

Effects of AMINOSA on PR2 and PR3 Enzyme Activities in Marigold (*Tagetes erecta*) Seedlings

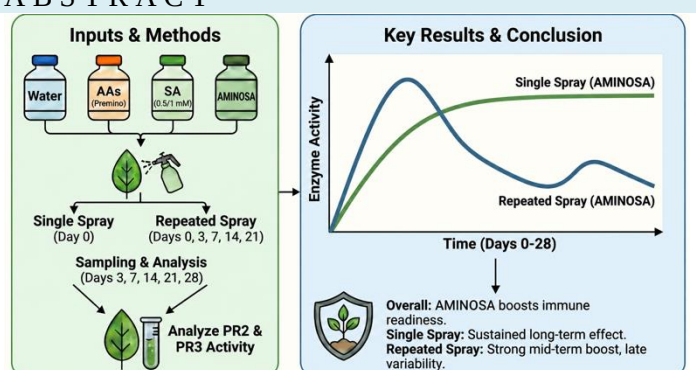
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ABSTRACT



This preliminary biochemical screening study assessed whether AMINOSA, a combined formulation of amino acids (27% w/v; AAs) and salicylic acid (SA), alters PR2 (β -1,3-glucanase) and PR3 (chitinase) activity trends in marigold seedlings. Twenty-one-day-old *Tagetes erecta* L. cv. 'Cana Yellow' seedlings were foliar-sprayed with water, AAs, SA (0.5 or 1 mM), or AMINOSA at the corresponding SA concentrations

under either a single-spray program (sampling at 3, 7, 14, 21, and 28 days) or a repeated-spray program (sprays at 0, 3, 7, 14, and 21 days; sampling at 7, 14, 21, and 28 days), giving 46 treatment combinations. Leaves from 10 plants per treatment were pooled into one composite sample at each time point. The total protein content was quantified by BCA assay, and PR2 and PR3 activities were measured colorimetrically and expressed as U/mg protein. AMINOSA, especially with 0.5 mM SA, tended to produce higher PR2 and PR3 activity trends than AAs alone and water. Repeated spraying produced stronger mid-term PR2 peaks but greater late-stage variability. As each treatment-timepoint was represented by a single pooled composite sample without independent replicated means, the data were interpreted descriptively as trend-level biochemical profiling and were not subjected to inferential statistical analysis. AMINOSA with 0.5 mM SA under single spraying emerged as a promising option for sustaining later-stage PR2/PR3 trends, whereas repeated spraying may be further evaluated for a stronger mid-term PR2 response. These findings suggested that time-course PR2/PR3 profiling may help select candidate formulations and spray programs. The next step is replicated pathogen-challenge validation, followed by greenhouse or field confirmation.

Keywords: 3-glucanase, Amino acids, Chitinase, Marigold, Salicylic acid, β -1

1. Introduction

Marigold (*Tagetes erecta* L.) is a high-value ornamental crop cultivated widely for cut flowers and landscape use. However, its production is frequently constrained by fungal and soil-borne diseases that reduce seedling vigor, delay growth, and ultimately lower flower quality (Abdel-Wahed & Abdel Rahman, 2022; Sharma et al., 2025). Although chemical control remains common, reliance on fungicides alone is increasingly limited by cost and the risk of resistance development, reinforcing the need for safer and more sustainable disease-management options (Corkley et al., 2022). Consequently, approaches that enhance plant immune readiness using biostimulants and defense elicitors are of practical interest for integrated marigold production (Reglinski et al., 2023).

Amino acid-based products are widely used in agriculture due to their high nutritional value and their potential to support physiological and metabolic functions related to growth and stress tolerance.

As biostimulants, amino acids and protein hydrolysates can influence plant development through improved nutrient uptake, enhanced chlorophyll synthesis, osmotic adjustment, antioxidant activity, and hormone-like signaling interactions (Rouphael & Colla, 2020; Sible et al., 2021; Navarro-León et al., 2022; Sun et al., 2024). The amino acid product AMINOSA used in this study has a broad amino acid profile, including glutamic acid, proline, glycine, arginine, tryptophan, methionine, aspartic acid, and lysine.

Salicylic acid (SA) is a central phytohormone in plant immunity, particularly in systemic acquired resistance and defense priming. Exogenous SA can enhance plant responsiveness to pathogen attack and has shown practical potential in crop protection and stress management. For example, SA treatment enhanced disease resistance in *Panax vietnamensis* through the regulation of secondary metabolite production (Hou et al., 2024), and field priming with exogenous SA improved antioxidant-related responses under production conditions (Thepbandit et al., 2024). However, the effectiveness of exogenous SA depends strongly on dose, timing, and plant physiological status, and excessive SA may impose growth-defense trade-offs (Ding & Ding, 2020; van Butselar & Van den Ackerveken, 2020; Spoel & Dong, 2024; Salinas et al., 2025).

