukey's test ($p \leq 0.01$). Vertical bars represent the standard error.

chlorophyll *a* concentration by 33.2% compared with untreated plants (Fig. 1 a). However, chlorophyll *b* levels were reduced in all treatments relative to the control (Fig. 1 b).

Salicylic acid (SA) application increased polyphenol (61.1%) and flavonoid (26.0%) concentrations relative to untreated plants (Fig. 2 e, f). On the other hand, the application of FA showed significant increases in the concentration of total soluble sugars (147.3%), lycopene (78.9%), β -carotene (78.8%), lutein (78.7%), polyphenols (91.6%), and flavonoids (85.7%), respectively, compared to the control (Fig. 2 a-f). However, the FA+SA combination increased the concentrations of soluble sugars (152.2%), polyphenols (37.3%), and flavonoids (76.5%) relative to untreated plants (Fig. 2 a, 2 e, 2 f).

Growth parameters increased with both the isolated application of FA and SA and with the combination of elicitors. Increases of 18.7% (FA), 28.9% (SA), and 46.8% (FA+SA) were observed in the shoot dry weight, 68.9% (FA), 66.6% (SA), and 76.7% (FA+SA) in the root dry weight and 26.5% (FA), 34.8% (SA), and 51.5% (FA+SA) in the total dry weight, respectively in relation to the control (Fig. 3 a, 3 b, 3 c). However, fruit weight at SA application increased by 8.5%, and the FA+SA combination increased by 4.9%, compared with untreated plants (Fig. 3 d).

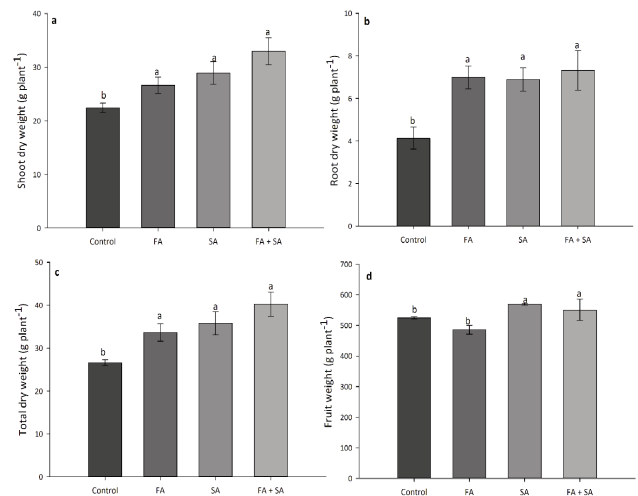


Fig. 3 - Shoot dry weight (a), root dry weight (b), total dry weight (c), and fruit weight (d) in *Solanum lycopersicum* plants treated with ferulic acid (FA, 0.5 mmol L⁻¹), salicylic acid (SA, 0.5 mmol L⁻¹), and the combination of FA and SA (0.5 mmol L⁻¹). Different letters indicate significant differences by Tukey's test ($p < 0.05$). Vertical bars represent the standard error.

Increases in leaf area, leaf number, fruit size, and fruit diameter were observed in plants treated with FA of 82.1%, 8.2%, 39.6%, and 44.1%, respectively, compared to untreated plants (Fig. 4 a, 4 b, 4 e, and 4 f). Plants treated with SA showed increases in leaf area (90.3%), plant height (12%), fruit size (32.3%),

