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Influence of phosphite and phosphate fertilisers at three different pH levels under a floating system on the growth, yield, and nutrient concentration of broccoli

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Abstract: This study was conducted to determine the effects of phosphite and phosphate fertilisers on broccoli's yield, growth, and nutritional status *(Brassica oleracea* L. var. *italica* cv. 'Fiorentino'). In factorial combinations, experiments were conducted at three pH levels (5.5, 7.0, and 8.5) and three phosphorus source levels [Pi (phosphate), Phi (phosphite), and Pi + Phi]. Phi had a statistically significant negative effect on yield, producing immature and button-like flower heads. Decreases in chlorophyll index, Fv/Fm [ratio of variable fluorescence (Fv) to maximum fluorescence (Fm)], and leaf area were observed using the Phi treatment. These results indicate that fertilisation with Phi alone did not affect plant growth and yield. P (phosphorus) and K (potassium) concentrations in plants were increased by root fertilisation with Phi (floating system), but did not affect growth characteristics; increased Pi uptake in Phi-treated plants increased P and K concentrations in the sink source, resulting in reduced growth, phytotoxicity, and no head formation. While it may appear that Phi is upsetting the balance of solution nutrients and stressing the plants, the Phi-induced stress condition was identified by measuring proline levels and electrolyte leakage. Thus, phosphite could not be used as a P source for plants. However, Phi could be used as a plant nutrient source combined with conventional Pi fertilisers.

Keywords: accumulation; head formation; proline; toxicity; uptake

Broccoli (*Brassica oleracea* var. *italica*) is an important winter vegetable plant belonging to the *Brassicaceae* family and is consumed worldwide due to its high vitamin C content (Latté et al. 2011). It also contains antioxidants, anti-cancer properties, and other health benefits (Latté et al. 2011).

Phosphite (Phi; PO_3^{3}; valence of +3), a reduced form of phosphate (Pi; PO_4^{3}; valence of +5) that are being widely marketed either as an agricultural fungicide, biostimulant or as a superior source of plant P nutrition (Gómez-Merino, Trejo-Téllez 2015; Sutradhar et al. 2019). Estrada-Ortiz et al. (2013) found

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positive effects of Phi on strawberry fruit quality and induction of plant defence mechanisms, which have also been reported in several plant species and cultivars (Gómez-Merino, Trejo-Téllez 2015). Numerous publications indicate that leaves and roots can absorb Phi well, but have no benefit for plants as a P fertiliser source (Vinas et al. 2020). When tomato, pepper, strawberry, spinach, and maize plants were treated with Phi instead of Pi fertiliser, they showed a notable decrease in growth (Bozzo et al. 2004; Moor et al. 2009; Thao, Yamakawa 2009). Different viewpoints have been expressed on the uptake, transport, and metabolism of Phi (Orbović et al. 2008; Ratjen, Gerendás 2009), and it appears that Phi is readily transported by the xylem and phloem (Thao, Yamakawa 2009). According to earlier studies, it seems that while Phi can be readily absorbed and transferred by plants, it cannot be oxidised or metabolically processed (Carswell et al. 1997; Förster et al. 1998; Ticconi et al. 2001). The study's findings demonstrated that plant cells are unable to oxidise Phi to Pi (Danova-Alt et al. 2008). In research, McDonald et al. (2001a) proposed that Phi could exacerbate the consequences of Pi deficit by misleading cells lacking Pi into assuming they have enough Pi. Therefore, the accumulation and toxicity of Phi in plants are likely linked to decreases in Pi assimilation and/or the inability to metabolise Phi or its oxidation to Pi in the cell. Recently, reports and research papers have emphasised the bio-stimulant role of Phi (Gómez-Merinio, Trejo-Téllez 2015; Sutradhar et al. 2019). Foliar application of potassium phosphite to soybean plants under water deficit and high radiation stress-induced plant tolerance to stress conditions (Batista et al. 2023). According to molecular and enzymatic research findings, Phi in plants disrupts the enzymes involved in Pi metabolism and reduces the induction of Pi-dependent enzymes (Ticconi, Abel 2004; Varadarajan et al. 2002). In comparison to conventional Pi fertilisation, Moor et al. (2009) found that the application of Phi had no effect on the growth or yield of strawberries but improved fruit quality by promoting the synthesis of ascorbic acid and anthocyanins. The negative effects of Phi on plant growth and yield have been demonstrated in a number of studies with annual plants, including *Spinacia oleracea* (Thao et al. 2008), *Cucurbita pepo* (Ratjen, Gerendás 2009), *Brassica rapa* (Thao, Yamakawa 2010), *Cucumis sativus* (Constán-Aguilar et al. 2014) and *Solanum lycopersicum* (Vinas et al. 2020). Numerous studies have shown that Phi has been found to display systemic effects and high chemical stability in plant tissues, though it also shows great mobility throughout the whole plant. This mobility facilitates the penetration and transport of the foliar-applied Phi to the rest of the plant, including the roots (Lee et al. 2005; Ávila et al. 2012). The effect of Phi on plant growth is strongly dependent on the Pi nutrient status of the plants, and those insufficiently fertilised with Pi are at high risk of the deleterious effects of Phi (Thao, Yamakawa 2009). P-limited plants are susceptible to Phi and show toxicity symptoms such as reduced growth and chlorosis of the leaves (Ratjen, Gerendás 2009; Thao et al. 2009).

This study aimed to investigate the effects of the supply of Phi and Pi at different levels of pH in a hydroponic system (floating) on the growth, yield, P status, and nutrient concentrations of broccoli plants.