Pathogenesis-related (PR) proteins are widely used as biochemical markers of induced defense responses. In particular, PR2 (β -1,3-glucanase) and PR3 (chitinase) are relevant because their enzymatic activities contribute to antifungal defense and are often associated with SA-related signaling (Ali et al., 2018; dos Santos & Franco, 2023). Measuring PR2 and PR3 activities therefore provides a practical biochemical readout that complements symptom-based evaluation and supports mechanistic interpretation of treatment effects.

Despite the established roles of SA signaling, PR proteins, and amino acid-based biostimulants, time-course information on PR2 and PR3 responses after SA supplementation in an amino acid formulation remains limited in marigold, especially when comparing single- and repeated-spray programs. This gap hinders evidence-based selection of formulation–application combinations for subsequent biological validation.

Accordingly, this study aimed to determine whether AMINOSA, a formulation combining amino acids (AAs) and SA, alters PR2 (β -1,3-glucanase) and PR3 (chitinase) activity trends in marigold seedlings across SA concentrations and sampling times under single- and repeated-spray programs. It was based on pooled composite samples, this work was designed as a preliminary biochemical screening study, and the results should therefore be interpreted as trend-level biochemical profiling. These findings may help guide formulation and spray-program selection and prioritize candidate treatments for subsequent validation in more rigorous biological assays and under production-relevant conditions.

2. Research method

2.1 Research materials

Marigold seedlings (*Tagetes erecta* L. cv. ‘Cana Yellow’) at 21 days old were used as plant material. AMINOSA was defined as a combined amino acid–salicylic acid formulation, consisting of an amino acid component (27% w/v; coded as AAs) supplemented with SA. The amino acid component was applied at 1 mL/L water, and SA was prepared at final concentrations of 0.5 and 1 mM. The combined treatments are hereafter referred to as AMINOSA 0.5 mM and AMINOSA 1 mM according to the final SA concentration. Total proteins were extracted using the Minute™ Total Protein Extraction Kit for Plant Tissues (SN-009) supplemented with Halt™ Protease Inhibitor Cocktail (EDTA-free, 1×). Protein concentration was determined using the Pierce™ BCA Protein Assay. PR2 (β -1,3-glucanase) and PR3 (chitinase) activities were measured using the Abbexa Beta-1,3-Glucanase Assay Kit (abx097994) and Abbexa Chitinase Assay Kit (abx298854), respectively.

2.2 Research procedure

The experiment comprised 46 treatment-timepoint combinations defined by spray solution, spray program, and sampling day. In the single-spray program, seedlings received one application at day 0 and were sampled at 3, 7, 14, 21, and 28 days after the first spray. The treatments in this program were water, AAs, SA 0.5 mM, SA 1 mM, AMINOSA 0.5 mM, and AMINOSA 1 mM, giving 30 treatment–timepoint combinations. In the repeated-spray program, seedlings were sprayed at 0, 3, 7, 14, and 21 days after the first spray and sampled at 7, 14, 21, and 28 days. Treatments in this program were SA 0.5 mM, SA 1 mM, AMINOSA 0.5 mM, and AMINOSA 1 mM, giving 16 treatment–timepoint combinations. The full treatment structure was enumerated in Table 1.

Table 1. Enumeration of the 46 treatment-timepoint combinations used in the preliminary biochemical screening study.

No.	Spray program	Spray solution	SA concentration	Spray day(s) after first spray	Sampling day after first spray
1	Single spray	Water	-	0	3
2	Single spray	Water	-	0	7
3	Single spray	Water	-	0	14
4	Single spray	Water	-	0	21
5	Single spray	Water	-	0	28
6	Single spray	AAs	-	0	3
7	Single spray	AAs	-	0	7
8	Single spray	AAs	-	0	14
9	Single spray	AAs	-	0	21
10	Single spray	AAs	-	0	28
11	Single spray	SA	0.5 mM	0	3
12	Single spray	SA	0.5 mM	0	7
13	Single spray	SA	0.5 mM	0	14
14	Single spray	SA	0.5 mM	0	21
15	Single spray	SA	0.5 mM	0	28
16	Single spray	SA	1 mM	0	3
17	Single spray	SA	1 mM	0	7
18	Single spray	SA	1 mM	0	14
19	Single spray	SA	1 mM	0	21
20	Single spray	SA	1 mM	0	28
21	Single spray	AMINOSA	0.5 mM	0	3
22	Single spray	AMINOSA	0.5 mM	0	7
23	Single spray	AMINOSA	0.5 mM	0	14
24	Single spray	AMINOSA	0.5 mM	0	21
25	Single spray	AMINOSA	0.5 mM	0	28
26	Single spray	AMINOSA	1 mM	0	3
27	Single spray	AMINOSA	1 mM	0	7
28	Single spray	AMINOSA	1 mM	0	14
29	Single spray	AMINOSA	1 mM	0	21
30	Single spray	AMINOSA	1 mM	0	28
31	Repeated spray	SA	0.5 mM	0, 3	7
32	Repeated spray	SA	0.5 mM	0, 3, 7	14
33	Repeated spray	SA	0.5 mM	0, 3, 7, 14	21
34	Repeated spray	SA	0.5 mM	0, 3, 7, 14, 21	28
35	Repeated spray	SA	1 mM	0, 3	7
36	Repeated spray	SA	1 mM	0, 3, 7	14
37	Repeated spray	SA	1 mM	0, 3, 7, 14	21

