

Accumulation and distribution of micronutrients in banana cv. Williams (*Musa* AAA Simmonds) with different doses of nitrogen

Acumulación y distribución de micronutrientes en banano cv. Williams (*Musa* AAA Simmonds) bajo diferentes dosis de nitrógeno



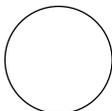
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Banana fruits cv. Williams grown for export in the CENIBANANO experimental field (Carepa, Colombia).

Photo: J. Torres-Bazurto

ABSTRACT

This research evaluated the effect of nitrogen fertilization (0, 161, 321.8, or 483 kg ha⁻¹) on the accumulation of Fe, Cu, Zn, Mn, and B in banana plants cv. Williams in two production cycles in Uraba, Colombia. The micronutrient accumulation models for the plants were obtained with a multivariate approach for differences between cycles, using a randomized complete block design with repeated measurements over time. The N doses with greater accumulation of Cu, Fe, Mn, Zn and B in the plants with fruits were 321.8 and 483 kg ha⁻¹. The average sequence of micronutrient extraction by whole plants was Fe>Mn≥Zn>B>Cu in the first cycle and Fe>Cu>Mn≥Zn>B in the second cycle. The micronutrient accumulation was organ-specific and varied depending on the stage of development. Fe was the major micronutrient extracted by the corm, pseudostem, and bunch. The leaves had the greatest accumulation of Mn. The higher fertilizer doses generated a major response in terms of micronutrient extraction by the banana plants.



Additional keywords: fertilization; nutrient partitioning; mineral nutrition; Musaceae.

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RESUMEN

La presente investigación evaluó el efecto de diferentes dosis de fertilización nitrogenada (0, 161, 321,8 o 483 kg ha⁻¹) sobre la acumulación de Fe, Cu, Zn, Mn y B en plantas de banano cv. Williams durante dos ciclos productivos en el Urabá, Colombia. Los modelos de acumulación de micronutrientes en plantas se obtuvieron mediante un enfoque multivariado de las diferencias entre ciclos, empleando un diseño de bloques completos al azar con medidas repetidas en el tiempo. Las dosis de N que mejor comportamiento mostraron para la acumulación de Cu, Fe, Mn, Zn y B en las plantas, incluidos los frutos, fueron 321,8 y 483 kg ha⁻¹. La extracción de micronutrientes por plantas completas tuvo el siguiente orden Fe>Mn≥Zn>B>Cu para el primer ciclo y Fe>Cu>Mn≥Zn>B para el segundo ciclo. La acumulación de micronutrientes fue específica a cada órgano y varió según la etapa de desarrollo, siendo el Fe el principal micronutriente extraído por el cormo, el pseudotallo y el racimo, mientras que las hojas presentaron la mayor acumulación de Mn. Las mayores dosis de fertilizantes resultaron en la mayor respuesta en la extracción de micronutrientes por las plantas de banano.

Palabras clave adicionales: fertilización; partición de nutrientes; nutrición mineral; Musaceae.

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INTRODUCTION

Musa sp. crops are vital for the diet of approximately 400 million people worldwide (Perea, 2003). In Colombia, the income of about 150 thousand families depends on the production of banana for export (Red Agrícola, 2020), and Uraba is the principal producing region of the country. About 85% of the current Uraba economy is based on the production of export-type bananas, mainly the Cavendish subgroup *Musa* AAA Simmonds (Red Agrícola, 2020). However, various socio-economic and technical problems affect the cultivation of bananas in the area (Gutiérrez *et al.*, 2017; AUGURA, 2020).

One of the serious technical challenges that affect banana productivity is the adjustment of fertilizer doses (Alcaraz and Jiménez, 2018; Torres *et al.*, 2019). In Uraba, nitrogen is the key element in fertilizer doses based on crop requirements per stage of development (Torres *et al.*, 2019; AUGURA, 2020). Nitrogen is assimilated by banana plants mainly in the form of nitrate or ammonium (Sánchez and Mira, 2013; Keshavan *et al.*, 2014). The requirements of banana crops are considered high, exceeding 200 kg ha⁻¹ year⁻¹ of N (Sánchez and Mira, 2013). According to Marschner (2012), the optimal content of N for adequate plant growth is between 2 and 3% of dry weight (DW). Robinson and Galán (2012) reported an optimal N content in banana leaves ranging from 2.5 to 3% DW.

