

Exploring the Potential of Bio-Stimulant from Organic Waste: Improving Plant Growth and Disease Resistance

Lakshmiprabha Selvaraj¹, Kannikaparameswari Nachimuthu², Ashanath Fowmitha Nazeer³, Sumathi Ramasamy⁴, Natchiappan Senthikumar^d

¹⁻²Dr. N.G.P College of arts and Science, Coimbatore, Tamil Nadu, India

³⁻⁵ICFRE- Institute of Forest Genetics and Tree Breeding, Coimbatore, Tamil Nadu, India

Abstract:

This study aimed to develop a natural biostimulant (F2) from fruit peels and floral wastes and evaluate its effect on plant growth. Methanolic extracts of pineapple, pomegranate peels, and red rose, paneer rose, and chrysanthemum petals were used to formulate F2. Phytochemical screening confirmed the presence of bioactive compounds such as alkaloids, flavonoids, tannins, saponins, terpenoids, phenols, glycosides, and quinones. HPLC analysis identified quercetin and related flavonoids, particularly in F2 and floral waste extracts. Application of the F2 biostimulant on *Solanum melongena* under greenhouse conditions significantly improved plant height, leaf number, and leaf coloration compared to untreated controls. Treated plants also showed reduced disease severity. These growth enhancements are attributed to the antioxidant, antimicrobial, and metabolic-stimulating properties of the phytoconstituents. Overall, the results demonstrate that the F2 biostimulant offers a sustainable and eco-friendly approach to improving crop productivity and plant health using agro-waste resources.

Keywords — Bio-stimulant, Organic waste, phytochemical analysis, HPLC identification.

I. INTRODUCTION

Agriculture depends on synthetic pesticides and fertilizers for maximum yields, but excessive use of pesticides leads to soil pollution, water contamination and destruction of useful microbes [1]. The search for the ecofriendly alternatives has been driven due to the high costs and health hazards associated with pesticide use [2]. Organic waste derived bio-stimulant, made from fruit peel and petal residues, offer a sustainable solution by enhancing plant growth, Nutrient absorption, and stress resistance without harming the environment. These natural compounds promote agricultural productivity while supporting a more sustainable Farming system [3].

This study examined organic waste-based biostimulants for enhancing plant growth, pathogen resistance, and reduce synthetic inputs. Utilizing natural bioactive compounds from organic waste, these bio- stimulant stimulated the root development,

improved nutrient absorption, and increased stress tolerance. *Solanum melongena* (eggplant) was the model crop to assess growth, leaf and shoot development, and disease resistance under the greenhouse conditions [4].

Preliminary findings showed that plants treated with organic waste, Bio-stimulant exhibited enhanced growth resilience. Key phytochemical like alkaloids, flavonoids and terpenoids promoted plant health and defence against pathogens. HPLC analysis identified quercetin as crucial for growth and disease resistance. These results highlight bio-stimulant potential to reduce reliance on synthetic inputs while supporting soil health and sustainability.

II. Materials and Methods

A. Preparation of F2 sample

The Ingredients of F2 samples used in this study, are Fruit peels of *Ananas comosus* (pineapple) and

Punica granatum (pomegranate), along with floral wastes including *Rosa indica* (Red Rose), *Rosa damascena* (Paner Rose), and *Chrysanthemum indicum* (Chrysanthemum) were collected from local markets in Coimbatore, Tamil Nadu. The collected samples were thoroughly washed with distilled water to remove dirt and surface impurities, then air-dried at room temperature to eliminate residual moisture. The dried fruit/Flower wastes were ground into a fine powder using an electric grinder [5]

B. Extraction of samples

The powdered samples were mixed with methanol in a 1:10 (w/v) ratio and boiled for 3 hours to release the bioactive compounds. The resulting mixture was cooled to room temperature and filtered through a muslin cloth to remove solid residues. The filtrate was then evaporated to dryness using rotary evaporator to obtain the crude extract, and stored at 4°C for further use.

C. Phytochemical Screening

The individual Fruit/Flower waste and the mixed sample (F2) were subjected to phytochemical screening to detect the presence of bioactive phytochemicals. Based on the colour intensity, the results were tabulated.

D. Metabolimic study of Bio-stimulant using HPLC Analysis:

Bioactive compounds present in the individual sample and mixed sample (F2) were identified using HPLC Analysis. A HITACHI HPLC instrument was employed to carry out the analyses. The instrument was interfaced with an Agilent 7000 Ultra High Definition (UHD) Accurate Mass. A reversed-phase C18 analytical column (Agilent Zorbax Eclipse Plus, 1.8 μm , 4.6 \times 150 mm) with a guard cartridge of the same packing was utilized for separation. The flow rate was 0.5 mL/min, and the mobile phases were composed of 100% acetonitrile as solvents A and 1% phosphoric acid, 10% acetic acid, 5% acetonitrile in water as solvent B, respectively. The injection volume was 20 μL at 260 nm [6].