No.	Spray program	Spray solution	SA concentration	Spray day(s) after first spray	Sampling day after first spray
38	Repeated spray	SA	1 mM	0, 3, 7, 14, 21	28
39	Repeated spray	AMINOSA	0.5 mM	0, 3	7
40	Repeated spray	AMINOSA	0.5 mM	0, 3, 7	14
41	Repeated spray	AMINOSA	0.5 mM	0, 3, 7, 14	21
42	Repeated spray	AMINOSA	0.5 mM	0, 3, 7, 14, 21	28
43	Repeated spray	AMINOSA	1 mM	0, 3	7
44	Repeated spray	AMINOSA	1 mM	0, 3, 7	14
45	Repeated spray	AMINOSA	1 mM	0, 3, 7, 14	21
46	Repeated spray	AMINOSA	1 mM	0, 3, 7, 14, 21	28

Seedlings were maintained in an indoor controlled laboratory environment under a 12 h light/12 h dark photoperiod at 25–28°C. During cultivation, plants were kept in a randomized arrangement within the growing area. To minimize spray drift and cross-contamination during treatment application, plants from each treatment group were transferred to separate trays only at the time of spraying, and each solution was applied using a dedicated hand sprayer. Trays were positioned sufficiently far apart to minimize spray drift between treatments. After application, plants were kept separated until the leaf surfaces were dry and were then returned to the growing area in the randomized arrangement. Spray solutions were applied at 1 mL per plant with uniform foliar coverage.

At each sampling date, leaves 1–6 counted from the lowerest leaves were collected from 10 plants per treatment and pooled to form one composite sample per treatment per timepoint. Thus, pooling was performed as 10 plants $\times$ 1 treatment $\times$ 1 timepoint = 1 pooled composite sample. Samples were ground in liquid nitrogen, and total protein was extracted according to the SN-009 protocol with protease inhibitor supplementation. Protein concentration was quantified by BCA assay at 562 nm using a microplate reader and used to normalize PR2 and PR3 enzyme activities. Enzyme assays were performed according to the manufacturers' instructions, with absorbance measured at 540 nm for β -1,3-glucanase and 585 nm for chitinase. Activities were calculated using the kit-provided formulas and standards and are reported as U/mg protein.

No technical replicates were performed during enzyme assays. Therefore, each pooled composite sample generated one analytical value for total protein, PR2 activity, and PR3 activity. Because the dataset lacked independent replicated means, it was not subjected to inferential statistical analysis. Instead, results were interpreted descriptively as trend-level biochemical profiling to identify promising treatment and spray-program candidates for subsequent replicated pathogen-challenge experiments and later greenhouse or field validation.

3. Results and discussion

3.1 PR2 (β -1,3-glucanase) response under the single-spray program

Under the single-spray program (Figure 1), the water control provided a relatively stable baseline trend for PR2 activity, with a transient dip at 14 days. Amino acids (AAs) showed PR2 activity trends similar to those of water across sampling dates, indicating that AAs alone produced limited PR2 induction under the present conditions. Such context-dependent responses are commonly reported for biostimulant inputs across products, crops, and application conditions (Rouphael & Colla, 2020; Sible et al., 2021). In contrast, the SA treatments showed clearer PR2 induction trends than both water and AAs. SA 0.5 mM increased from early to mid sampling dates and tended to peak around 14–21 days after spraying, whereas SA 1 mM showed a weaker early trend with a later rise toward 21–28 days. This pattern is consistent with dose- and timing-dependent SA signaling with regulatory feedback (Ding & Ding, 2020; van Butselaar & Van den Ackerveken, 2020; Spoel & Dong, 2024; Salinas et al., 2025). Among the combined treatments, AMINOSA 0.5 mM maintained elevated PR2 trends from 7 to 21 days