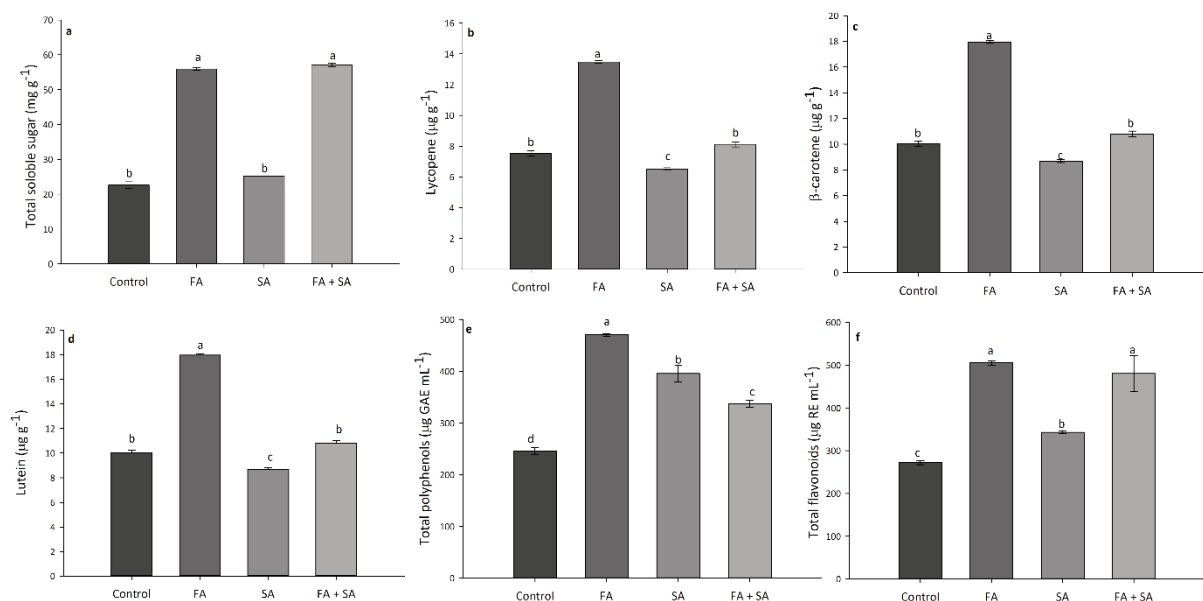


Fig. 2 - Total soluble sugar (a), lycopene (b), β -carotene (c), lutein (d), total polyphenols (e), and total flavonoids (f) concentrations in *Solanum lycopersicum* plants treated with ferulic acid (FA, 0.5 mmol L⁻¹), salicylic acid (SA, 0.5 mmol L⁻¹), and the combination of FA and SA (0.5 mmol L⁻¹). Different letters indicate significant differences by Tukey's test ($p < 0.05$). Vertical bars represent the standard error.

