The effect of soil properties on the relation between soil management and Fusarium wilt expression in Gros Michel bananas

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Abstract
Aims This study looks whether the response of soil management (liming and nitrogen fertilization) on the incidence of Fusarium wilt (Foc Race 1) in Gros Michel banana (Musa AAA) varies with different soil properties.

Methods The effect of inoculation with Foc Race 1 was studied in a factorial greenhouse trial with soil samples from eight representative soil types from the Costa Rican banana region, two pH levels; and three levels of N-fertilization. After an 8-week period, plant biomass and a disease index were measured.

Results There were significant effects of soil pH and N, and their interactions on disease expression. Low pH levels and high N-fertilization increased the disease expression. The response to changes in soil pH and N-fertilization differed considerably between the different soils.

Conclusions Although soil pH and N influence Fusarium wilt in banana, each soil differs in its response to these soil properties. This complicates the development of standard soil management strategies in terms of e.g., N-fertilization and liming to mitigate or fight the disease.

Keywords Costa Rica · Liming · Nitrogen · Panama disease · Plant nutrition · Soil fertility, Soil pH

Introduction
Fusarium wilt of banana (FWB), also known as ‘Panama disease’ (caused by the soil-borne fungus Fusarium oxysporum f. sp. cubense or Foc), is one of the most critical diseases affecting banana production. Foc Race 1 devastated the subgroup Gros Michel (Musa AAA), which was the main cultivar exported from Latin America and the Caribbean (LAC) during the first half of the 20th century (Ploetz and Churchill 2011; Pocasangre et al. 2017; Dita et al. 2018; Magdama et al. 2020). Foc Race 1 remains a serious problem in small-scale production systems in LAC where the specific traits of the Gros Michel banana are preferred (Pocasangre et al. 2011). The gradual shift of the production systems to the cultivars of the subgroup Cavendish (Musa AAA), which are resistant to Foc Race 1, was a temporary solution to the...
problem (Harper 1950; Stover 1961, 1962; Ploetz 1990; Perez-Vicente 2004). A new, more aggressive strain of the fungus denominated Foc Tropical Race 4 (TR4) is spreading over the world and has recently been reported in LAC (García-Bastidas et al. 2020). Most of the varieties produced in LAC, including the Gros Michel and the Cavendish subgroups, are susceptible to Foc TR4. The spread of Foc TR4 in LAC would have a tremendous impact given the economic and social importance of banana production in the region (Aurore et al. 2009; Pocasangre et al. 2011; Dita et al. 2013).

Conventional control options such as fungicides, replanting or crop rotation are ineffective in controlling or eradicating the disease (Ploetz 2006, 2015; Ordoñez et al. 2015). Other alternatives, such as the evaluation of partially resistant cultivars or breeding new resistant cultivars (Su et al. 1986; Hwang and Ko 2004; Dale et al. 2017), may take a long time to be available for practical implementation. In the short run, it is important to develop a control package that allows farmers to face the disease.

Soil management in agriculture mainly focuses on crop production and rarely considers crop disease control. However, specific soil conditions can suppress diseases in agricultural crops (Janvier et al. 2007). Managing soil properties can (i) influence the soil microbiome and as a result change disease pressure or (ii) influence the crop nutritional status and change the crop’s predisposition to diseases (Dordas 2008; Ghorbani et al. 2008; Huber et al. 2012). Already in 1946, there were reports that liming, fertilization, and crop rotation may be a “cure” to FWB (Taylor (1946) cited by Jones and Morrison 1952). Studies also included flooding of infected areas to aim for soil disinfection or a reduction of the fungus population (Stover 1961). However, through the production of chlamydospores Foc can survive extreme conditions like the anaerobic conditions under flooding (Ploetz 2015). Recently, flooding and irrigation are increasingly being attributed to also increase the spread of the disease (Salacinas 2019). After the shift from Gros Michel to the resistant Cavendish cultivars, research on FWB control was limited for more than 30 years. However, since reports of the new and more aggressive strain Foc TR4 (Stover 1986) were published, research on controlling this disease came back. Studies show that Foc Race 1 and Foc TR4 respond in a similar way and that soil properties play an important role in conducing or suppressing the fungus in banana Domínguez et al. 2008; Orr and Nelson 2018; Segura et al. 2021a, b). However, the results in the literature are found to be inconsistent. This seriously hampers the translation of research results into operational management recommendations. Bananas are grown under a wide variety of agro-ecological conditions (Jaramillo and Vásquez 1990; Stoorvogel and Segura 2018). One possible reason for these inconsistencies could be that the interactions between FWB and soil properties differ with agro-ecological conditions. Soil types have long been known to play a role in this relationship (Stotzky et al. 1961).