and remained high at 28 days, indicating a more sustained PR2-associated response than AAs alone. AMINOSA 1 mM showed a gradual increase over time and approached AAs levels by 28 days, but it did not consistently exceed AAs at the mid sampling dates. These trends suggest that the AMINOSA formulation may have contributed to maintenance of PR2-associated responses under the moderate SA dose, whereas this tendency was less evident at the higher dose. This interpretation is biologically plausible because amino acid-based biostimulants can modulate plant physiological performance and stress responses in a context-dependent manner, while SA responses themselves are strongly influenced by dose and timing (Ding & Ding, 2020; Rouphael & Colla, 2020; Sible et al., 2021; Spoel & Dong, 2024). However, this possible mechanistic contribution remains to be verified directly in marigold. Overall, PR2 trends indicate that 0.5 mM SA drove the main increase, and its combination with amino acids in AMINOSA may have supported maintenance of elevated PR2 toward later time points.

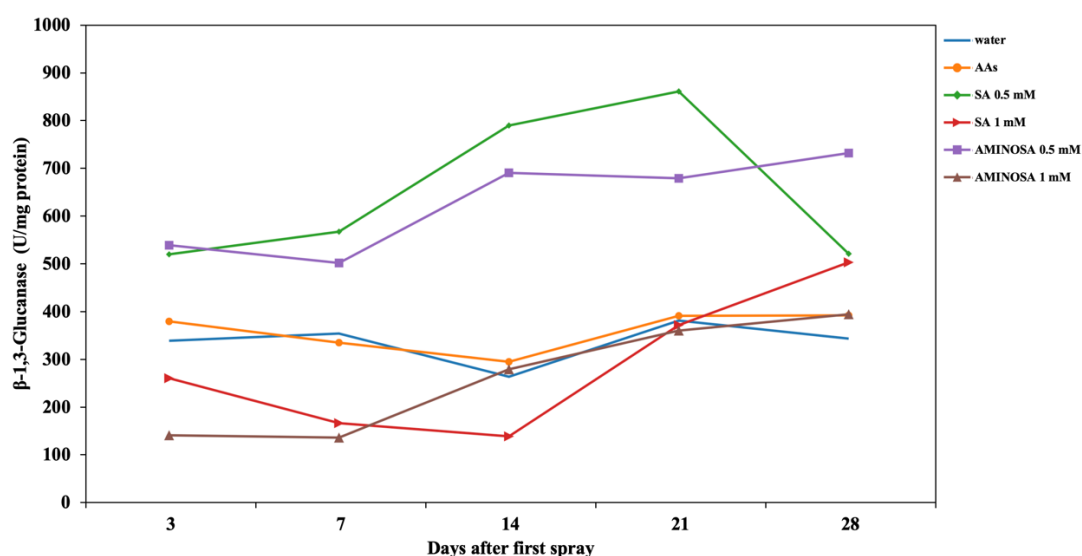


Figure 1. Time-course of β -1,3-glucanase (PR2) activity (U/mg protein) in marigold seedlings under the single-spray program.

3.2 PR3 (chitinase) response under the single-spray program

Under the single-spray program (Figure 2), the water control provided the baseline trend for PR3 activity. Amino acids (AAs) showed PR3 activity trends slightly higher than those of water across the time course, indicating a mild PR3-associated response under the present conditions. In contrast, SA-containing treatments produced larger PR3 shifts than AAs alone. SA 0.5 mM showed high PR3 trends at 3-14 days, followed by a marked dip at 21 days and a rebound by day 28. SA 1 mM showed a high early PR3 level at day 3 but declined progressively toward later sampling dates, reaching a low level by day 28. This pattern is consistent with dose- and timing-dependent SA signaling, in which stronger or prolonged SA inputs may be followed by attenuation and potential defense-associated costs (van Butselar & Van den Ackerveken, 2020; Spoel & Dong, 2024; Salinas et al., 2025). Among combined treatments, AMINOSA 0.5 mM showed high PR3 trends at 3 and 14 days, dipped at 7 and 21 days, and remained elevated at day 28, whereas AMINOSA 1 mM declined steadily after day 3 and showed the lowest late-stage PR3 level. Compared with PR2, the PR3 time course appeared more dynamic, showing clearer dip-and-rebound or progressive decline patterns across treatments. This may suggest that PR3-associated activity showed greater temporal variation in relation to treatment timing and SA dose, such that early induction was not always maintained through the later sampling dates. Such temporal fluctuation is consistent with the plasticity of defense-related protein responses under induced-resistance conditions (Ali et al., 2018; dos Santos & Franco, 2023; Reglinski et al., 2023). Within this more variable pattern, the later elevation retained under AMINOSA 0.5 mM is consistent with the possibility that amino acids in the formulation helped support maintenance of PR3-associated activity after the initial