MATERIAL AND METHODS

Plant materials and growth conditions. The seeds (*Brassica oleracea* L. var. *italica* cv. 'Fiorentino') were germinated for 20 days in plastic boxes filled with perlite. The seedlings were then transferred to black buckets containing 8 L of aerated nutrient solution. The nutrient solution contained (in mg/L): 65 KNO_3, 32 $Ca(NO_3)_2·4H_2O$, 9 $MgSO_4·7H_2O$, 8 NH_4NO_3. The micronutrients were 0.2 FeEDTA, 0.08 $MnSO_4·4H_2O$, 0.05 $ZnSO_4·5H_2O$, 0.02 $MoO_4·2H_2O$, 0.08 H_3BO_3, and 0.02 $CuSO_4·5H_2O$. The experiment consisted of 9 treatments with three pH values (5.5, 7.0, 8.5) factorially combined with three P sources (Pi, Phi, and Pi + Phi), each replicated four times. Plants were treated with three P sources: Pi (2.5 mg/L, KH_2PO_4), Phi (2.5 mg/L, liquid NPK growth products 0 : 29 : 26 "TKO" PHOSPHITE), and a combination of phosphite with traditional phosphate Pi + Phi (2.5 mg/L, 50% Pi + 50% Phi). Solutions were changed weekly in the first 4 weeks, and after that, changed every fifth day. Plants were harvested at 10 weeks after transplanting.

Analysis of the sample. The leaves were removed to measure weight. After weighing (fresh weight, FW) the leaves, stems, roots, and heads, the dry weight (DW) was determined after drying the samples at 80 °C for three days. The leaf area was measured using a leaf area meter (Li-Cor, model Li-1300, USA). Chlorophyll index and chlorophyll fluorescence were measured (2 or 3 leaves for any plant) after four months in the fully developed leaves us-

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ing the chlorophyll meter apparatus (SPAD-502, Minolta, Japan) and chlorophyll fluorescence meter (Hand pea, Hansatech, UK), respectively. The proline content in fully developed leaf tissues was measured using ninhydrin (Bates et al. 1973).

The procedure of electro-leakage measurement was based on the method of Lutts et al. (1996). Electrical conductivity, EC_1 and EC_2, are the measured electrical conductivity before and after boiling the samples; electro-leakage was calculated as EC_1/EC_2 and expressed as a percentage. The concentrations of total P in the samples were analysed colourimetrically using a spectrophotometer (Motic, CL-45240-00, China) at a wavelength of 436 nm after staining with an ammonium molybdate-vanadate solution (Gericke, Kurmies 1952), and K was analysed using a flame photometer (Flame Photometric 410, Iran) (Varley 1966). The concentration of total N (nitrogen) in the youngest fully expanded leaves was determined by the Kjeldal method (Chapman, Pratt 1962) and the Zn (zinc) content by an atomic absorption spectrophotometer (Perkin Elmer, Model 110, USA).

Statistical analysis was performed using analysis of variance in SPSS version 21.0 software, and means were separated by Duncan's test at the 5% level. The average values results of the trait were obtained from two or three plants as replicates.

RESULTS

The vegetative characteristics (fresh weight and dry weight of leaf, root, and stem) of the P source at different pH values are shown in Table 1. The results showed that broccoli's growth response depended on both P and pH. Plants treated with both P sources showed intermediate results (Table 1). In Figure 1, the results show that the application of Phi resulted in a significant decrease in yield, but Pi at a pH of 5.5 had the highest fresh weight (yield). Increasing the pH from 5.5 to 8.5 in the Pi and Pi + Phi treatments decreased the yield from 237.25 g/plant to 158.50 g/plant in the Pi source. When Phi was applied at all three pH values, the yield of broccoli was less than 40 g/plant, and this treatment resulted in the formation of small heads, and some plants did not have heads (inflorescence). Using Phi reduced the plants' leaf area

Table 1. The effect of P source in different pH levels on the vegetative characteristics of the broccoli plant

Treatment	Fresh weight (g/plant)			Dry weight (g/plant)		
	leaf	root	stem	leaf	root	stem
Pi						
pH 5.5	332.67^{a}	167.33^{a}	155.00^{a}	40.12^{a}	6.93^{bc}	14.04^{abc}
pH 7	268.00^{b}	154.00^{ab}	124.33^{bc}	24.82^{b}	7.20^{abc}	10.98^{bcd}
pH 8.5	258.67^{b}	139.00^{bc}	138.67^{ab}	25.16^{b}	8.81^{a}	14.57^{ab}
Phi						
pH 5.5	187.00^{c}	136.67^{c}	114.67^{bc}	19.87^{b}	8.01^{ab}	11.04^{bcd}
pH 7	266.33^{b}	129.67^{c}	111.33^{c}	21.93^{b}	6.83^{bc}	10.16^{cd}
pH 8.5	162.00^{c}	99.00^{d}	82.00^{d}	17.02^{b}	5.49^{c}	10.44^{cd}
Pi + Phi						
pH 5.5	291.00^{ab}	143.67^{bc}	159.67^{a}	25.17^{b}	8.77^{a}	16.47^{a}
pH 7	266.00^{b}	101.33^{d}	84.67^{d}	19.79^{b}	6.07^{c}	7.83^{d}
pH 8.5	248.00^{b}	105.00^{d}	99.00^{cd}	18.75^{b}	6.41^{bc}	8.03^{d}
ANOVA						
P source	16 410.25**	3 572.25**	3 153.92**	283.31^{ns}	1.76^{ns}	19.38*
pH	6 267.81**	2 774.37**	1 012.03**	156.30^{ns}	2.22^{ns}	40.63**
P source × pH	4 452.87**	341.42**	2 709.53**	63.19*	6.06**	22.09**
Error	590.00	82.51	210.00	21.85	0.84	4.47