This study aims to evaluate the role that soil properties plays on the effect of liming and N-fertilization on the incidence of FWB. Samples of eight representative soil types from the Costa Rican banana regions are evaluated studying the incidence of Fusarium wilt by Foc Race 1 in Gros Michel bananas in a large greenhouse experiment. The results may help to better identify the role of the evaluated soil properties according to the soil types in explaining the inconsistencies in previous results and to support the development soil management strategies to reduce the impact of FWB by Foc Race 1 and help to identify research strategies to similarly develop strategies to control FWB by Foc TR4.

Materials and methods

Banana production in Costa Rica is concentrated in the perhumid Atlantic zone (Fig. 1). Soil conditions vary considerably in the area. Soil types were selected during a survey in the banana regions and they represent the soil variation that was found in the region (Klinkert 2014). The survey included the Caribbean lowlands, which include over 40,000 has of large intensive production of Cavendish cultivars for export (Segura et al. 2015). In addition, the Turrialba region, which has a more extensive production of Gros Michel cultivars for local markets (Ramirez et al. 2010), was incorporated into the study. Soils in the Caribbean lowlands are highly variable (Lopez and Solís 1991; Segura et al. 2015). Soils of the region to the East of the Reventazón river (Fig. 1) are predominantly sedimentary with a high clay content and high fertility. Soils of the region to the West of the Reventazón river originate
from volcanic ashes with a low clay content and medium fertility. Soils in the Turrialba region are deep, well-drained, tropical red soils with a high clay percentage and medium fertility (Dijkshoorn et al. 2005). The climate is tropical and humid with an average annual rainfall of 3000-3500 mm distributed throughout the year.

Selected soils were described in situ and large topsoil samples (0-30 cm) were taken. Two soils were selected from the West of the Caribbean lowlands (S1 and S6), four soils were selected from the larger East of the Caribbean lowlands (S2, S3, S4 and S5), and two soils were selected from the Turrialba region (S7 and S8). The soils were analyzed for pH, acidity, organic matter (SOM), and the concentrations of Ca, Mg, K, P, Zn, Cu, Fe, Cu, and Mn, using the extraction solution Mehlich III (Díaz-Romeu and Hunter 1978; Mehlich 1984). The distribution, location and main properties of the soils are presented in Fig. 1; Table 1.

The experiment was performed in a greenhouse made from insect gaze (in which the plants are exposed to most of the environmental conditions except direct rain and sunlight) at the experimental station of CORBANA in La Rita (132 m.a.s.l., 10°15’54” latitude N, 83°46’26” longitude W, maximum temperature of 35 °C and minimum temperature of 17 °C, average temperature of 28 °C with an 85% relative humidity and approx. 12 h of daylight). The factorial design included topsoil samples from 8 soil types, with and without inoculation of Foc Race 1, 2 levels of soil pH, 3 levels of N-fertilization, and 3 replications resulting in a total of 288 pots. Two contrasting soil pH levels were tested: (1) pH\textsubscript{low} with a pH of 5.2 or lower, and (2) pH\textsubscript{high} with a pH equal to or higher than 6.0. Soil pH was adjusted
to these target levels by applying a hydrochloric acid solution (10% HCl) to decrease pH or lime (CaCO₃) to increase soil pH. Soil pH was adapted before any other treatment were applied. In each case, the acid or alkaline units of solution required to achieve the lower and the higher pH were calculated before liming and/or acidifying. Both processes liming and acidification were done through manually mixing the soil and the amendments. Soil pH was analyzed before each treatment and eight days after the liming or the acidification treatments. In the low pH treatment, pH levels between 4.0 and 5.1 were measured and in the high pH treatment, pH levels between 6.2 and 6.8 were measured.