SA-triggered response (Ding & Ding, 2020; Rouphael & Colla, 2020; Sible et al., 2021; Spoel & Dong, 2024), although this possible contribution remains to be verified directly in marigold. Overall, these PR3 trends indicate that moderate SA (0.5 mM), particularly in AMINOSA, tended to support higher PR3 levels at later sampling dates than SA 0.5 mM alone.

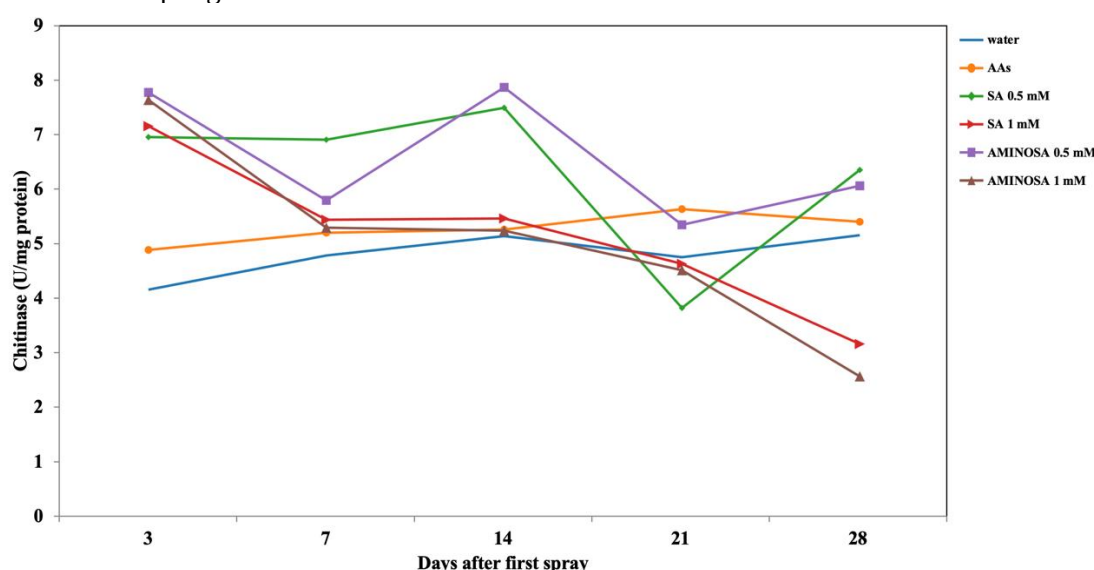


Figure 2. Time-course of chitinase (PR3) activity (U/mg protein) in marigold seedlings under the single-spray program.

3.3 Effects of repeated spraying on temporal patterns of PR2 and PR3 activities

Repeated spraying altered the time-course trends of PR2 and PR3 compared with single spraying (Figures 3-4), and the direction of change depended on whether SA was applied alone or in combination with AAs and on SA concentration.

For PR2 (Figure 3), repeated spraying at SA 0.5 mM produced a sharper mid-term elevation, peaking at day 14, than the corresponding single-spray program, but PR2 then declined markedly toward days 21-28. In contrast, SA 0.5 mM (single) increased more gradually, reached a later maximum around day 21, and remained higher at day 28 than the repeated program. At SA 1 mM, repeated spraying generated a pronounced trough at day 14 followed by a transient rise around day 21, whereas the single-spray program showed a later increase toward days 21-28, indicating that repeated application did not consistently enhance PR2 at the higher SA dose.

For PR3 (Figure 4), repeated spraying at SA 0.5 mM reduced the late-stage rebound observed under single spraying and maintained mid-range activity at days 21-28, whereas SA 1 mM (repeated) maintained relatively steady mid-range PR3 trends compared with the clear late decline observed under SA 1 mM (single). These patterns are consistent with dose- and timing-dependent SA signaling, in which repeated stimulation can amplify responses at specific windows but may also be followed by attenuation and defense-associated costs (Ding & Ding, 2020; Spoel & Dong, 2024).

For PR2 (Figure 3), repeated spraying with AMINOSA 0.5 mM produced very high early-to-mid PR2 trends (7-14 days), exceeding the corresponding single-spray levels, but it declined sharply at day 21 with only partial recovery by day 28. In contrast, AMINOSA 0.5 mM (single) maintained elevated PR2 trends from mid to late time points and remained high at day 28, suggesting a more sustained late-stage PR2 trend under single spraying. At AMINOSA 1 mM, repeated spraying did not improve PR2 relative to single spraying and tended to show lower late-stage levels.

