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RESEARCH ARTICLE

Pseudomonas sp. and rooting agents: A sustainable alternative for fig (*Ficus carica* L.) production

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ABSTRACT

Plant growth-promoting rhizobacteria have proven to be a sustainable alternative for improving agricultural establishment and production. This is particularly important for the propagation and production of fig trees (*Ficus carica* L.), a crop that stands out for the high nutritional value, fiber content, and bioactive compounds of its fruit. This study evaluated the effect of *Pseudomonas* sp. and a commercial rooting agent on the productive and biochemical variables of 'Black Mission' fig trees in a greenhouse. Before transplanting the fig cuttings, 1×10^8 colony-forming units mL^{-1} *Pseudomonas* sp., commercial rooting agent (indole-3-butyric acid [IBA]), a combination of both, and a control were applied; the crop was managed in polyethylene bags, increasing their volume from 3 to 30 L. Subsequently, *Pseudomonas* sp. was applied at the same dose each month to plants previously treated with this rhizobacteria. With the combination of *Pseudomonas* sp. and IBA, there was a higher bacterial concentration in the rhizosphere (61.4%), an increase in the number of fruits (5.2%), yield (67.8%), total soluble solids (6.6%), and P concentration in leaves (14.6%) compared to the control. The individual application of *Pseudomonas* sp. increased fruit weight (13.4%), as well as polar (7.9%) and equatorial (6.4%) diameters compared to the control. However, the control had the highest anthocyanin levels compared to the other treatments (29.3%). *Pseudomonas* sp., alone or in combination with the rooting agent, is a sustainable option for improving fig production and quality.

Key words: Biostimulants, fig tree, indole-3-butyric acid, plant growth-promoting rhizobacteria, roots.

INTRODUCTION

In recent decades, the development of agricultural biosafety products has evolved from the use of agrochemicals to more sustainable systems designed to avoid negative impacts on soil microflora and fauna (Igiehon et al., 2024). This change has become increasingly relevant due to the worsening of climate change, which has intensified the effects of abiotic stress, negatively affecting plant growth and productivity (Chieb and Gachomo, 2023). As a result, plant growth-promoting rhizobacteria (PGPR) have emerged as a key tool for mitigating these challenges. The PGPR have proven their efficiency in improving plant establishment and production through various mechanisms, such as the production of growth hormones and the solubilization of minerals, which facilitate their absorption by plants (Vejan et al., 2016). Among these mechanisms, the ability to produce low molecular weight organic acids stands out, which are fundamental in the solubilization of soil phosphate, increasing its availability to plants (Chen et al., 2021).

Microorganisms in the rhizosphere play a biostimulant role in plant development and adaptation, facilitating processes such as biological N fixation, nutrient solubilization, and resistance to stress conditions, reinforcing their importance as key allies in sustainable agriculture (Bhattacharyya and Jha, 2012).

The production of indole-3-acetic acid (IAA) by rhizobacteria has been linked to the promotion of plant growth, particularly in processes such as root initiation and elongation (Nosheen et al., 2018). The IAA and indole-3-butyric acid (IBA) are among the most important plant hormones in the auxin family, which regulates various aspects of plant development, including growth, differentiation, and response to environmental stimuli (Bunsangiam et al., 2021). Furthermore, its function is not limited to plants, as it also plays a crucial role in the development of microorganisms and in plant-microorganism interactions. In this way, PGPR play an essential role in the interaction between plants and microorganisms, promoting an increase in fruit yield and quality. In particular, the genus *Pseudomonas* regulates plant growth, with positive effects on key agronomic variables in this crop (Gándara-Ledezma and Gutiérrez-Coronado, 2023), and contributes to the control of phytopathogens through the secretion of phytohormones and other secondary metabolites, such as flavonoids and specific enzymes, for example, phenylalanine ammonia lyase (Sah et al., 2021).

Radix 10 000 is a plant growth regulator, formulated as an impregnable powder, containing 1.0% IBA as the active ingredient, one of the most effective auxins in promoting adventitious or lateral root formation. The IBA can be absorbed by any part of the plant. It acts on its own and through its transformation into IAA, which also regulates root growth (Interie, 2014).