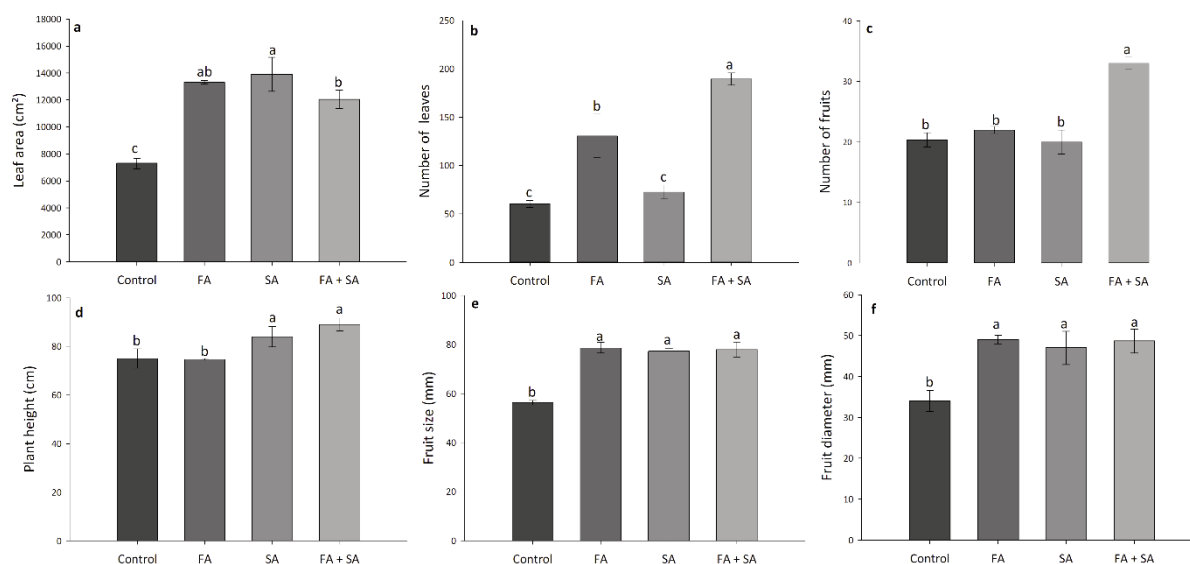


Fig. 4 - Leaf area (a), number of leaves (b), number of fruits (c), plant height (d), fruit size (e), and fruit diameter (f) in *Solanum lycopersicum* plants treated with ferulic acid (FA, 0.5 mmol L⁻¹), salicylic acid (SA, 0.5 mmol L⁻¹), and the combination of FA and SA (0.5 mmol L⁻¹). Different letters indicate significant differences by Tukey's test (p < 0.05). Vertical bars represent the standard error.

and fruit diameter (38.2%), respectively, compared to the control (Fig. 4 a, 4 d, 4 e, 4 f). The combination of FA+SA showed increases in leaf area (64.9%), number of leaves (213.8%), number of fruits (62.3%), plant height (18.7%), fruit size (38.5%), and fruit diameter (43.1%), respectively, compared to the control (Fig. 4 a-f).

FA and SA treatments significantly increased allometric traits in tomatoes (Fig. 5). The highest values were observed in the isolated and combined application of FA and SA, which resulted in an increase of 41.9%, 56% and 44.1% in the root: shoot ratio (Fig. 5 a), 68.7%, 48.4% and 18.5% in the leaf area ratio (Fig. 5 b) and 78.9%, 60.4% and 24.4% in the specific leaf area (Fig. 5 c), respectively in comparison to the untreated plants. However, the leaf mass ratio of the treated plants did not differ significantly from that of the control plants (Fig. 5 d).

In Pearson's correlations of *S. lycopersicum* under FA and SA elicitation (Fig. 6), the glow matrix highlights how the elicitors FA, SA, and their combination (FA+SA) influence different aspects of plant metabolism and development, revealing potential underlying physiological mechanisms. Under the FA application, significant associations were observed among growth, physiological, and biochemical variables (Fig. 6 a). FrD and FrS were associated with RDW, LA, and TDW (p < 0.01), suggesting that improvements in plant growth were

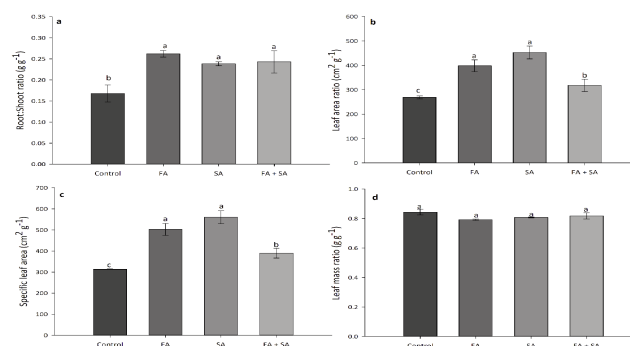


Fig. 5 - Root: shoot ratio (a), leaf area ratio (b), specific leaf area (c), and leaf mass ratio (d) in *Solanum lycopersicum* plants treated with ferulic acid (FA, 0.5 mmol L⁻¹), salicylic acid (SA, 0.5 mmol L⁻¹), and the combination of FA and SA (0.5 mmol L⁻¹). Different letters indicate significant differences by Tukey's test (p < 0.05). Vertical bars represent the standard error.

accompanied by better fruit development. FLV, TSS, LYC, BETA, and chlorophyll-related traits also showed significant associations (p < 0.001), indicating that FA influenced both photosynthetic activity and antioxidant metabolism. Chl a and Chl t were significantly associated with carotenoid accumulation (p < 0.001), reinforcing the relationship between pigment biosynthesis and fruit quality. In contrast, FrW showed few significant associations with the evaluated variables, suggesting a more isolated response compared with the other production traits.