For PR3 (Figure 4), repeated AMINOSA 0.5 mM showed larger oscillations than the single program, including a pronounced drop at day 14 and further fluctuations through day 28, whereas AMINOSA 0.5 mM (single) maintained comparatively higher PR3 trends at later time points. Repeated AMINOSA 1 mM also showed a deep mid-term reduction and only partial recovery, while the single program declined steadily toward day 28.

This greater late-stage variability may reflect non-linear response dynamics after repeated elicitation rather than a simple cumulative strengthening effect. Repeated SA exposure may create a limited window of enhanced biochemical responsiveness, after which defense-associated enzyme output becomes less stable because of attenuation, feedback regulation, and shifts in growth–defense balance over time (Ding & Ding, 2020; van Butselaar & Van den Ackerveken, 2020; Hönig et al., 2023; Spoel & Dong, 2024). The stronger fluctuation observed in some AMINOSA treatments also suggests that formulation context may influence temporal response stability, although this interpretation remains provisional because each treatment–timepoint was represented by one pooled composite sample.

Taken together, repeated spraying tended to boost mid-term enzyme activity, most clearly for PR2 around day 14 in several 0.5 mM programs. However, it also tended to increase late-stage variability (days 21–28), especially in AMINOSA treatments, suggesting that repeated application may not sustain elevated PR2/PR3 trends across the full-time course.

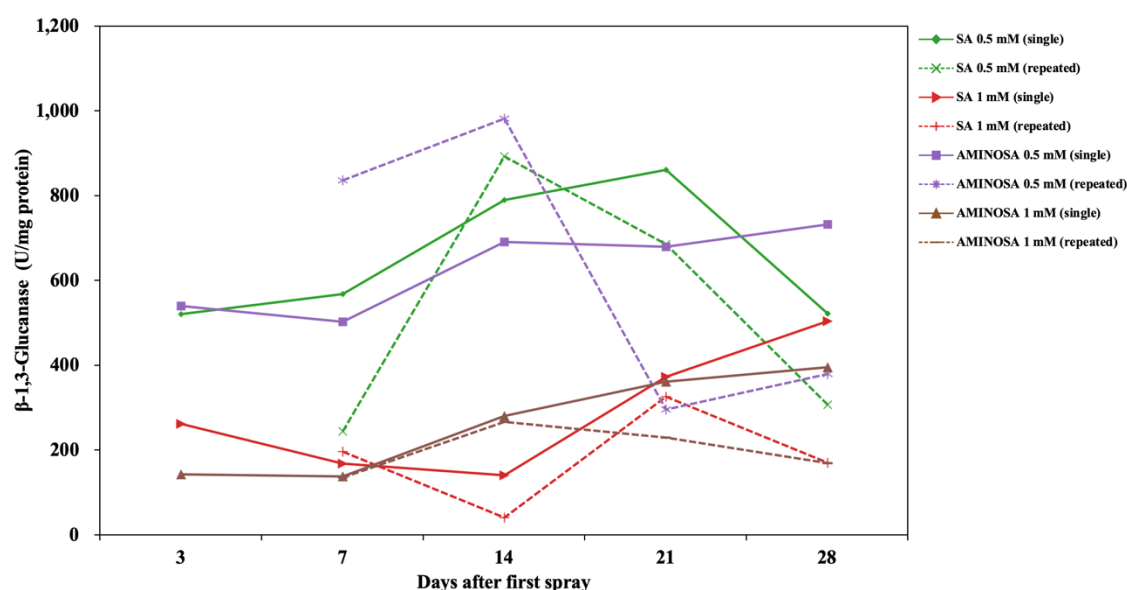


Figure 3. Comparison of PR2 activity (U/mg protein) between single and repeated spray programs in SA-containing treatments.