E. Experimental design

The experiment was carried out according to randomized complete block design with four

replicates. It was comprised of four treatments, viz., 1) The control 2) *Balanites aegyptiaca* seed oil 3) Tree PAL^H 4) kraft solution 5) F2 sample (Table 1). *Solanum melongena* (eggplant) was chosen for this experiment because of its economic value and susceptibility to frequent pathogens. *Solanum melongena* plants were purchased from the SIV nursery, Kuppepalayam, Coimbatore and utilized for the experimental setup in the greenhouse. The plants were cultivated in a soil blend made up of 60% loamy soil, 30% organic compost, and 10% sand to ensure proper aeration and nutrition [7].

Table 1. Field Test Using *Solanum melongena* under field condition

S. No	Plants	No. of plants	Dosage
1	Control	4	Water
2	<i>Balanites aegyptiaca</i> oil Treatment (Organic Biostimulant)	4	0.5 ml in 1 liter of water
3	Tree PAL ^H Treatment (IFGTB Product)	4	0.5 ml in 1 liter of water
4	Kraft Perfekt Treatment (Marketable Product)	4	1 ml in 1 liter of water.
5	F2 Organic Biostimulant treatment	4	1 ml in 1 liter of water.

F. Measurement of Plant height, No.of leave, No.of leaves fallen and its colour change

Four week after foliar spray and soil drench at regular time intervals application of control, BA oil (*Balanited aegyptiaca* seed oil), Tree PAL^H, Kraft solution and F2 samples, Plant height, number of leaves, number of leaves fallen and its colour change were evaluated. Also disease severity was assessed by visual rating and detection of the pathogen [8].

G. Statistical analysis

The results were expressed as Mean \pm SD. The data were statistically analyzed using SPSS version 20.0 by means of one-way ANOVA followed by Duncan's test. Mean values were considered statistically significant when $p < 0.05$.

III. Results and Discussion

A. Phytochemical screening of F2 sample and Individual waste matters.

The phytochemical analysis of the F2 sample revealed a diverse array of bioactive compounds, including alkaloids, flavonoids, tannins, saponins,

glycosides, terpenoids, phenols, and quinones, indicating a rich profile of secondary metabolites with potential biostimulant activity (Table 2)[9]. These phytoconstituents are known to enhance plant growth and development by stimulating physiological and metabolic processes. Alkaloids and flavonoids, for instance, contribute to enhanced antioxidant defence and enzyme regulation in plants, improving stress tolerance and vitality. Tannins and saponins can play roles in strengthening plant immunity [10], while glycosides and terpenoids are associated with promoting nutrient uptake and hormonal balance [11]. The presence of phenols and quinones suggests a role in modulating oxidative stress and supporting overall plant vigor [12]. Collectively, the abundance of these bioactive compounds supports the efficacy of the F2 sample as a natural biostimulant, capable of improving plant growth, health, and resilience in a sustainable manner.

indicating the presence of quercetin or structurally similar flavonoids. The F2 sample, a formulated organic biostimulant extract, exhibited multiple peaks with high intensity, suggesting a synergistic enrichment of phenolic and flavonoid compounds. This diverse phytochemical profile implies enhanced antioxidant potential and greater biostimulant efficacy. The elevated peak intensities, particularly in the floral extracts and F2 formulation, support their potential application in promoting plant growth, stress tolerance, and pathogen resistance. These results are consistent with preliminary phytochemical screening, further reinforcing the role of such natural formulations as eco-friendly biostimulants in sustainable agriculture.

Table 2. The phytochemical screening of F2 Sample

S. No	Name of the test	Presence/Absence in methanol
1	Alkaloids	Present
2	Flavonoids	Present
3	Tannins	Present
4	Saponins	Present
5	Quinone	Present
6	Sterol	Absent
7	Phenol	Present
8	Anthocyanin	Absent
9	Protein	Absent
10	Carbohydrate	Absent
11	Steroids	Absent
12	Terpenoids	Present
13	Glycosides	Absent

B. HPLC Analysis and Discussion

The HPLC analysis of methanolic extracts derived from fruit peel, floral wastes and F2 demonstrated the presence of key bioactive flavonoids, with special emphasis on quercetin identification (Figure 1). The quercetin standard showed a distinct peak at approximately 5.9 minutes, which was used as a reference marker. Among the tested samples, significant peaks aligning with the quercetin retention time were observed in red rose petals, paneer rose petals, and notably the F2 sample,