This is particularly important for the propagation and rooting of fig tree (*Ficus carica* L.) cuttings and their production, due to the consumption of agrochemicals that serve the same purpose. Fig cultivation stands out due to growing demand for its fruit in national and international markets, with Mexico having the potential to position itself as a leading global producer (Soberanes-Pérez et al., 2020). This species contributes directly to food security and plays an essential role in human nutrition, hence its strategic importance in current agri-food systems (FAO, 2023). Figs, both fresh and dried, are notable for their nutritional value and functional potential. Various studies have shown that these fruits contain significant amounts of fiber, essential minerals, and bioactive compounds such as flavonoids and phenolic acids, which are antioxidants. It has been observed that dark, ripe figs have higher concentrations of these compounds, especially in the skin of the fruit, suggesting their relevance in preventing oxidative damage and promoting intestinal health (Arvaniti et al., 2019). Therefore, the objective was set to evaluate the effect of *Pseudomonas* sp. and a commercial rooting agent on the productive and biochemical variables of 'Black Mission' fig trees, thereby contributing to the reduction in the use of agrochemicals as a sustainable alternative.

MATERIALS AND METHODS

Study area

The experiment was conducted in a zenithal greenhouse located in the Department of Horticulture at the Universidad Autónoma Agraria Antonio Narro (UAAAN) in Saltillo (25°23'36" N, 101°00'02" W; 1785 m a.s.l.), Coahuila, Mexico. During the period between March 2023 and December 2024, temperatures ranged from 18 to 40 °C throughout the growing cycle, with an average photosynthetically active radiation (PAR) of 594 µmol m^{-2} s^{-1}.

Plant material

Fig tree (*Ficus carica* L.) 'Black Mission' was used, which is highly nutritious due to its rich content of vitamins, minerals, fiber, and antioxidants. The cuttings were obtained from plants in production at the UAAAN Department of Horticulture. The treatments were applied to the cuttings, using five replicates per treatment with eight plants each.

Management and treatments

Fig tree cuttings were obtained from pruning material from healthy and productive mother plants. From this material, cuttings were selected based on uniformity of diameter, length, and number of buds, ensuring visual and morphological homogeneity among the propagules. Initially, the cuttings were stored in refrigeration at 10 °C for 1 wk, a cold stratification technique that helps induce rooting and sprouting of new plants.

Four treatments were established under a randomized complete block design: 1) Control, 2) commercial rooting agent (Radix 10 000, Intercontinental Import Export, Salamanca, Guanajuato, Mexico), 3) *Pseudomonas* sp. at a concentration of 1×10^8 colony forming units (CFU) mL^{-1}, and 4) *Pseudomonas* sp. at the same concentration combined with the commercial rooting agent indole-3-butyric acid (IBA).

Before transplanting, the cuttings that received the *Pseudomonas* sp. treatments were immersed in a solution containing this rhizobacterium at the aforementioned concentration for 30 min. For the rooting agent treatments, the product was applied by immersing the base of the cutting in the IBA rooting agent, and for the control, no prior treatment was applied. The cuttings were planted in 3 L polyethylene bags, using a substrate composed of peat moss, loamy soil, and perlite, in a ratio of 40:30:30. Soil was used to promote better adaptation of *Pseudomonas* sp., as it is a rhizobacterium native to this type of soil.

In July 2023, the plants were transplanted into 15 L pots and pruned. Subsequently, the plants were transplanted into 30 L containers in July 2024, and throughout the entire cultivation period, from planting to fruiting, they were fertilized with a 60% Steiner solution (60% milliequivalent content: 7.2 NO_3, 0.6 H_3PO_4, 4.2 K, 5.4 Ca, 2.4 Mg, 4.2 S, electrical conductivity (EC) 1.2 dS m^{-1}), prepared with the following fertilizers: Potassium nitrate (KNO_3), calcium nitrate ($Ca(NO_3)_2 + 4H_2O$), magnesium nitrate ($Mg(NO_3)_2 + 6H_2O$), monopotassium phosphate (KH_2PO_4), a mixture of micronutrients (Ultrasol micro mix, Sociedad Química y Minera de Chile-SQM Comercial de Mexico S.A. de C.V., Zapopan, Jalisco, Mexico), 85% nitric acid (HNO_3) and 90% sulfuric acid (H_2SO_4). In addition, *Pseudomonas* sp. was applied at same concentration to the base of the stem every month for 18 mo to the treatments previously treated with the same rhizobacteria.