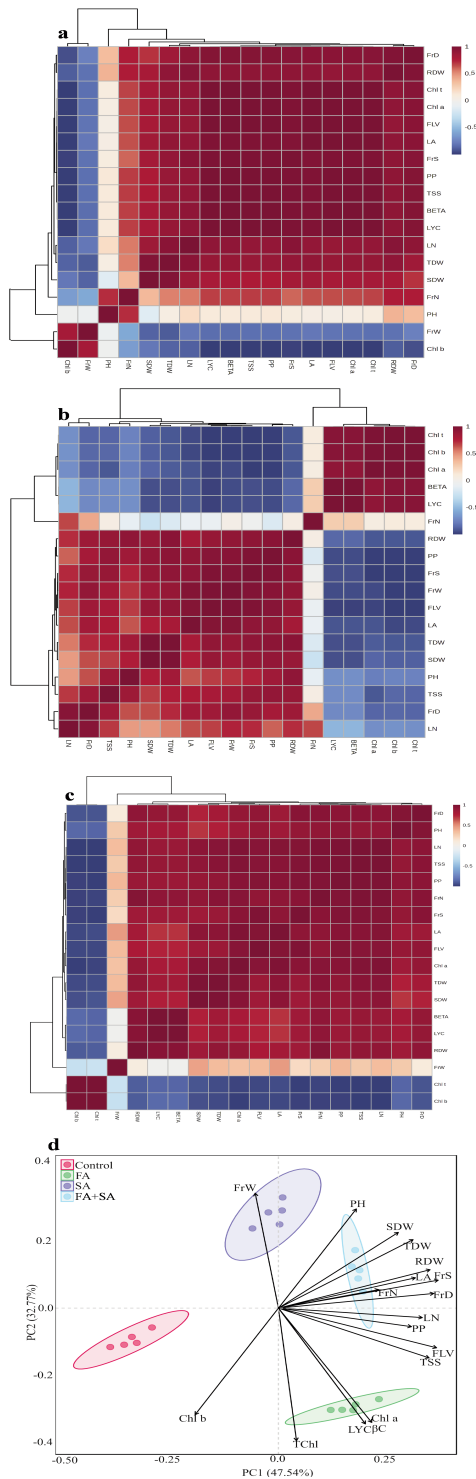


Fig. 6 - Pearson's correlations between the variables studied: (a) correlation related to FA; (b) correlation with SA; (c) joint correlation of FA+SA, and (d) principal component analysis (PCA) between treatments. Chl a (chlorophyll a), Chl b (chlorophyll b), TChl (total chlorophyll), TSS (total soluble sugar), FLV (flavonoids), PP (polyphenols), LYC (lycopene), BETA (β -carotene), SDW (shoot dry weight), RDW (root dry weight), TDW (total dry weight), FrW (fruit weight), LA (leaf area), LN (number of leaves), FrN (number of fruits), PH (plant height), FrS (fruit size), FrD (fruit diameter).

Under SA treatment (Fig. 6 b), chlorophyll variables were significantly associated with growth and biochemical traits, including FLV, TSS, RDW, and fruit-related variables ($p < 0.01$). LYC and BETA were also associated with chlorophyll contents ($p < 0.001$), indicating coordinated changes between photosynthetic pigments and antioxidant compounds. FrS and FrW were associated with FLV, LA, and TDW ($p < 0.01$), suggesting that SA application promoted simultaneous responses in plant growth and fruit quality. PH and TSS also showed significant associations with productive traits ($p < 0.05$).

For the combined FA+SA treatment (Fig. 6 c), strong associations were observed among growth, productivity, and biochemical variables. FrD, FrN, FrS, and TSS were significantly associated with chlorophyll-related traits, FLV, and biomass accumulation variables ($p < 0.001$), indicating an integrated response of plant metabolism under the combined application. LYC and BETA were associated with chlorophyll content, RDW, and TDW ($p < 0.01$), suggesting a close relationship between antioxidant metabolism and plant growth. In contrast, FrW did not show significant associations with most variables, indicating a distinct response pattern relative to the other production parameters.

When the two compounds were applied simultaneously, the correlation patterns reflected a combination of the effects observed in the individual treatments. There is a strong correlation between antioxidant compounds and growth parameters, suggesting a combined effect between FA and SA. It is observed that the correlation between biomass and antioxidant compounds is more balanced than in individual treatments, indicating that the plant may be balancing investment in defense and growth.