P – phosphorus; Pi – phosphate; Phi – phosphite; ns – non-significant; *,**significance at 0.05, 0.01 probability level, respectively, established by Duncan's multiple range test; $^{a–d}$means followed by the same superscript letter within the columns are not significantly different

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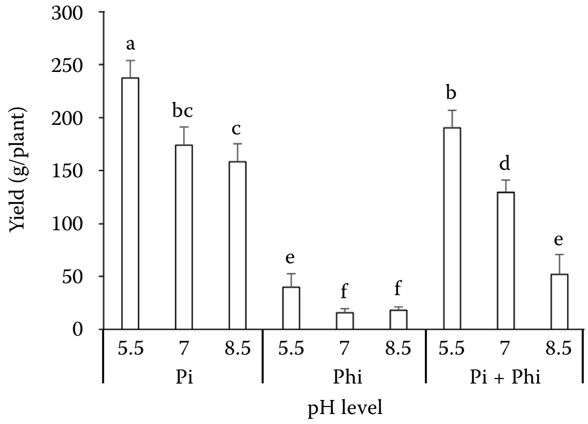


ANOVA (yield)

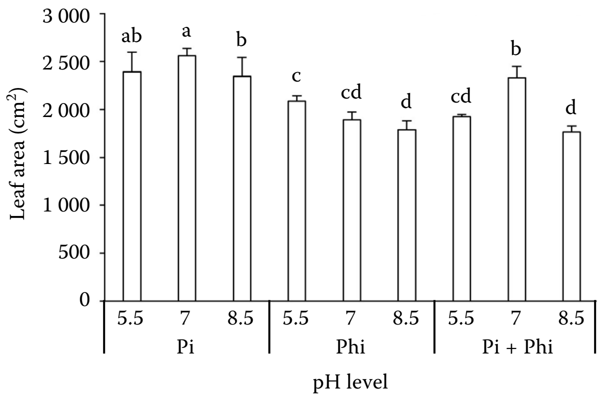
P source	73 592.361**
pH	12 168.361*
P × pH	12 033.736*
Error	3 362.500

Figure 1. The effect of Phi and Pi in different pH levels on broccoli yield (g/plant)

P – phosphorus; Pi – phosphate; Phi – phosphite; *,**significance at 0.05, 0.01 probability level, respectively; a–f different lowercase letters represent significance

compared to the Pi. The treatments with equal ratios of Pi and Phi (Pi + Phi) also reduced leaf area at pH 5.5 and 8.5, except at pH 7 (Figure 2).

The chlorophyll index data showed that Pi at pH 5.5 had the highest value for this parameter (63.13 SPAD unit). The pH of 8.5 produced the least amount of chlorophyll in all three P-feeding scenarios. There was a 50% rise in the highest chlorophyll index (Pi, pH = 5.5) compared to the lowest (Phi, pH = 8.5) (Figure 3A). The chlorophyll index was observed to decrease with increasing pH in the Pi and Pi + Phi treatments. When Phi was used as the P fertiliser source for broccoli in the floating system, chlorophyll fluorescence was less than 0.8. However, for Pi and Pi + Phi treatments at pH 5.5 and 7, these values were above 0.8 and not significantly different from each other (Figure 3 B).

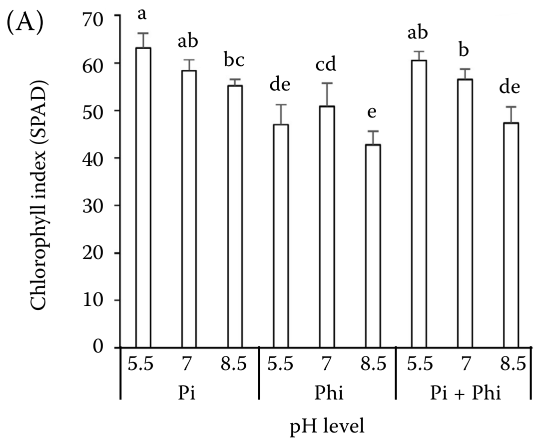


ANOVA (chlorophyll index)

P source	678 575.60**
pH	195 759.54**
P × pH	83 810.148**
Error	13 584.470

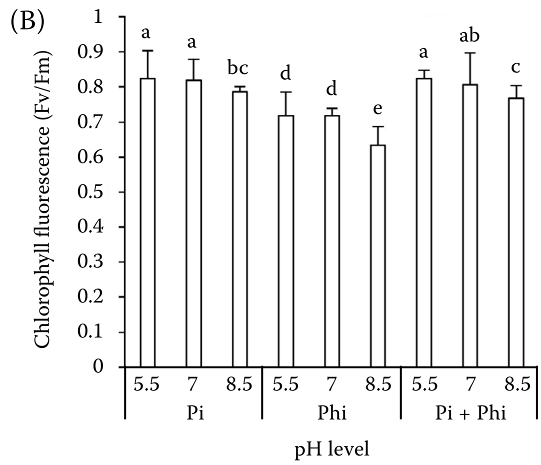
Figure 2. The effect of Phi and Pi in different pH levels on the leaf area of the broccoli plant

P – phosphorus; Pi – phosphate; Phi – phosphite; *,**significance at 0.05, 0.01 probability level, respectively; a–d different lowercase letters represent significance