Hardened, approximately 3-month-old, tissue culture banana plants (Musa AAA, subgroup Gros Michel) were used in the experiment. The plants grew in a standard potting mix before the experiment. Banana plants in young stages from tissue culture, such as the ones used in the experiment, are more sensitive to Foc infestation (Brake et al. 1995). This condition ensured the plant’s response to the disease according to the treatments. Two levels of Foc Race 1 inoculation were achieved by root dipping (Dita et al. 2010; García-Bastidas et al. 2014, 2016; Ordoñez et al. 2016): (1) In₀: a control, 30 min in clean water, and (2) In₁: the inoculated group, 30 min in a solution of water with 10⁶ conidia mL⁻¹ of Foc Race 1. The fungus strain was collected from Costa Rican soils, tested, and cultivated by CORBANA’s Center of Biological Control. It should be noted that plants in In₀ could still be infected with Foc Race 1 present in the soil samples. It is widely accepted that Costa Rican banana soils are infested with Foc Race 1. As the soil samples were not sterilized before the experiment in order to maintain a certain level of the natural soil microbiome, the plants in the control would be exposed to Foc Race 1 if it is present at the soil. Immediately after the inoculation, plants were separately planted in 2 L pots (one plant per pot). Three levels of N were achieved through weekly differentiated N-fertilization with ammonium nitrate (AN, 33.5% N): (1) Nlow with no N addition; (2) Nmed with 0.08 g N plant⁻¹ week⁻¹ (0.64 g N plant⁻¹ in the total experimental period) supplied through 0.24 g of AN plant⁻¹ week⁻¹; and (3) Nhigh with 0.25 g N plant⁻¹ week⁻¹ (2.00 g N plant⁻¹ in total experimental period) of N supplied through 0.75 g of AN plant⁻¹ week⁻¹. These N levels were respectively achieved through applications of 300 mL of solutions of AN in water with concentrations of respectively 0.00 g L⁻¹ N, 0.14 g L⁻¹ N and 0.43 g L⁻¹ N, two times week⁻¹. Nmed emulated the average N requirement of plants during the first 10 weeks after planting in real field conditions. No other nutrients or agro-chemicals (e.g., fungicides, insecticides, etc.) were applied to the plants.
The experimental period was eight (8) weeks long and at the end of this period, total (above ground plus roots) fresh biomass (g plant\(^{-1}\)) was measured. In addition, a non-intrusive way to measure the disease according to the management of the soil properties was following the development of the wilting. A disease index (DI) was obtained adapting the McKinney’s formula (McKinney 1923) that was also used by Haddad et al. (2018) and Rocha et al. (2020) in the same way. However, in this case, it was based on the number of sick plants and the wilted leaves: \[ \text{DI}(\%) = 100 \times \frac{\sum (p_{\text{dis}}/p_{\text{tot}}) (l_{\text{dis}}/l_{\text{tot}})}{\text{total of plants}} \]

where; \( p_{\text{dis}} = \) number of diseased plants; \( p_{\text{tot}} = \) total of plants; \( l_{\text{dis}} = \) number of leaves with symptoms; and \( l_{\text{tot}} = \) total number of leaves. The presence of the typical symptoms of the wilting of the leaves in previously inoculated plants is reported as a valid element to corroborate the presence of the disease in bananas (Dita et al. 2010; García-Bastidas et al. 2014, 2016, 2020; Hung et al. 2018). Plant biomass from In\(_0\) and In\(_1\) and the wilting per plant data were analyzed using a factorial analysis of variance, which considered involved factors and their interactions: soils, inoculation, soil pH and N for biomass, and only the soil, pH and N for the DI. The differences in the results according to the factors were evaluated through a Tukey’s analysis.

**Results**

Natural effect of soil and soil pH in not inoculated plants

The control group (In\(_0\)) showed the effect of the soil, the pH and N management in the plant biomass (Fig. 2). There were considerable differences in the mean biomass per plant according to soil (P < 0.001). Bananas grown on soils from the West and Turrialba showed the best mean performance with the plants grown on pH\(_{\text{high}}\) being significantly (P < 0.001) larger than plants grown on pH\(_{\text{low}}\). However, two soils gave more biomass in plants from pH\(_{\text{low}}\). The N level did

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**Fig. 2** Biomass from control, not inoculated (In\(_0\)) Gross Michel (Musa AAA) banana plants after eight weeks growing in a greenhouse experiment with eight representative soils from the Costa Rican banana regions (West, East and Turrialba) with two soil pH levels (low and high) and three N-fertilization levels (low, medium and high). Bars represent the error standard.
not significantly affect the biomass over the eight soils \( (P \geq 0.730) \). The effect of N fertilization differed per soil where in some cases the increased nitrogen levels resulted in an improved performance but in other cases there was a decline in the performance with the increases in N.