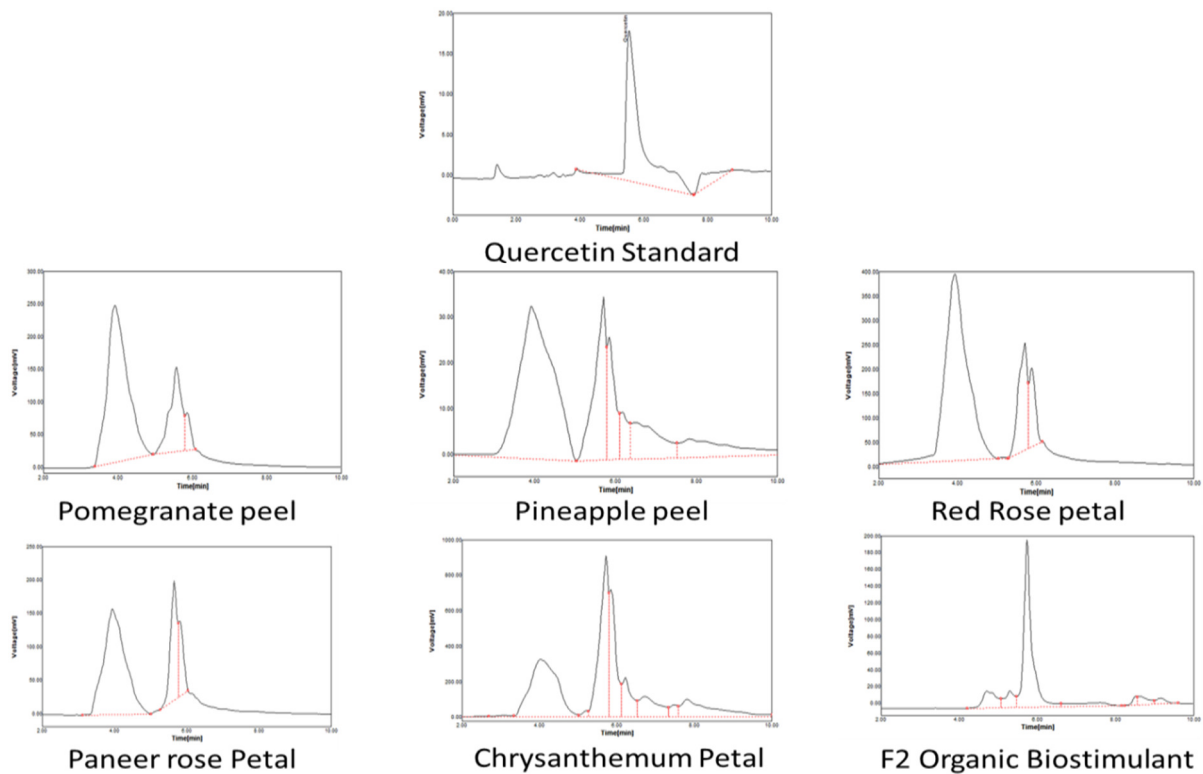


Figure 1. HPLC Chromatogram

C. Measurement of Plant height, No.of leave, No.of leaves fallen and its colour change

The application of the formulated F2 biostimulant on *Solanum melongena* under controlled greenhouse conditions significantly enhanced plant growth and physiological parameters compared to untreated controls (Figure 2). Over a four-week period, biostimulant-treated plants exhibited notable improvements in plant height, number of leaves, and leaf coloration. The average plant height of treated plants increased consistently across the weeks, reaching a maximum mean of 37.33 cm by the fourth week compared to 26.67 cm in control plants. Similarly, the number of leaves increased from 8.00

to 16.67 in treated plants, indicating a higher rate of vegetative growth. Enhanced leaf greenness and pigmentation were also observed, suggesting improved chlorophyll content and photosynthetic efficiency. Disease severity was considerably lower in treated plants, correlating with the presence of antimicrobial phytochemicals identified in the biostimulant [13]. These effects can be attributed to the bioactive constituents such as flavonoids, phenols, and saponins present in the extract, which are known to promote plant vigor, boost immune responses, and stimulate nutrient uptake [14]. Overall, the results underscore the efficacy of the biostimulant in promoting healthy growth and resilience in brinjal plants, thereby supporting its application in sustainable agriculture (Figure 3).