ANOVA (chlorophyll index)

P source	334.1**
pH	181.0**
P × pH	26.04*
Error	8.309



ANOVA (chlorophyll fluorescence)

P source	0.040**
pH	0.010**
P × pH	0.001*
Error	0.001

Figure 3. The effect of Phi and Pi in different pH levels on (A) chlorophyll index and (B) chlorophyll fluorescence of broccoli plant

P – phosphorus; Pi – phosphate; Phi – phosphite; *,**significance at 0.05, 0.01 probability level, respectively; a–e different lowercase letters represent significance

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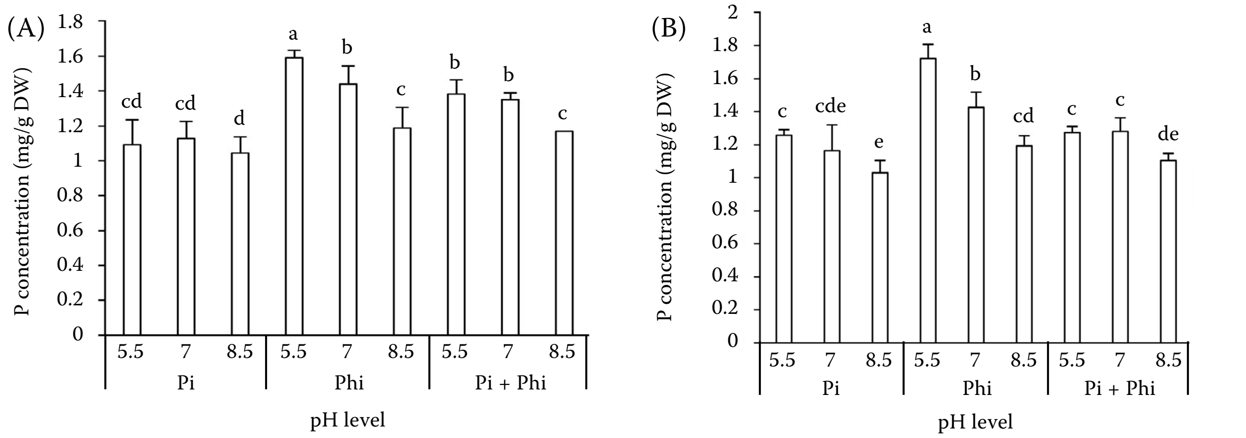
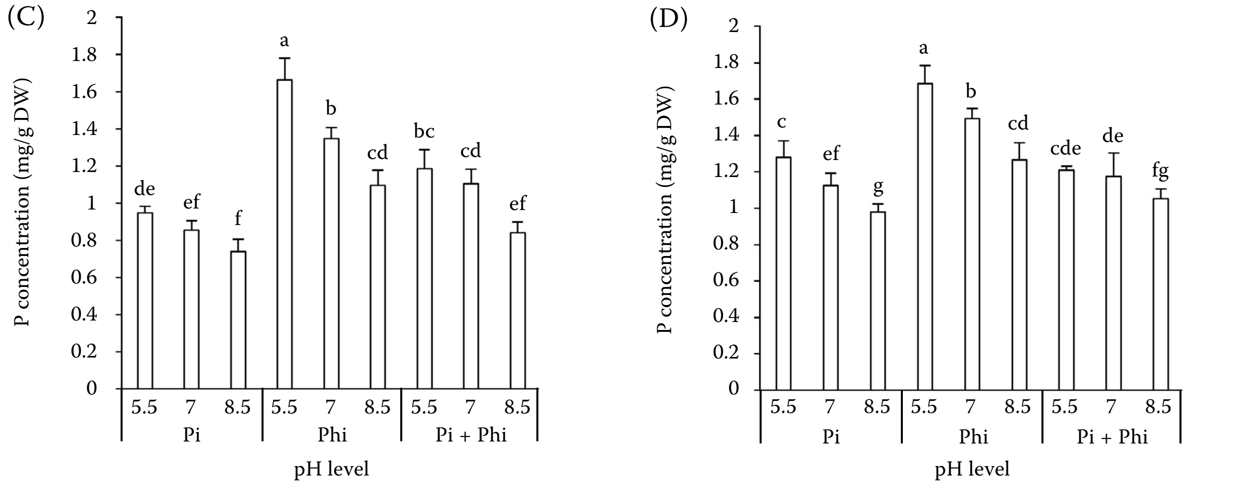
The Phi at pH 5.5 has been shown to have the highest concentration of P in all four measured parts (leaf, head, stem, and root), with an amount of more than 1.5 mg/g DW in each measured component (Figure 4). P status in broccoli leaves showed significance in the three pH levels in the Phi treatment, but this was not the case with the Pi treatment (Figure 4A). Results for head P concentrations showed significant differences between pH 5.5 and 8.5 for all three P sources, but only Phi showed a significant difference between pH 7 and 5.5 (Figure 4B). The broccoli stem section had the lowest P concentration of any other part of this plant in the Pi fertiliser source. Therefore, in all three pH levels, the amount of P was less than 1 mg/g DW (Figure 4C). P concentrations in broccoli roots completely depended on pH; in all three P treatments, pH 5.5 showed the highest concentration (Figure 4D). In general, the P concentration of the four broccoli parts depended on the P source used and was determined from lowest to highest P content in the form of Pi, Pi + Phi, and Phi (Figure 4). Similar to the P concen-



ANOVA (P concentration in leaf)
P source 0.236**
pH 0.121**
P × pH 0.024**
Error 0.004