Interaction of inoculation, soil pH and N on the plant biomass and disease index (DI)

Except for S2, S3 y S4 in \( \text{pH}_{\text{low}} \), the biomass in \( \text{In}_{1} \) was significantly lower than \( \text{In}_{0} \) for almost all the soils and pH levels (Table 2). The detrimental effect of the interaction between \( \text{pH}_{\text{low}} \) and inoculation was evident in plants from the three regions. Despite following the trend of a higher biomass in \( \text{pH}_{\text{high}} \), the mean biomass in \( \text{In}_{1} \) was contrastingly lower against the control for both pH levels. The effect of the inoculation according to the soil pH levels and expressed as a decline in plant biomass is presented in Fig. 3. It is showed that for most soils a pH increase led to higher loss in biomass due the disease. Differences in pH levels resulted in a very different response to the inoculation of the disease for the different soils.

Inoculation led to a significant decline in plant biomass in almost all combinations of soil and N level (Table 3). With increasing N-fertilization, typically, the decline in biomass due to inoculation increased. A higher effect of the interaction of the soil and the inoculation was evident in all soils in the three N levels. The interaction of the inoculation and the N level was expressed in different trends according to the soils and N levels and the average biomass (Fig. 4). In some cases, the loss in biomass due the N-level was higher in \( \text{N}_{\text{low}} \) as it took place in S6, S3 S4 and S7. The higher loss in biomass with \( \text{N}_{\text{med}} \) only took place in S2 and S8. Soils 1 (S1) and 5 (S5) showed the higher loss in biomass according to the N levels in \( \text{N}_{\text{high}} \).

The single effect of soil pH was significant in the DI expression with a higher average wilting in \( \text{pH}_{\text{high}} \) in all soils. The higher DI is offset by the increased performance of the plant at \( \text{pH}_{\text{high}} \) still leading to a net benefit of the increased pH. The DI according to the N levels (average from all the soils) was not significant, and its trend was more erratic. Plants from \( \text{pH}_{\text{high}} \) remained more biomass and expressed a lower DI. The interaction pH x N was significant in the DI and average biomass (Fig. 5). There was a lower biomass in \( \text{N}_{\text{high}} \) and \( \text{pH}_{\text{low}} \). From the possible combinations, a higher DI took place in \( \text{N}_{\text{med}} \) and \( \text{pH}_{\text{high}} \).

**Table 2** Decline in biomass (g±s.e.) of Gros Michel (Musa AAA) banana plants eight weeks after the inoculation with Foc Race 1 and growing in a greenhouse experiment with eight representative soils from the Costa Rican banana regions (West, East and Turrialba) with two soil pH levels (low and high)

| Code | Region and soil | \( \text{pH}_{\text{low}} \leq 5.2 \) | Decline 1 | \( \text{pH}_{\text{high}} > 6.0 \) | Decline 2 |
|------|-----------------|-----------------|----------|-----------------|----------|
|      | \( \text{In}_{0} \) | \( \text{In}_{1} \) | \( (\text{In}_{0} - \text{In}_{1}) \) | \( \text{In}_{0} \) | \( \text{In}_{1} \) | \( (\text{In}_{0} - \text{In}_{1}) \) |
| West | S1              | 38.5±1.3        | 6.5±0.4  | 32.9***          | 12.6±1.9 | 19.7***         |
|      | S6              | 21.6±0.8        | 11.5±0.5 | 10.1**           | 18.0±1.3 | 12.3**          |
| East | S2              | 13.0±0.7        | 6.5±0.5  | 6.5              | 26.0±1.7 | 11.0±0.7        | 15.0**         |
|      | S3              | 12.1±0.9        | 7.2±0.6  | 4.9              | 42.1±2.4 | 24.4±1.2        | 23.4***         |
|      | S4              | 4.4±0.2         | 1.7±0.1  | 2.7              | 27.8±0.8 | 15.6±1.3        | 12.2**          |
|      | S5              | 12.5±0.8        | 2.5±0.2  | 10.0**           | 40.5±0.1 | 23.7±1.5        | 16.8***         |
| Turrialba | S7      | 40.5±1.7        | 10.3±0.9 | 30.2***          | 37.1±1.1 | 16.5±1.2        | 20.6***         |
|      | S8              | 27.3±0.9        | 12.9±1.2 | 14.4***          | 39.0±0.8 | 13.8±0.7        | 25.5***         |
|      | Mean            | 21.2±0.6        | 7.3±0.2  | 13.9*            | 34.4±0.7 | 17.0±0.5        | 17.4**          |