PCA was performed to explore the multivariate patterns among morphophysiological traits and the FA, SA, and FA+SA treatments (Fig. 6 d). The variables were distributed along two principal components that together explained 80.31% of the total variance (PC1: 47.54%; PC2: 32.77%). The control treatment was positioned in the lower-left quadrant, in close association with Chl b, indicating that untreated plants were characterized mainly by basal levels of photosynthetic pigments, without an expressive increase in biomass accumulation or fruit-related traits. Plants treated with SA were located in the upper-left quadrant and positioned near FrW, suggesting a greater contribution of this variable to the SA treatment profile and indicating enhanced

fruit production. In contrast, the FA treatment was grouped in the lower-right quadrant and positioned close to Chl a, TChl, LYC, and β C, indicating that these variables contributed strongly to the FA-associated response pattern, especially regarding pigment accumulation and fruit quality-related traits. The combined treatment (FA+SA) was distributed in the upper-right quadrant and was associated with variables related to growth and productivity, including PH, FrN, SDW, RDW, TDW, LA, FrS, and FrD. This distribution suggests that the combined application elicited broader morphophysiological responses associated with vegetative growth and reproductive performance.

4. Discussion and Conclusions

One of the most pronounced responses observed was the increase in photosynthetic pigments, particularly chlorophyll a and total chlorophyll, mainly in plants treated with FA and FA+SA (Fig. 1 a and c). Increased chlorophyll content is commonly associated with greater photosynthetic efficiency and improved carbon assimilation, which directly supports vegetative growth and reproductive development. Similar responses were reported by Linić *et al.* (2021), who observed improved photosynthetic performance in Chinese cabbage treated with SA and FA under stress conditions. In addition, FA has been associated with the maintenance of photosynthetic apparatus stability and protection against reactive oxygen species (Fiorito *et al.*, 2024; Khan *et al.*, 2024). Previous studies on tomato plants treated with 1 mmol L⁻¹ FA and SA also reported increases in chlorophyll accumulation (Gorni *et al.*, 2021 a). However, the present study demonstrates that similar physiological responses can be achieved using a lower concentration (0.5 mmol L⁻¹), suggesting that reduced doses are sufficient to stimulate pigment biosynthesis without imposing high metabolic costs. This finding is relevant because higher elicitor concentrations may overstimulate defense pathways and compromise plant growth under some conditions (Queiroz *et al.*, 2023).

The increase in chlorophyll content was accompanied by higher accumulation of soluble sugars in plants treated with FA and FA+SA (Fig. 2 a). This relationship is expected because chlorophyll

accumulation improves light capture and photosynthetic activity, increasing carbon fixation and carbohydrate production (Fiorito *et al.*, 2024; Khan *et al.*, 2024). Similar increases in chlorophyll and carbohydrate contents after FA and SA application were also observed by Linić *et al.* (2021) and Gorni *et al.* (2021 b). Nevertheless, our results suggest that FA-containing treatments promoted a more balanced relationship between photosynthetic metabolism and biomass production, reinforcing the idea that FA may act directly on primary metabolism rather than exclusively on stress signaling pathways.

Also evident was the accumulation of carotenoids such as lycopene, β -carotene, and lutein, as well as flavonoids and polyphenols (Fig. 2 b-f). These compounds are associated with antioxidant activity and fruit nutritional quality, and their accumulation indicates activation of secondary metabolism. Similar responses have been reported in tomato fruits treated with phenolic elicitors (Gorni *et al.*, 2021 a; Shah *et al.*, 2021). However, the stronger responses observed in FA-containing treatments suggest that FA may have a more direct role in modulating the phenylpropanoid pathway and carbon allocation toward antioxidant biosynthesis. As a hydroxycinnamic acid derivative, FA participates in cell wall metabolism and can influence enzymes associated with phenolic synthesis (Antonopoulou *et al.*, 2022). This mechanism may explain the simultaneous increase in carotenoids, flavonoids, and biomass accumulation observed in the present study.