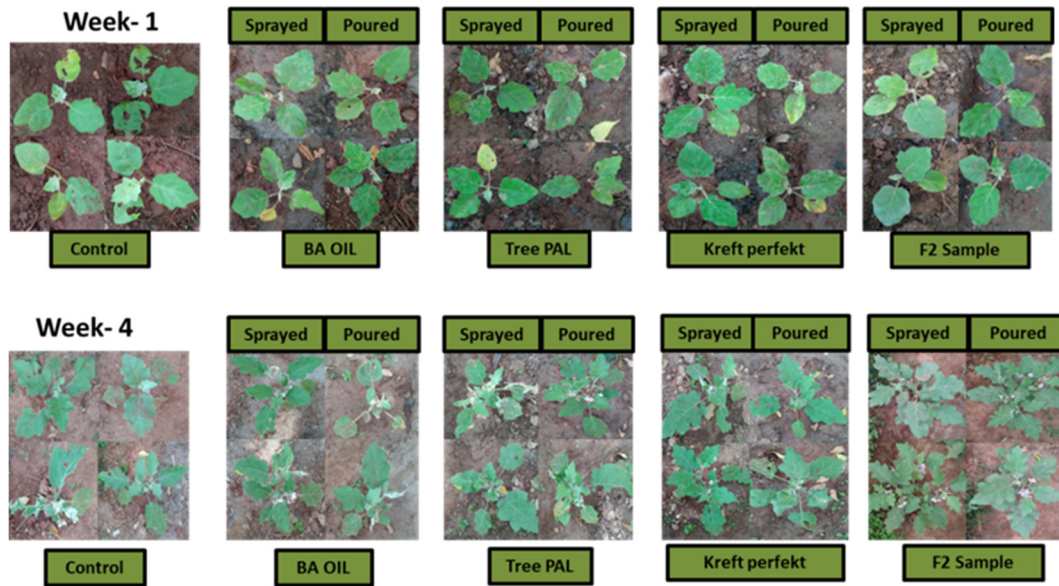


Figure 2: Analysis of plant growth condition

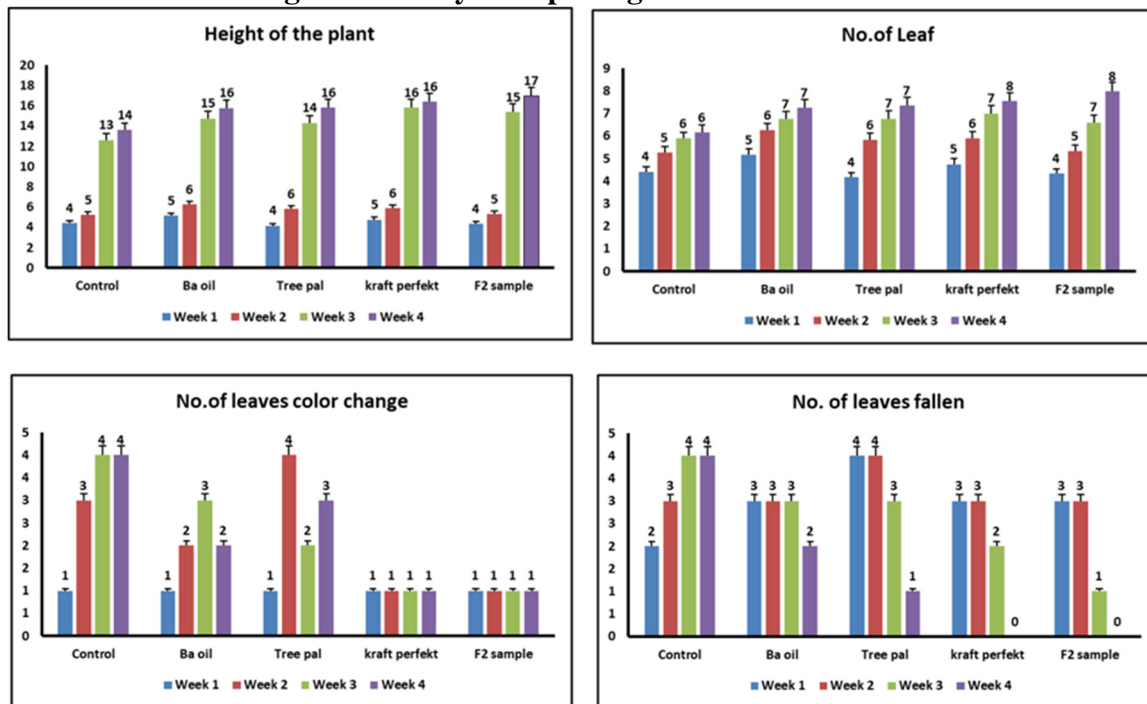


Figure 3: Analysis of plant growth condition

IV. Conclusion

The comprehensive analysis of the F2 biostimulant formulation, derived from individual fruit and floral waste extracts, confirms its potent bioactive profile and promising role in sustainable agriculture. Phytochemical screening revealed the presence of key secondary metabolites such as alkaloids, flavonoids, tannins, saponins, terpenoids, phenols, glycosides, and quinones, all known for their roles in