**Discussion**

Soil properties were found to have a strong effect on plant performance under natural (not inoculated) and inoculated conditions. Losses in biomass in \( \text{In}_{1} \) can be attributed to the detrimental effect of Fusarium wilt on the plants. Biomass in the inoculated plants was lower because of the loss of the tissues produced by the disease and the minor biomass production due
the effect of the disease. Soil pH_{high} increased the biomass per plant for both In_{0} and In_{1} treatments. Besides, pH x inoculation interactions on the effect of the disease were stronger in pH_{low}. This could imply a direct (Almeida et al. 2018), indirect effect, or both, of soil pH on the incidence of the disease. The role of pH as an indicator of soil health in banana and its influence in soil suppressiveness is known (Pattison et al. 2008; Geense et al. 2015; Segura et al. 2015). Low soil pH (less than 5.2) apparently can stimulate the pathogen activity in the soil due to a detrimental effect on soil diversity. However, this was not evaluated in this trial. The lower pH also limits plant nutrient and water uptake (White 2012). This effect can increase banana predisposition to diseases led both by a higher Foc activity and a limited capacity to perform physiological processes against the infestation under a lower (than 5.2) soil pH.

Despite the differences according to the interaction of the different soils and inoculation, and the general effect of pH on plant response, the detrimental interaction of lower pH x inoculation was higher in various soils. The highest effect of the disease expressed as a lower biomass found in the eastern region could

| Region and soil | Code | N_{low} | N_{med} | N_{high} | decline^1 | decline^2 | decline^3 |
|----------------|------|---------|---------|----------|------------|------------|------------|
| West           | S1   | 36.5±1.8| 10.1±0.9| 26.4***  | 38.5±2.5   | 7.8±0.9    | 30.7***    |
|                | S6   | 26.4±1.3| 15.7±0.8| 10.7*    | 22.4±0.9   | 19.9±1.3   | 2.5        |
| East           | S2   | 18.5±1.3| 12.5±1.1| 6.0      | 15.4±0.9   | 8.5±0.7    | 6.9        |
|                | S3   | 24.5±3.4| 19.2±2.4| 11.0     | 26.5±2.1   | 16.5±1.3   | 10.0*      |
|                | S4   | 14.4±1.3| 11.2±1.5| 22.3     | 17.9±2.1   | 9.2±1.1    | 8.7        |
|                | S5   | 27.6±2.3| 11.0±1.2| 16.6**   | 26.9±3.1   | 11.3±1.2   | 15.6**     |
| Turrialba      | S7   | 35.0±1.3| 15.9±0.8| 19.1**   | 41.4±1.5   | 14.4±0.9   | 27.0***    |
|                | S8   | 33.0±1.6| 12.8±2.0| 20.2***  | 31.6±1.1   | 16.5±1.5   | 15.1**     |

*** P≤ 0.001; ** P≤ 0.010; * P≤ 0.050
be due to the specific characteristics of these soils. Although all of them have a high natural fertility, plants from those soils were more sensible to the disease, especially in pH_low. The natural soil properties as a package can play a role in the plant status and it can also define the plant’s predisposition to the disease. Previous reports indicated that chemical and physical soil conditions would be linked to soil conduciveness of Fusarium wilt (Scher 1980; Domínguez et al. 2008). However, generalization this by analyzing the interaction of soil properties and the disease in one single soil can lead to misunderstandings and inconsistent conclusions. Interactions of the soils with pH, N and other abiotic (and biotic) properties, such as e.g. the higher clay content, Fe and Mn, would be involved in the plant’s response. This knowledge is crucial when considering soil management to deal with or control Fusarium wilt in banana.