Evaluated variables

The variables were measured during the reproductive stage of each harvest. The number of fruits per plant (FN) and average fruit weight (FW) were evaluated using a Steren scale; polar diameter (PD) and equatorial diameter of the fruit (ED) were evaluated using a Steren vernier caliper; total soluble solids (TSS) were measured using an ATC refractometer. Yield was calculated per plant (FY) in grams, according to the following formula:

$$FY = FN \times FW$$

To determine total anthocyanins (ANT), an extractive solution was prepared consisting of methanol with 0.1% hydrochloric acid (HCl) (v/v), which was left to stand for 24 h to ensure its chemical stability. The following day, fresh figs were harvested, ensuring uniformity in size and degree of ripeness for each treatment. The epicarp (skin) of each fruit was manually removed, as it contains the highest concentration of anthocyanins. The epicarp samples were immersed in the prepared extracting solution. The tissue was then macerated; the resulting mixture was filtered and measured in a volumetric flask. The extract was stored refrigerated at 4 °C for 24 h to minimize oxidative processes. The absorbance of the extract was then measured using spectrophotometry (Jenway 6320D, Cole-Parmer Instrument Company, Vernon Hills, Illinois, USA) at a wavelength of 532 nm, corresponding to maximum absorption for anthocyanins. The ANTs were determined using a standard curve constructed with cyanidin-3-glucoside, expressing the results in milligrams per 100 grams of fresh sample (mg 100 g^{-1}). The formula used for the calculation was as follows:

$$\text{ANT (mg 100 g}^{-1}) = \frac{\text{Abs} \times V_{ext} \times 1000}{\varepsilon \times L \times m}$$

where Abs is absorbance measured at 532 nm, V_{ext} is extract volume (mL), ε is molar extinction coefficient of cyanidin-3-glucoside (26 900 L mol^{-1} cm^{-1}), L is spectrophotometer cell length (1 cm), m is fresh sample mass (kg).

To determine P content in leaves, the colorimetric method using spectrophotometry was used. One gram of dry sample was incinerated in a muffle furnace (Thermo Fisher Scientific, Asheville, North Carolina, USA) at 500-600 °C until clear ashes were obtained, which were dissolved in 50 mL 50% HCl. From this solution, 1 mL aliquot was taken and transferred to a test tube washed with P-free detergent and rinsed with distilled water. Subsequently, 5 mL 5% ammonium molybdate solution and 2 mL 2-hydroxy-1-aminonaphthalene-4-sulfonic acid (ANSA) reagent were added, stirring to homogenize the mixture. The reaction was allowed to stand for 20 min at room temperature, allowing the blue molybdenum complex to develop. The solution was transferred to a clean cell and the absorbance was measured at 650 nm using a UV-Vis spectrophotometer model UV-1100 (DLAB Scientific, Shunyi District, Beijing, China), using distilled water as a blank. The P concentration was obtained by interpolation on a calibration curve constructed from standard phosphate solutions, and the results were adjusted according to the dilution and dry weight of the analyzed sample.

The Kjeldahl method was used to determine N in fruits. This method is based on the conversion of organic N into ammonium ions (NH_4^+) through acid digestion, followed by its transformation into ammonia (NH_3) through alkalization, and subsequent distillation and titration. Dry ground samples (0.5 g) were weighed and placed in heat-resistant digestion tubes. Each sample was added 4 mL digesting mixture composed of

concentrated sulfuric acid (H_2SO_4), potassium sulfate (K_2SO_4), mercury oxide (HgO), and a Cu and Se catalyst. Digestion was carried out under controlled heating until a lime green color was achieved, indicating the complete conversion of organic N to NH_4^+ ion. After cooling, the digested solution was distilled in the presence of 50% sodium hydroxide (NaOH), releasing NH_3, which was captured in a flask containing 2% boric acid and a few drops of mixed indicator. Finally, the captured NH_3 was quantified by direct titration with 0.025 N H_2SO_4, recording the volume consumed until a color change was observed. The results were calculated using the following formula, expressed as a percentage of total N based on dry weight.

$$\text{N}\ (\%) = \frac{(\text{Vm} - \text{Vb}) \times \text{N} \times 14.007 \times 100}{\text{P}}$$

where Vm is volume of acid consumed in the titration of the sample (mL), Vb is volume of acid consumed in the blank (mL), N is normality of H_2SO_4 used in titration (eq L^{-1}), 14.007 is atomic weight of N (g mol^{-1}), P is weight of the sample analyzed (mg).