enhancing plant physiological functions, stress resistance, and overall vigor. HPLC profiling further substantiated the presence of quercetin-like flavonoids in F2 and its individual components, particularly floral wastes, highlighting the synergistic antioxidant and growth-promoting potential of the formulation. The greenhouse trial on *Solanum melongena* demonstrated significantly improved plant height, leaf number, and coloration

in F2-treated plants compared to controls, with reduced disease severity, validating the formulation's bioefficacy. Together, these findings suggest that the F2 biostimulant harnesses the power of natural phytoconstituents to enhance crop performance, offering an eco-friendly, value-added solution for integrated plant health management.

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REFERENCES

1. Tudi, M., Ruan, H. D., Wang, L., Lyu, J., Sadler, R., Connell, D., Chu, C., & Phung, D. T. (2021). *Agriculture development, pesticide application and its impact on the environment*.
2. Bourguet, D., & Guillemaud, T. (2016). *The hidden and external costs of pesticide use*.
3. Du Jardin, P. (2015). *Predicting bankruptcy using neural networks and other classification methods: The influence of variable selection techniques on model accuracy*. *Neurocomputing*, **168**, 102–112.
4. Sudheesh, S., Sandhya, C., Koshy, A. S., & Vijayalakshmi, N. R. (1999). *Antioxidant activity of flavonoids from Garcinia cambogia and Garcinia indica*. *Phytotherapy Research*, **13**(5), 364–366.
5. Azizi, M., & Alavi, M. (2019). Utilization of fruit and vegetable wastes as biostimulants: A sustainable approach. *Waste and Biomass Valorization*, **10**(10), 2979–2992.
6. Hamade, K., Fliniaux, O., Fontaine, J. X., Molinié, R., Petit, L., Mathiron, D., ... & Mesnard, F. (2024). NMR and LC-MS-based metabolomics to investigate the efficacy of a commercial bio stimulant for the treatment of wheat (*Triticum aestivum*). *Metabolomics*, **20**(3), 58.
7. Yuniati, N., Kusumiyati, K., Mubarok, S., & Nurhadi, B. (2023). Assessment of biostimulant derived from moringa leaf extract on growth, physiology, yield, and quality of green chili pepper. *Sustainability*, **15**(9), 7113.
8. Saporta, R., Bou, C., Frías, V., & Mulet, J. M. (2019). A method for a fast evaluation of the biostimulant potential of different natural extracts for promoting growth or tolerance against abiotic stress. *Agronomy*, **9**(3), 143.
9. Mrid, R. B., Benmrid, B., Hafsa, J., Boukcim, H., Sobeh, M., & Yasri, A. (2021). Secondary metabolites as biostimulant and bioprotectant agents: A review. *Science of the Total Environment*, **777**, 146204.
10. Iqbal, N., & Poór, P. (2024). Plant protection by tannins depends on defence-related phytohormones. *Journal of Plant Growth Regulation*, 1-18.
11. Kapoor, R., Anand, G., Gupta, P., & Mandal, S. (2017). Insight into the mechanisms of enhanced production of valuable terpenoids by arbuscular mycorrhiza. *Phytochemistry Reviews*, **16**, 677-692.
12. Hajam, Y. A., Lone, R., & Kumar, R. (2023). Role of plant phenolics against reactive oxygen species (ROS) induced oxidative stress and biochemical alterations. In *Plant phenolics in abiotic stress management* (pp. 125-147). Singapore: Springer Nature Singapore.
13. Barbieri, R., Coppo, E., Marchese, A., Daglia, M., Sobarzo-Sánchez, E., Nabavi, S. F., & Nabavi, S. M. (2017). Phytochemicals for human disease: An update on plant-derived compounds antibacterial activity. *Microbiological research*, **196**, 44-68.
14. El-Saadony, M. T., Zaber mawi, N. M., Zaber mawi, N. M., Burollus, M. A., Shafi, M. E., Alagawany, M., ... & Abd El-Hack, M. E. (2023). Nutritional aspects and health benefits of bioactive plant compounds against infectious diseases: a review. *Food Reviews International*, **39**(4), 2138-2160.