Gorni *et al.* (2021 a) and Linić *et al.* (2021) previously demonstrated positive effects of SA and FA on phenolic metabolism in tomato plants. However, most previous studies evaluated the isolated action of these compounds or used higher concentrations. In the present study, the combined application at 0.5 mmol L⁻¹ led to simultaneous increases in photosynthetic pigments, bioactive compounds, and growth-related traits, suggesting that lower doses may be sufficient to induce coordinated physiological responses. Rather than suggesting a purely additive effect, the results indicate that FA and SA may act through complementary metabolic pathways. SA is primarily associated with defense signaling and regulation of oxidative stress, whereas FA appears to be more closely linked to carbon metabolism, cell expansion, and fruit development.

In physiological terms, SA acts as an important

signaling molecule involved in abiotic stress responses and antioxidant metabolism, favoring the synthesis of flavonoids and phenolic compounds (Ali, 2021; Ramos-Sotelo and Figueroa-Pérez, 2023). Its action also includes regulation of photosynthetic activity and protection of the photosynthetic apparatus (Aires *et al.*, 2022; Saleem and Fariduddin, 2024). SA-mediated signaling is strongly associated with ROS homeostasis and activation of defense-related pathways (Pacheco and Gorni, 2021). However, activation of defense metabolism often requires high energetic investment, which can reduce carbon allocation to growth and reproduction (Watts *et al.*, 2023). This may explain why SA-treated plants showed weaker associations with biomass and fruiting variables compared with FA-treated plants.

In contrast, FA appears to act more directly on primary metabolism and developmental processes. FA has been associated with cell division, tissue elongation, and fruit development (Niharika *et al.*, 2021; Antonopoulou *et al.*, 2022). In addition, its participation in cell wall biosynthesis and enzymatic regulation may contribute to greater biomass accumulation and improved reproductive development. Unlike SA, whose effects are more closely linked to defense signaling, FA was associated more strongly with vegetative growth and fruit-related parameters, suggesting distinct physiological roles for the two elicitors.

The correlation analyses reinforced these differences between elicitors (Fig. 6 a-c). SA was predominantly associated with antioxidant protection and stress-related responses, whereas FA was more strongly associated with biomass accumulation and reproductive development. These results support the physiological trade-off between defense and growth proposed by Watts *et al.* (2023). Interestingly, the combined FA+SA treatment promoted a broader response pattern, with simultaneous gains in growth, pigment accumulation, soluble solids, and antioxidant compounds, suggesting a more integrated metabolic adjustment.

PCA further supported this interpretation by grouping FA+SA-treated plants in a region associated with multiple agronomic and biochemical variables (Fig. 6 d). This distribution indicates that the combined application promoted a more balanced interaction between primary and secondary metabolism, allowing plants to maintain growth while activating protective mechanisms. Similar

integrated responses between growth and defense metabolism have been discussed by Pang *et al.* (2024) and Fiorito *et al.* (2024) in studies involving phenolic signaling compounds.

Finally, the dose used in the present study (0.5 mmol L⁻¹) proved effective, with no visible phytotoxic effects. Compared with the previous study using 1 mmol L⁻¹ (Gorni *et al.*, 2021 a), the lower concentration promoted comparable increases in photosynthetic pigments and antioxidant-related compounds while maintaining balanced growth and productivity responses.

These findings reinforce the importance of dose adjustment in elicitor-based strategies, as excessive stimulation of defense pathways may impair plant metabolism and carbon allocation (Queiroz *et al.*, 2023). Overall, the results indicate that low-dose applications of FA and SA represent a promising approach to improving both the agronomic performance and nutritional quality of tomato fruits.

In conclusion, this study demonstrates that the combined application of ferulic acid (FA) and salicylic acid (SA) at 0.5 mmol L⁻¹ promoted simultaneous improvements in plant growth and in the accumulation of nutritionally important compounds in tomato fruits. These findings suggest that the combined use of these phenolic compounds can effectively enhance both yield-related and quality-related traits in tomato plants. Overall, FA and SA may represent a promising and sustainable agronomic strategy to improve fruit quality and plant performance, although further studies under different environmental conditions and cultivation systems are needed to confirm the consistency and applicability of these effects.

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