Bacterial concentration was estimated by measuring turbidity in a UV-Vis spectrophotometer model UV-1100 (DLAB Scientific) at 600 nm. To do this, different serial dilutions were adjusted in sterile saline solution, recording the optical density (OD) of each dilution. At the same time, the viable OD count was performed using colony-forming units (CFU mL^{-1}). Once the curve was generated, bacterial concentrations were determined indirectly using spectrophotometric readings.

Statistical analysis

A randomized block design was used to control the variability of relative humidity within the greenhouse. These gradients were associated with a wet wall located on the north side and an extractor fan on the south side. The data were analyzed for each variable. First, it was determined that the data were normal according to the Shapiro-Wilks test, and then an ANOVA and Duncan's mean comparison test ($p \leq 0.05$) were performed using the statistical software InfoStat 2020 (Grupo InfoStat, Universidad Nacional de Córdoba, Córdoba, Argentina).

RESULTS AND DISCUSSION

Number and fruit weight

Treatment with *Pseudomonas* sp. + indole-3-butyric acid (IBA) showed a significant increase of 5.2% in number of fruit (FN) compared to the control, while the control, IBA, and *Pseudomonas* sp. treatments showed the lowest values and were statistically equal to each other (Table 1). This suggests an improvement attributed to the combined effect of rhizobacteria and commercial rooting agents, favoring the absorption of essential nutrients and the production of phytohormones that stimulate reproductive development, as it solubilizes P through organic acids and phosphatase enzymes, and synthesizes phytohormones such as auxins, gibberellins, and cytokinins, promoting root production and plant growth (Beltrán-Pineda and Bernal-Figueroa, 2022). These results are consistent with the effect obtained by Villaseñor-Tulais et al. (2023), who reported a 65.5% increase in tomato fruit yield with *P. fluorescens*, while Camacho-Rodríguez et al. (2022) observed an increase of up to 96% in jalapeño peppers with *Serratia liquefaciens*.

The application of *Pseudomonas* sp. alone promoted a significant increase in fruit weight (FW), registering a 13.4% increase compared to control. Although the *Pseudomonas* sp. + IBA treatment did not show significant differences compared to other treatments (Table 1). These results suggest that inoculation with rhizobacteria, either individually or in combination with the IBA rooting agent, has a positive effect on fruit weight. This is attributed to improved nutrient availability and physiological stimulation of fruit filling, as in the work of Gándara-Ledezma and Gutiérrez-Coronado (2023), who reported a 30% increase in fruit weight when applying bacterial consortia. Similarly, in the work of Soberanes-Pérez et al. (2020), increases of 5-20 g were observed in figs under the application of bioregulators that can be produced by plant growth-promoting rhizobacteria (PGPR), and Dong et al. (2023) reported increases of more than 42% in tomatoes with the application of *Bacillus*.

Table 1. Effect of *Pseudomonas* sp. and commercial rooting agent (indole-3-butyric acid, IBA) on the production and quality of fig crop. FN: Number of fruits; FW: fruit weight; PD: polar diameter; ED: Equatorial diameter; FY: fruit yield per plant; TSS: total soluble solids; ANT: anthocyanins; BC: Bacterial concentration; CFU: colony forming units. Values on the right correspond to the standard error. Means with different letters within the same column indicate a significant difference (Duncan, $p \leq 0.05$).

	Variable				
Treatment	FN	FW	PD	ED	FY
		g	mm	mm	g plant^{-1}
Control	18.83 ± 1.48^{b}	26.25 ± 0.48^{b}	41.03 ± 0.27^{b}	35.47 ± 0.37^{b}	492.28 ± 28.80^{c}
IBA	18.00 ± 2.16^{b}	26.99 ± 1.15^{b}	41.00 ± 0.66^{b}	35.72 ± 0.48^{b}	480.60 ± 43.40^{c}
Pseudomonas sp.	22.00 ± 1.95^{b}	29.77 ± 0.08^{a}	44.27 ± 0.25^{a}	37.74 ± 0.23^{a}	654.63 ± 57.08^{b}
Pseudomonas sp. + IBA	28.67 ± 1.25^{a}	28.83 ± 0.83^{b}	43.19 ± 0.81^{a}	37.51 ± 0.12^{a}	826.34 ± 40.65^{a}
Treatment	TSS	ANT	N	P	BC
	°Brix	mg 100 g^{-1}	%	mg 100 g^{-1}	10 ^{8} CFU mL^{-1}
Control	17.75 ± 0.28^{b}	87.60 ± 1.84^{a}	0.07 ± 0.01^{a}	913.04 ± 13.77^{b}	18.71 ± 0.67^{c}
IBA	18.52 ± 0.20^{a}	66.34 ± 5.19^{b}	0.07 ± 0.01^{a}	993.02 ± 29.18^{ab}	20.43 ± 0.82^{bc}
Pseudomonas sp.	18.45 ± 0.14^{a}	69.56 ± 5.12^{b}	0.09 ± 0.01^{a}	1003.00 ± 30.95^{ab}	21.61 ± 0.37^{b}
Pseudomonas sp. + IBA	18.92 ± 0.35^{a}	54.35 ± 6.83^{b}	0.09 ± 0.01^{a}	1046.18 ± 17.19^{a}	28.59 ± 0.53^{a}