ANOVA (P concentration in head)
P source 0.216**
pH 0.217**
P × pH 0.032**
Error 0.007



ANOVA (P concentration in stem)
P source 0.624**
pH 0.317**
P × pH 0.028*
Error 0.009

ANOVA (P concentration in root)
P source 0.356**
pH 0.193**
P × pH 0.013**
Error 0.003

Figure 4. The effect of Phi and Pi in pH levels on the concentrations of P in the (A) leaf, (B) head, (C) stem and (D) root of broccoli plant

P – phosphorus; Pi – phosphate; Phi – phosphite; DW – dry weight; *,**significance at 0.05, 0.01 probability level, respectively; $^{a–g}$different lowercase letters represent significance

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tration, the K concentration was highest in the Phi at pH of 5.5, with concentrations 62.64, 43.69, 23.53, and 36.23 mg/g DW measured in the leaf, head, stem, and root parts, respectively (Figure 5). When the pH was increased from 5.5 to 7, the Pi source did not show significant changes in K concentrations in broccoli leaves, but changes in the Phi source were observed (Figure 5A). In broccoli heads compared to leaves at pH 5.5, K concentration decreased from 62.46 to 43.65 mg/g DW for Phi and from 54.91 to 31.95 mg/g DW for Pi + Phi, but increased from 25.66 to 34.26 mg/g DW for Pi (Figures 5A, B). In the head, K concentration was significant at the pH level of the Pi source. In general, K concentrations varied in different parts of the broccoli plants. The lowest K content was found in the stem section of all three P sources, with a range of 10 to 23.53 mg/g DW between the lowest (Pi, pH = 8.5) and highest (Phi, pH = 5.5) concentrations (Figure 5C). The amount of K in the roots was 26 mg/g DW in both Pi and Pi + Phi treatments at pH 5.5 and 36 mg/g DW in the Phi (Figure 5D).

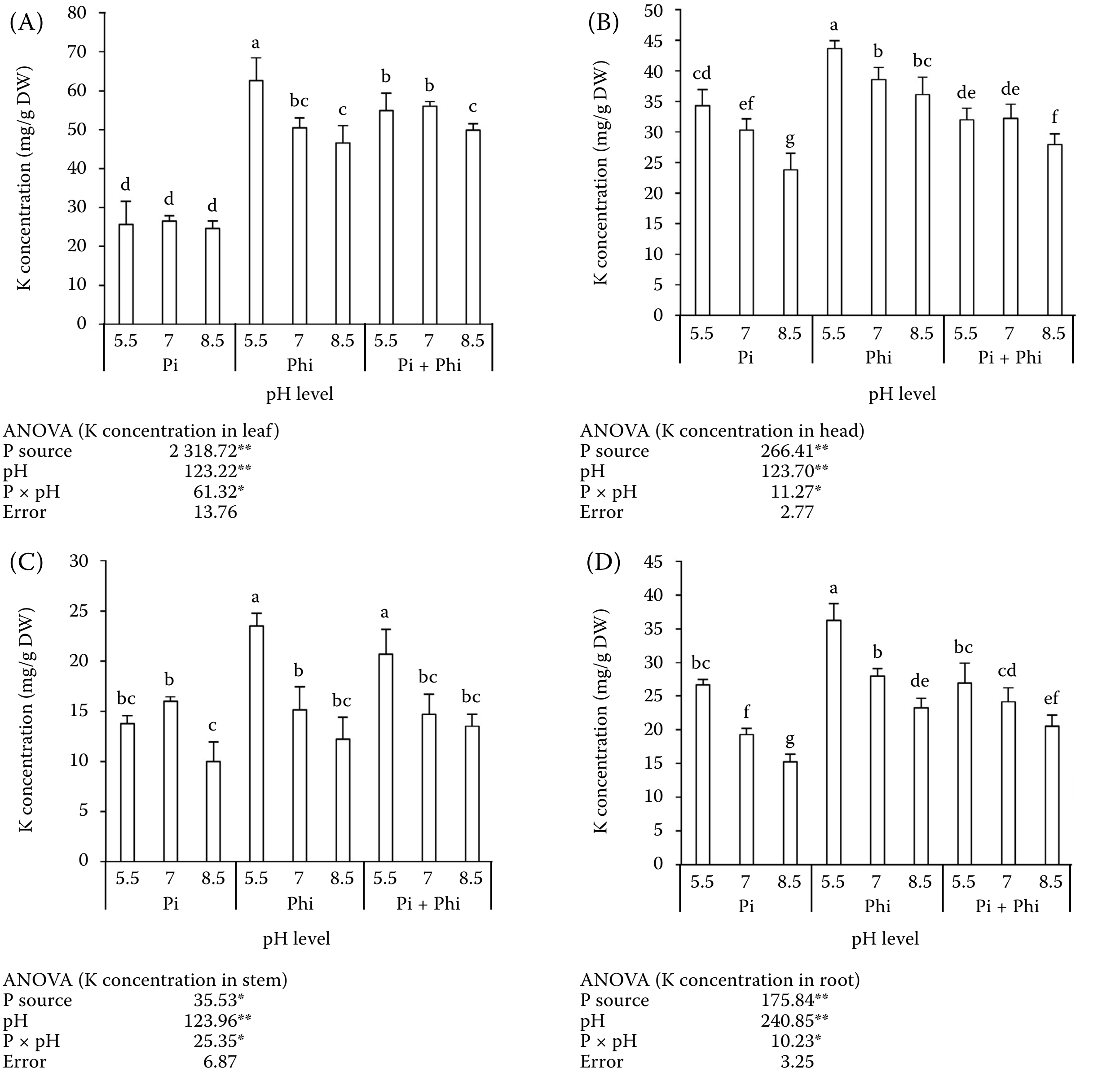

Figure 5. The effect of Phi and Pi in pH levels on the concentrations of K in the (A) leaf, (B) head, (C) stem and (D) root of broccoli plant