Despite the disturbance of the soil samples during sampling and processing, the plant’s response to the disease can be related to specific ecological condition. Soils with a lower concentration of SOM in the eastern region, for instance, showed the highest detrimental effect of the disease. Interactions of the SOM and other biotic soil properties (not analyzed) would be playing a role in this result. In the western region,
plant response to inoculation showed considerable variation. In fact, the best plant performance under infected and pH_{low} conditions took place on soils with the highest soil SOM content (S6, S7 and S8). Results allow us to define different degrees of risk to the disease, according to the studied soils. However even in a same banana region, the magnitude of the response to the disease according to the management of pH and N differed depending on the soil. Nevertheless, practices such as maintaining a higher pH (with liming or just avoiding soil acidification) and increasing SOM through management appear to be preventive measures that can decrease plant predisposition to the disease. This seems to be a standard recommendation for all banana locations could be complex or not applicable. With this information, strategies based on soil management to prevent and deal with Foc can be implemented, in this case specifically for the Costa Rican reality.

The interaction of soil type with pH and N management in the disease expression should be studied thoroughly. How and where this interaction is more significant in the disease can define the strategy that should be implemented in the crop management. Soil pH_{low} and N_{high} treatments predisposed the plant to be more easily infected and more affected by the disease. It appears that soil pH is a primary factor in the plant’s predisposition to the disease in most of the studied soils. N inputs have important impacts in soil conditions. It is likely that the differences in the disease according to the N levels are related to the residual of the nature N in the soil samples (not evaluate). The SOM concentration in the soil could play a role as a N-organic source.

At the same time, N ammonia sources are recognized as a main cause for a drop in pH in agricultural soils. Besides the effect of the N source in the diseases (Huber and Watson 1974; Stover and Simmonds 1987). Therefore, an integral management of soil properties to alleviate or prevent Fusarium wilt should include pH management and choosing less acidity N sources. In addition, it is necessary to include a soil N analysis in order to define more accurate N recommendation for banana plantations.

More integral soil conditions can be playing a role in plant response to the disease. Furthermore, the role that the soil origin played in the plant’s response to disease was evident. Although it agrees with the response in terms of suppression or conduction of the disease according to the soil type in Australia (Bowen et al. 2019), the results that we found allowed us to see a complex interaction of the soil (may be the soil type), its integral properties and the incidence of the disease. The influence of specific soil properties (chemical, physical, and microbiological) linked to the soil origin in each banana region can be part of the scenario of the natural banana’s response to the disease. This could be the reason why previous studies have shown an erratic behavior of the disease according to soil management. Probably those results could depend on the region where plantations are established in the different banana locations. The specific study of at least soil by its origin and its conditions on banana predisposition to Fusarium wilt in each location or banana region is necessary to better understand the role of soil properties in FWB. Even ecological and environmental aspects of each region can be playing a role in the incidence of the disease.

Conclusions

Soil properties like pH and N play an important role in banana predisposition to Fusarium wilt and in the disease incidence. However, this relation is strongly influenced by soil origin, i.e., other soil inherent properties. This complexity clearly hampers the development of uniform management recommendations. Nevertheless, general trends are found. An increase in soil pH supports, in almost all cases, a reduction of Fusarium wilt incidence in banana. For the development of more specific soil management strategies, it appears to be necessary to view the soil as an integral system of physical, chemical, and biological properties rather than looking at individual soil properties with their thresholds. This complexity could be the cause for the inconsistencies that were found in the literature with respect to the role of soil properties in suppressing or conducing FWB.

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Author contribution All the authors took part in conceptualization of the research and editing the manuscript and consent its publication. RS and JAS performed the greenhouse experiment in Costa Rica. RS and JJS performed the results analysis and RS wrote the manuscript. JSS and JS made important inputs to improve the final version of the manuscript.

Declarations

Ethical statement This manuscript is not submitted to another journal. The manuscript is original, and it is not published elsewhere partially or in full, in any form or language. Besides, it does not concern an expansion of previous work. The study is not split up into several parts to increase the quantity of submissions and submitted to various journals or to one journal over time. Results are presented clearly, honestly, and without fabrication, falsification, or inappropriate data manipulation. Data was collected from the greenhouse experiment and managed with statistical software with total honesty and transparency. No data, text, or theories by others are presented as if they were the author’s own. All collected data, and the performed analysis are available (as a supplementary file). Proper acknowledgements to other works are given. This piece of work respects third parties’ rights such as copyright and/or moral rights.

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