Polar and equatorial diameter

Inoculation with *Pseudomonas* sp. in combination with IBA significantly increased polar diameter (PD) by 7.89% and 5.26% compared to the control and the IBA-only treatment (Table 1). This reflects the influence of rhizobacteria, which can promote fruit morphological development through cell division and elongation. The results are consistent with Martínez et al. (2019), who observed increases in melon fruit size with *P. fluorescens* attributed to better absorption of N, P, and Zn. Bona et al. (2015) also reported higher production and size in strawberry fruits with *Pseudomonas* spp. and mycorrhizae compared to non-inoculated plants.

Treatments with *Pseudomonas* sp. alone and in combination with IBA performed similarly, but showed a significantly higher equatorial diameter (ED) than the control and the treatment with rooting agent IBA by 6.39% and 5.75% (Table 1), respectively, suggesting that rhizobacteria improve reproductive structures by promoting nutrient absorption and the production of phytohormones, such as auxins and cytokinins, which directly influence cell division and expansion (Calvo et al., 2014; Egamberdieva et al., 2017). The results are consistent with Gándara-Ledezma and Gutiérrez-Coronado (2023), who also observed improvements in the size and weight of fig fruits with microbial consortia composed of *P. fluorescens*, *Bacillus subtilis*, *Bacillus cereus*, and *Trichoderma harzianum*.

Yield per plant

The fruit yield per plant (FY) was 67.8% higher when using the *Pseudomonas* sp. + IBA mixture compared to the control, followed by *Pseudomonas* sp. alone, which exceeded the control by 33%. There was nonsignificant difference between the IBA treatment and the control (Table 1). Garcia-Seco et al. (2015) reported a significant increase in fruit yield in *Rubus* sp. after inoculation with *P. fluorescens*, which they attribute to improvements in vegetative development, flowering, and photosynthetic efficiency of the plant. Similarly, Condori et al. (2024) and Altunlu et al. (2024) reported improvements of 8%-22% with PGPR in corn and tomatoes under stress. Chiquini-Medina et al. (2021) reported a 32% increase in habanero chili peppers with *Pseudomonas* sp., which coincides with the results of the present study.

Total soluble solids

The treatments with *Pseudomonas* sp. and IBA did not show significant differences between each other for the total soluble solids (TSS) variable; however, all significantly outperformed the control, showing an increase of 4.33%, 3.94%, and 6.59%, respectively, which indicates that these rhizobacteria and the rooting agent can increase their soluble sugar content, which is related to better nutrition, greater photosynthetic activity, and C metabolism (Calvo et al., 2014; Egamberdieva et al., 2017). Pii et al. (2018) observed greater sweetness in strawberries with *Azospirillum brasilense*, probably due to a reduction in the total concentration of citrate,

which increases the perception of sugars, while Gashash et al. (2022) reported increases of up to 26% in the TSS content in tomatoes, improving the organoleptic quality of the fruit.