P – phosphorus; Pi – phosphate; Phi – phosphite; K – potassium; DW – dry weight; *,**significance at 0.05, 0.01 probability level, respectively; a–g different lowercase letters represent significance

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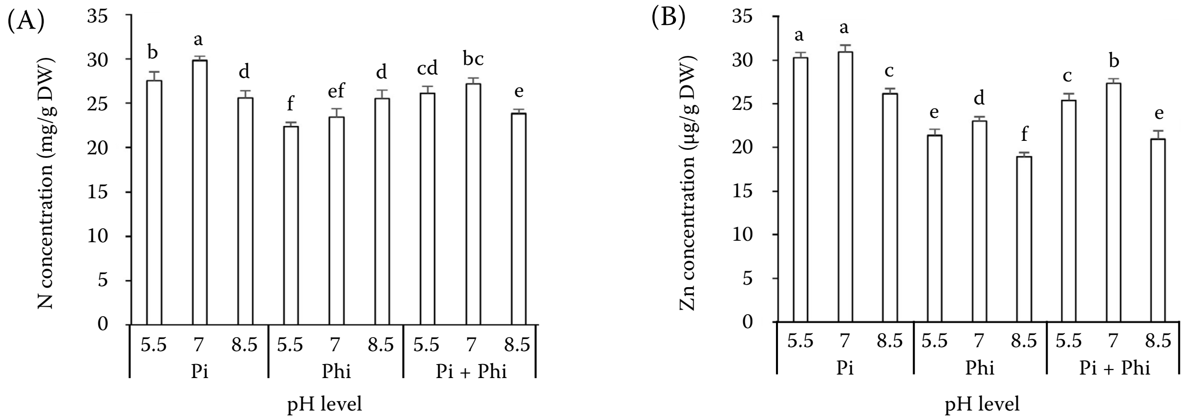
Results showed that there were significant differences in N content between the P source and pH levels (Figure 6A). In general, higher N concentration was observed in treatments with pH 5.5 and 7 Pi. The use of Phi decreased the absorption of Zn in plant leaves. Zn at pH = 5.5 had a 16.45 % and 29.44 % reduction in the Pi + Phi and Phi treatments compared to the Pi treatment (Figure 6B).

The proline and electrolyte leakage rates were significantly increased by applying Phi (Figure 7). The highest percentage of electrolyte leakage (31.33%) was observed in Phi at pH = 8.5 treatment, which shows a 100% increase in electrolyte leakage compared to the lowest (15.67%) treatment (Pi at pH = 5.5). The amount of proline in the presence of Phi as a source of P increased drastically compared to Pi. The Phi at pH = 5.5 and 8.5 increased the proline content by 0.82 mg/g DW compared to Pi in pH = 5.5 and 7.0 treatment, indicating a more than 4-fold increase in proline (Figure 7B). For both proline and electrolyte leakage parameters, the Pi + Phi treatment showed intermediate results between the Phi and Pi treatments.



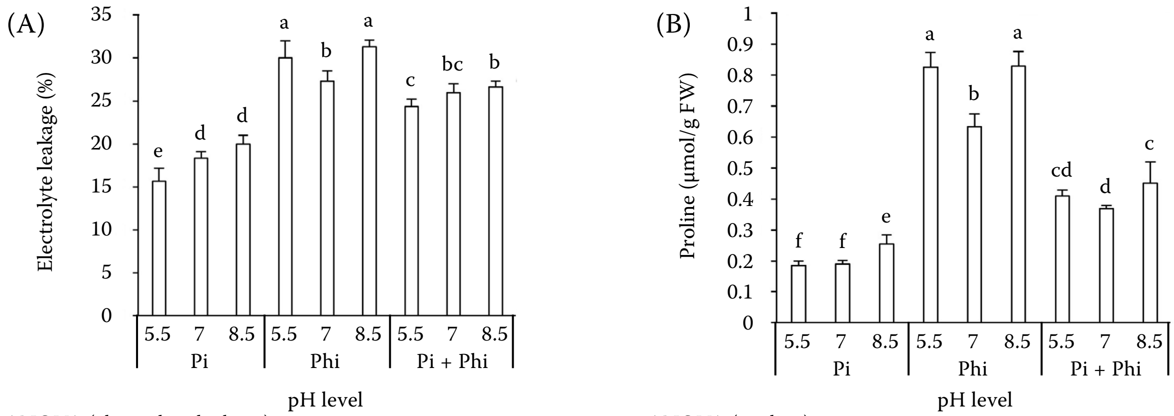
ANOVA (N concentration in leaf)

P source	33.67**
pH	8.42**
P × pH	10.96**
Error	0.44

ANOVA (Zn concentration in leaf)

P source	145.65**
pH	61.59**
P × pH	1.47**
Error	0.32

Figure 6. The effect of Phi and Pi in pH levels on the concentrations of (A) N and (B) Zn in the leaf of the broccoli plant
P – phosphorus; Pi – phosphate; Phi – phosphite; N –nitrogen; Zn – zinc; DW – dry weight; *,**significance at 0.05, 0.01 probability level, respectively; a–f different lowercase letters represent significance



ANOVA (electrolyte leakage)

P source	311.11**
pH	8.42**
P × pH	6.64**
Error	1.22

ANOVA (proline)

P source	0.70**
pH	0.03**
P × pH	0.02**
Error	0.001

Figure 7. The effect of Phi and Pi on pH levels on (A) percentage of electrolyte leakage and (B) concentration of proline in the broccoli plant
P – phosphorus; Pi – phosphate; Phi – phosphite; FW– fresh weight; *,**significance at 0.05, 0.01 probability level, respectively; a–f different lowercase letters represent significance

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DISCUSSION

Broccoli grown using two P sources (Pi and Phi) as fertilisers showed different effects on head formation and P uptake. More recently, other research has also found that Phi has a systemic effect in plant tissues, exhibiting high chemical stability but great mobility throughout the plant. (Gómez-Merino, Trejo-Téllez 2015; Sutradhar et al. 2019; Vinas et al. 2020).