Nutrient extraction curves in plants are useful for defining optimal doses of macro- and micronutrients

(Medina, 2010; Jeyabaskaran *et al.*, 2018). The extraction of mineral nutrients should be studied in banana plants taking into account the sink-source relationships, such as between the mother plant and suckers (Guimarães *et al.*, 2020; Turner *et al.*, 2020). Some regional studies on banana cv. Williams have defined N extraction levels at harvest, which could be used for determining fertilizer doses (Soto, 2001). In Colombia, one of the first studies on nutrient extraction by banana plants was done by Martínez (2006) for cvs. Gran Enano and Valery in Uraba, which posed the question: what is the effect of N fertilizer rates on the accumulation and distribution of micronutrients in banana plants?

This question should be answered by taking into account the importance of micronutrients in banana growth (Torres, 2016). Micronutrient applications affect the growth of leaves (Souza *et al.*, 2016), roots (Liu *et al.*, 2019), and pseudostems (Kumar *et al.*, 2020) and banana production (Robinson and Galán, 2012). The contents of Fe, Mn, Zn, Cu, and B in plant organs estimate the plant nutrient status with the DRIS method (Villaseñor *et al.*, 2020). The concentrations of Mn and B in banana leaves may correlate with chlorophyll contents (Arantes *et al.*, 2016), and micronutrient allocation to fruits is required for bunch development (Soto, 2001). However, excess Fe, Mn, and Cl in coastal tropical soils could negatively affect yields (Osorio, 2014; Villaseñor *et al.*, 2020).

Few studies have addressed micronutrient accumulation in banana plants as affected by macronutrient fertilizers. In Brazil, Borges *et al.* (2006) compared foliar contents of the microelements B, Cl, Cu, Fe, Mn, and Zn in 24 genotypes in two production cycles that had an annual addition of 100 kg N ha⁻¹, 40 kg P₂O₅ ha⁻¹, and 300 kg K₂O ha⁻¹. The genotypes differed in terms of micronutrient extraction by the leaves. In addition, each genotype had an accumulation that varied between the cycles, with lower levels of Cl, B, Fe, and Zn in the second cycle; the leaf contents substantially varied for Mn (43-574 mg kg⁻¹) and Fe 56-212 mg kg⁻¹ (Borges *et al.*, 2006). Robinson and Galán (2012) stated that genotype plays a key role in the absorption and foliar accumulation of micronutrients, and that these processes are influenced by edaphoclimatic conditions and fertilizer practices. The aim of this research was to evaluate the accumulation and distribution of the micronutrients Fe, Mn, Zn, Cu, and B in banana cv. Williams (*Musa* AAA Simmonds) as affected by different doses of N fertilizers in the Uraba region of Colombia.

MATERIALS AND METHODS

Experiment location and climatic conditions

This research was conducted in 2011-2012 in the Uraba region of the Antioquia department, Colombia, using the experimental field of AUGURA (Carepa, Antioquia), lots 3 and 4, 7°46'46" N and 76°40'20" W and 20 m a.s.l. The soils were fine Fluventic Eutrudepts, clay loam, over clay Fluvaquentic Eutrudept, and fine loam Vertic Endoaquept, according to USDA classifications (Soil Survey Staff, 2014). In the soils, the pH ranged from 5.2 to 6.2, the average N contents was 0.36%, the contents of Ca, Mg, and K were 15.8, 5.9, and 0.44 cmol_c kg⁻¹, respectively, and the P content was 8.25 mg kg⁻¹. The micronutrient contents (mg kg⁻¹) in the soils were 4.35-6.67 for Cu, 72.2-148.9 for Fe, 13.3-72.6 for Mn, 0.79-1.70 for Zn (DTPA extraction), and 0.22-0.34 for B (Azomethine-H method) (IGAC, 2006).