Anthocyanins

The control had 20.6% to 38% higher anthocyanins (ANT) content than the other treatments. The reduction in the latter could be associated with lower exposure to environmental or physiological stress, which in turn reflects a better metabolic condition of the plants treated with *Pseudomonas* sp. or IBA. This was because PGPR, such as *Pseudomonas* sp., can reduce stress in plants by improving nutrient absorption, inducing phytoalexin production, and increasing antioxidant activity (Egamberdieva et al., 2017). The results are consistent with Zulueta-Rodriguez et al. (2014), who reported a reduction of up to 25% in anthocyanin content compared to the control in poinsettia (*Euphorbia pulcherrima*) plants treated with *P. putida* strains. Anthocyanins, belonging to the flavonoid group, are pigments that give plants different colors and contribute to their defense against various types of environmental stress, helping to neutralize reactive oxygen species (Li and Ahammed, 2023). These actions can mitigate adverse conditions, which would explain the lower concentration of anthocyanins in the PGPR and IBA treatments.

Nitrogen and phosphorus content

Although nonsignificant differences were observed between treatments in N content. This result indicates a possible positive effect of *Pseudomonas* sp. on N nutrition, even if it did not reach significance. Studies such as that by Chen et al. (2021) showed that inoculating PGPR increased corn production by 8%-20% under low N and P conditions, thanks to a beneficial restructuring of the rhizosphere community. Similarly, El-Akhdar et al. (2025) observed that the combined use of compost and PGPR significantly improved the N content in corn under different salinity levels, suggesting greater absorption efficiency under adverse conditions.

The *Pseudomonas* sp. + IBA treatment increased the P content, exceeding the control by 14.58%. The treatment with *Pseudomonas* sp. alone showed an increase of 9.85%, while IBA alone showed an increase of 8.75%, both also higher than the control. This supports the effectiveness of PGPR in converting insoluble phosphates into assimilable forms. For example, Yadav et al. (2016) demonstrated that *P. fluorescens* solubilizes tricalcium phosphate, thus contributing to improved P availability. Rezakhani et al. (2019) showed that *Bacillus simplex* UT1 and *Pseudomonas* sp. FA1, combined with Si, significantly improved P absorption in wheat, showing notable increases in P use efficiency and optimizing its assimilation. Therefore, the results could be related to increased microbial activity and reflected in greater P nutritional efficiency in fig cultivation, as *Pseudomonas* produces organic acids such as gluconate and oxalate, phytohormones, and siderophores, which not only stimulate root growth but also mobilize P (Kalayu, 2019).

Bacterial concentration

As for bacterial concentration (BC), there was a significant increase in the *Pseudomonas* sp. + IBA treatment, exceeding the control by 61.4%. Similarly, the treatment with *Pseudomonas* sp. alone showed a 22% increase compared to the control. Meanwhile, the treatment with IBA alone showed an increase of 15.4%, but this was nonsignificant. This indicates successful and persistent bacterial colonization in the rhizosphere, evidencing a synergy between the rhizobacteria and the commercial product with IBA. *Pseudomonas* sp. is known for its ability to adhere to the root system and form biofilms, which facilitates beneficial interaction with the host plant. This coincides with observations by Ansari and Ahmad (2018), who demonstrated in wheat that the formation of biofilms by *P. entomophila* is a crucial advantage for root colonization. Similarly, Pastor et al. (2023) observed in tomatoes that inoculation with a microbial consortium of *Trichoderma harzianum* and *P. putida* positively modified the structure of the soil bacterial community, showing increases in microbial density.

CONCLUSIONS

The combined application of *Pseudomonas* sp. with the rooting agent promoted significant improvements in biochemical and productive variables in fig cultivation, increasing the number of fruits, yield, total soluble solids, and foliar P content. Likewise, the individual application of *Pseudomonas* sp. increased fruit weight and

diameter. The use of *Pseudomonas* sp. alone or in combination with the rooting agent represents a sustainable biotechnological strategy for improving the production and quality of fig crop.

Author contribution

Conceptualization: Y.G.G-M., R.M-V. Methodology: V.R-T., R.M-V. Software: Y.G.G-M. Validation: R.M-V., V.R-T., A.H-P., I.G-P. Formal analysis: Y.G.G-M., R.M-V. Investigation: Y.G.G-M., R.M-V., A.H-P., I.G-P. Resources: R.M-V., V.R-T. Data curation: Y.G.G-M., A.H-P. Writing-original draft: Y.G.G-M. Writing-review & editing: R.M-V., V.R-T., A.H-P., I.G-P., Y.G.G-M. Visualization: Y.G.G-M. Supervision: R.M-V., V.R-T., A.H-P., I.G-P. Project administration: R.M-V., V.R-T. Funding acquisition: R.M-V., V.R-T. All co-authors reviewed the final version and approved the manuscript before submission.

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