Since the parameter of (Fv/Fm) is a measure of the efficiency of chloroplast organelles or chlorophyll molecules (Ibaraki, Murakami 2006), in experiments, the limit for plant tissue in the absence of physiological or pathological stress is 0.8, with values lower than 0.8 indicating tissue stress (Rostami et al. 2017). Plants treated with phosphite fertilisers showed chlorophyll fluorescence levels below 0.8 and also a lower chlorophyll index. Also, symptoms of toxicity appeared in the petiole area (Figures 8B, C).

In our experiments, the Phi treatment increased electrolyte leakage and proline content in fully developed leaves compared to the other treatments. This indicates that Phi induced stress conditions in the plants, upsetting the balance of soluble nutrients. Batista et al. (2023) reported that foliar spray of potassium phosphite reduced electrolyte leakage in soybean plants under water deficit stress combined with high irradiance under farm conditions. In addition, proline content was increased, suggesting osmotic regulation in response to stress. It appears that Phi induces a stress condition during the head-producing phase, which causes the plant to respond to this situation, and, as a result, the energy needed by the plant is used instead of producing heads to adapt to this status.

Many studies have reported that Pi-deficient plants are more sensitive to Phi application than plants with some supply of Pi and that the addition of Pi can overcome the negative effects of Phi (Singh et al. 2003; Lee et al. 2005). According to these results,

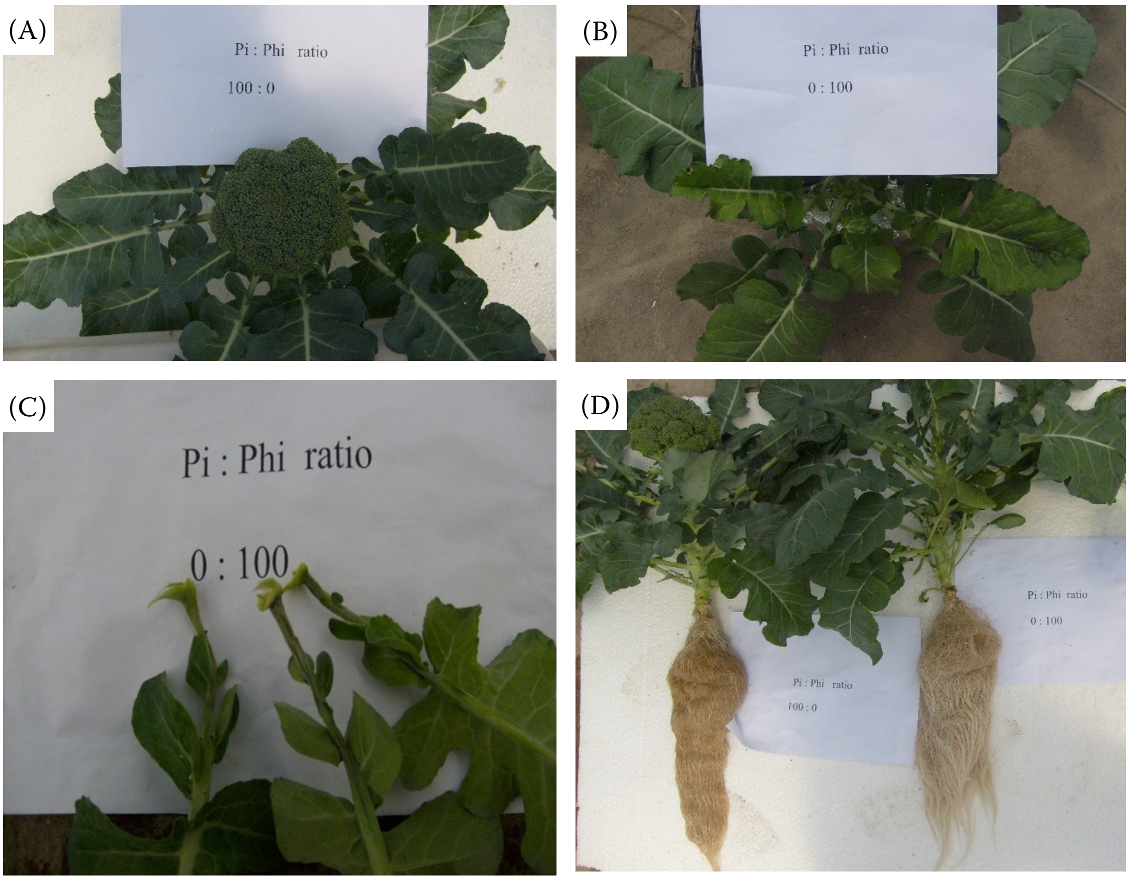

Figure 8. Effect of Phi and Pi ratio on head formation in broccoli: (A) Pi : Phi 100 : 0, (B) Pi: Phi 0 : 100, (C) symptoms of Phi toxicity in petiole and (D) whole plant growth in Phi and Pi after 10 weeks