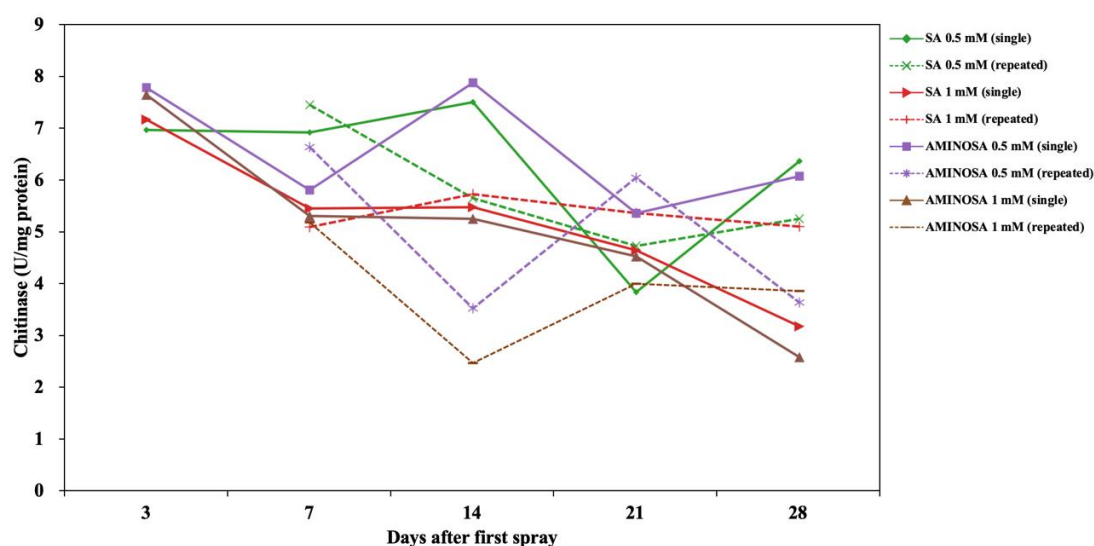


Figure 4. Comparison of PR3 activity (U/mg protein) between single and repeated spray programs in SA-containing treatments.

3.4 Implications for formulation and spray scheduling

Based on the time-course trends in Sections 3.1-3.3 (Figures 1-4), 0.5 mM SA consistently emerged as the more favorable dose than 1 mM for supporting PR2/PR3 activity trends, aligning with the concentration- and timing-dependent nature of SA responses and the potential for attenuation or defense-associated costs at stronger inputs (Ding & Ding, 2020; van Butselar & Van den Ackerveken, 2020; Spoel & Dong, 2024; Salinas et al., 2025). At this moderate dose, AMINOSA 0.5 mM was the most practical formulation candidate because it tended to sustain higher late-stage PR2/PR3 trends than SA alone, consistent with formulation- and context-dependent biostimulant performance (Rouphael & Colla, 2020; Sible et al., 2021; Sun et al., 2024).

For spray scheduling, repeated spraying can be considered when the goal is to maximize mid-term induction windows, notably PR2 around day 14, consistent with priming-like responses after re-stimulation (Mauch-Mani et al., 2017; Hönig et al., 2023). However, it may also increase late-stage variability, particularly in AMINOSA programs. Therefore, a pragmatic approach is to prioritize single-spray AMINOSA 0.5 mM when sustained later trends are desired, while reserving repeated spraying for situations where a stronger mid-term boost is required. As samples were pooled per treatment and time point, these implications should be treated as trend-level guidance to refine dose and intervals before replicated sampling and pathogen-challenge validation using PR2/PR3 as functional biochemical markers (Ali et al., 2018; dos Santos & Franco, 2023; Reglinski et al., 2023).

4. Conclusion

This study evaluated whether AMINOSA, a formulation combining amino acids (AAs) and salicylic acid (SA), shifts PR2 (β -1,3-glucanase) and PR3 (chitinase) activity trends in marigold seedlings under single- and repeated-spray programs using pooled composite samples. Overall, AMINOSA 0.5 mM was associated with higher PR2/PR3 activity trends than AAs alone. This pattern is consistent with stronger defense-associated enzyme responses during the seedling stage under the conditions tested. In terms of scheduling, repeated spraying was most useful for enhancing mid-term PR2 induction (around day 14) but was less suitable for maintaining stable late-stage trends (days 21-28) due to greater variability, particularly in AMINOSA treatments. The observed trend-level responses provide a basis for selecting candidate formulations and spray schedules to be taken forward into replicated pathogen-challenge experiments and future greenhouse/field validation under production-relevant conditions.