Pi – phosphate; Phi – phosphite

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in our experiments, the simultaneous use of Pi + Phi caused better results than using Phi alone. Ratjen and Gerendás (2009) showed that an increase in Phi concentration reduced the dry matter yield of zucchini (*Cucurbita pepo*) regardless of Phi application to roots or leaves. Phi has also been reported as a result of both root and foliar applications (lettuce, tomatoes, and bananas) by Bertsch et al. (2009), and their results suggested that in the root absorption, Phi is non-usable for the plant to fulfil its P needs, and tends to cause injury. Most studies designed for root absorption have shown that Phi tends to cause damage because plants do not absorb the Pi they need. The findings have been reported on P accumulation in the cells of sink tissues using Phi (Nartvaranant et al. 2004; Jost et al. 2015). Presumably, because of the short growing season in broccoli and the lack of enough time for the assimilation of Phi through Phi application, excessive P accumulates in the sink tissue and quickly affects the formation of heads, which could be a sign of toxicity. Plants grown under P-limiting conditions are very sensitive to Phi and show toxic symptoms such as leaf chlorosis and reduction in growth (Ratjen, Gerendás 2009; Thao et al. 2009; Thao, Yamakawa 2010). In addition, other negative effects reported with the use of Phi include inhibition of primary root growth, yellowing of young leaves, decreased root: shoot ratio, and accumulation of anthocyanins in mature leaves (Gómez-Merino, Trejo-Téllez 2015). The role of Phi as a biostimulant has been highlighted in a review study (Thao, Yamakawa 2009; Gómez-Merino, Trejo-Téllez 2015). Considering that some studies have mentioned that plants are unable to metabolise Phi, and it is transported quickly to the sink source by phloem (McDonald et al. 2001b).

The effects of Phi in soil and hydroponics are distinguished by the possibility of oxidation of Phi by soil microorganisms (Havlin, Schlegel 2021) and oxidation to Pi under soil conditions, which may be the reason for some positive reports of Phi under soil use conditions (Fontana et al. 2021; Kehler et al. 2021). Higher Pi concentrations were seen in all the plants after foliar sprays of KH_2PO_3, according to Schroetter et al. (2006). This could be because plant tissues partially oxidised and incorporated Phi to Pi in plants. According to most of the evidence, Phi has negative impacts on plants when they are starving or in a P deficit (Thao, Yamakawa 2009), but it can also have a few beneficial impacts on plants when they have an adequate supply of P (Glinicki et al. 2010; Lovatt 2013).

A clear relationship between P and K concentration of sink source and head formation was observed, suggesting that P and K may indirectly affect broccoli heads. It is most likely that in Phi treatment, the increased concentration of P and K in the leaves and heads may be responsible for reducing plant growth and toxicity effects in the absence of head formation. Our findings are consistent with those of Ávila et al. (2012), who discovered that plants with Phi treatment in oxisol soil showed increased P concentrations in shoot and root tissues. Presumably, the Phi treatment caused an antagonistic effect between P and Zn, and the results for P and Zn in leaves indicate that the absorption of Zn in the plant decreased as the P concentration in the leaves increased. According to Thao et al. (2009), adding Phi to the nutrient solution reduced the amounts of N, K, Ca (calcium), Mg (magnesium), Fe (ferrum), and Zn in the shoots of hydroponic lettuce grown in Pi deprivation.

Generally, Phi application is more effective when it is planned appropriately to plant requirements; Phi application depends on the genotype of the crop plant used, the type of soil and climate conditions in which the plant is grown, and cultural method, as well as the dose, rate, and source of Phi used (Trejo-Téllez, Gómez-Merino 2018; Havlin, Schlegel 2021). In general, it has been confirmed that when Pi is sufficient, the positive effects of Phi on plant metabolism are significantly more effective (Thao et al. 2008; Thao, Yamakawa 2009). Presumably, in the case of Phi, traditional Pi concentrations for plants do not apply; its concentration should be lower than Pi, and Phi cannot be used as the sole source of P but can be used in combination with Pi as a biostimulant in the form of a foliar spray or soil application. The rate of Pi uptake decreases as the pH of the external solution increases, which is explained by a reduction in the concentration of $H_2PO_4^-$, which is the substrate of the proton-coupled Pi symporter in the plasma membrane, in the pH range of 5.6–8.5; conversely, a decrease in pH can increase the activity of proton-coupled solute transporters and enhance anion uptake (da Silva Cerozi, Fitzsimmons 2016). Observations on the effect of pH on the absorption of elements, especially P, which is strongly influenced by environmental pH, Phi, like Pi, has its highest absorption by plants at pH 5.5 to 7, and furthermore, absorption decreases significantly when pH is higher than 7.

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CONCLUSION

The present study showed that Phi strongly inhibits the yield and the formation of immature broccoli heads as well as the formation of button-like flower heads. Compared to the usual Pi treatment, the Phi treatment leads to a higher concentration of P and K in the heads. As was discussed the Phi is strongly taken up by the plant cells and accumulates more systematically in the sink tissues. High P concentrations were found in all the plants when Phi was applied to the roots in the floating system.

The increased P uptake in the Phi-treated plants led to increased P concentration in the leaves, growth reduction, and phytotoxicity. We conclude that Phi does not provide P nutrition to broccoli plants via root application. We suspect that Phi is not metabolised by the plant cells and may not be used as a major source of P supply.

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