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References

- Abdel-Wahed, G. A., & Abdel-Rahman, T. F. M. (2022). Evaluation the activities of wood vinegar and some commercial bio-products in management of soil-borne fungi of two Tagetes varieties. **Egyptian Journal of Phytopathology**, 50(2), 104–123. <https://doi.org/10.21608/ejp.2022.173645.1075>
- Ali, S., Ganai, B. A., Kamili, A. N., Bhat, A. A., Mir, Z. A., Bhat, J. A., Tyagi, A., Islam, S. T., Mushtaq, M., Yadav, P., Rawat, S., & Grover, A. (2018). Pathogenesis-related proteins and peptides as promising tools for engineering plants with multiple stress tolerance. **Microbiological Research**, 212–213, 29–37. <https://doi.org/10.1016/j.micres.2018.04.008>
- Corkley, I., Fraaije, B., & Hawkins, N. (2022). Fungicide resistance management: Maximizing the effective life of plant protection products. **Plant Pathology**, 71(1), 150–169. <https://doi.org/10.1111/ppa.13467>
- Ding, P., & Ding, Y. (2020). Stories of salicylic acid: A plant defense hormone. **Trends in Plant Science**, 25(6), 549–565. <https://doi.org/10.1016/j.tplants.2020.01.004>

- dos Santos, C., & Franco, O. L. (2023). Pathogenesis-related proteins (PRs) with enzyme activity activating plant defense responses. **Plants**, 12(11), 2226. <https://doi.org/10.3390/plants12112226>
- Hönig, M., Roeber, V. M., Schmölling, T., & Cortleven, A. (2023). Chemical priming of plant defense responses to pathogen attacks. **Frontiers in Plant Science**, 14, 1146577. <https://doi.org/10.3389/fpls.2023.1146577>
- Hou, J., Ai, M., Li, J., Cui, X., Liu, Y., & Yang, Q. (2024). Exogenous salicylic acid treatment enhances the disease resistance of *Panax vietnamensis* by regulating secondary metabolite production. **Frontiers in Plant Science**, 15, 1428272. <https://doi.org/10.3389/fpls.2024.1428272>
- Mauch-Mani, B., Baccelli, I., Luna, E., & Flors, V. (2017). Defense priming: An adaptive part of induced resistance. **Annual Review of Plant Biology**, 68, 485–512. <https://doi.org/10.1146/annurev-arplant-042916-041132>
- Navarro-León, E., López-Moreno, F. J., Borda, E., Marín, C., Sierras, N., Blasco, B., & Ruiz, J. M. (2022). Effect of L-amino acid-based biostimulants on nitrogen use efficiency (NUE) in lettuce plants. **Journal of the Science of Food and Agriculture**, 102(15), 7098–7106. <https://doi.org/10.1002/jsfa.12071>
- Reglinski, T., Havis, N., Rees, H. J., & de Jong, H. (2023). The practical role of induced resistance for crop protection. **Phytopathology**, 113(4), 719–731. <https://doi.org/10.1094/PHYTO-10-22-0400-IA>
- Rouphael, Y., & Colla, G. (2020). Editorial: Biostimulants in agriculture. **Frontiers in Plant Science**, 11, 40. <https://doi.org/10.3389/fpls.2020.00040>
- Salinas, P., Velozo, S., & Herrera-Vásquez, A. (2025). Salicylic acid accumulation: Emerging molecular players and novel perspectives on plant development and nutrition. **Journal of Experimental Botany**, 76(7), 1950–1969. <https://doi.org/10.1093/jxb/erae309>
- Sharma, S., Das, A., Bharti, Singh, K. P., Panwar, S., Talukdar, A., Arzoo, E., Singh, A., Bisht, P., & Bhatia, R. (2025). Assessing resistance against *Alternaria* leaf spot in marigold genotypes (*Tagetes* spp.): A comprehensive *in vitro* and *in vivo* study. **Journal of Horticultural Sciences**, 20(2). <https://doi.org/10.24154/jhs.v20i2.3846>
- Sible, C. N., Seebauer, J. R., & Below, F. E. (2021). Plant biostimulants: A categorical review, their implications for row crop production, and relation to soil health indicators. **Agronomy**, 11(7), 1297. <https://doi.org/10.3390/agronomy11071297>
- Spoel, S. H., & Dong, X. (2024). Salicylic acid in plant immunity and beyond. **The Plant Cell**, 36(5), 1451–1464. <https://doi.org/10.1093/plcell/koad329>
- Sun, W., Shahrajabian, M. H., Kuang, Y., & Wang, N. (2024). Amino acids biostimulants and protein hydrolysates in agricultural sciences. **Plants**, 13(2), 210. <https://doi.org/10.3390/plants13020210>
- Thepbandit, W., Srisuwan, A., & Athinuwat, D. (2024). Priming of exogenous salicylic acid under field conditions enhances crop yield through resistance to *Magnaporthe oryzae* by modulating phytohormones and antioxidant enzymes. **Antioxidants**, 13(9), 1055. <https://doi.org/10.3390/antiox13091055>
- van Butselaar, T., & Van den Ackerveken, G. (2020). Salicylic acid steers the growth–immunity tradeoff. **Trends in Plant Science**, 25(6), 566–576. <https://doi.org/10.1016/j.tplants.2020.02.002>