The following climatic conditions were registered during the experiment: 23.2°C minimum and 32.3°C maximum air temperatures; 26.7°C average air temperature (with 27°C regional average); 87% relative air humidity, which coincided with the annual average; and 5 h d⁻¹ average solar brightness, totaling 1700 h year⁻¹. The annual precipitation was 845 mm during

the first production cycle (August 2011-April 2012) and 2,088 mm during the second production cycle (February 2011-December 2012). The rainfall was low from January to March (1.4-30.4 mm month⁻¹), which corresponded to a "dry season", typical for the region.

Plant material and treatments

Banana plants (*Musa* AAA Simmonds) from the AAA group, Giant Cavendish subgroup, Williams clone in the 6th productive cycle were used. The plants had an average height of 3.5 m. The sowing distance was 2.5 × 2.5 m, with approximately 1,600 plants/ha. The agronomic management for commercial banana plantations in Uraba was used, except for the fertilizer doses.

The fertilization system recommended by Cenibanao for the region was applied taking into account the plant nutrient requirements and the results of the soil analysis (Sánchez and Mira, 2013). The five treatments were: absolute control (no fertilizer applied), 0, 161, 321.8, and 483 kg ha⁻¹ of N. Additionally, all plants, except the absolute control, received the following amounts of fertilizers per ha: 87.1 kg P₂O₅, 678.8 kg K₂O, 50.5 kg CaO, 117.5 kg Mg, 64.2 kg S; 1.4 kg B, and 9.3 kg Zn. The sources of the mineral fertilizers were urea, K-Mag® (22% K₂O, 18% MgO, and 22% S), Fertiboro (10% B), Solufos (30% P₂O₅, 36% Ca, 5% S, and 8% Si), ZnO, K₂SO₄, and KCl. The fertilizer doses were fractionated within each production cycle, for a total of 17 fertilizer applications per year, with approximately 3 weeks between applications. The fertilizer applications started with the flowering of the crop, when fertilization of the ratoon traditionally begins in the region.

Experiment establishment

In the field, the soil-spatial variability was considered, establishing four in-space replicates (blocks), with the treatments randomized within the blocks. The blocks were distributed based on the soil taxonomic units, and each block had a complete set of treatments. Each treatment matched the unit called "botalon", which was 1563 m² area and had 250 banana plants. Fifteen plants were selected within each "botalon" by height (between 1.0 and 1.5 m), forming the experiment unit. The evaluations were performed after a certain number of weeks that corresponded to 50% of the plants reaching the following phases of

growth: vegetative growth (weeks 17-18), flower differentiation (weeks 22-23), flowering (weeks 39-40), bunch filling (weeks 43-44), and harvest (weeks 50-51). The fruit filling and harvest in the first production cycle overlapped with the vegetative growth and flower differentiation of the second cycle.

The sampling was done at least two weeks after the fertilizer applications. The plants were dissected into organs and dried in oven at 65°C for 48 h. The micronutrient contents were quantified in the Soil and water laboratory of the Facultad de Ciencias Agrarias, Universidad Nacional de Colombia (Bogota), using the standardized methods of analysis (IGAC, 2006).

Statistical analysis

A completely randomized block design was used with five fertilizer treatments and four replicates to assess the micronutrient accumulation in the plants. The multivariate analysis was used to evaluate the variance of the repeated measures design, where the treatments and replicates were two factors between-the-subjects, the production cycle was an intra-subject factor, and the blocks were associated with the soil spatial variability in the field, which helped adjust the model.

The micronutrient accumulations per organ and phenological stage were approached with a principal component analysis and multivariate analysis (Johnson and Wichern, 2007). To evaluate only the changes between the fertilizer treatments and eliminate the cycle effects, the differences between the cycles were considered for each variable. The principal component analysis determined the number of components for use in the multivariate models defined for the treatments to establish the variables with the best fit per model. The accumulation of Fe, Mn, Zn, Cu, and B in the plant organs was assessed to adjust three models. The first model described the phenological stages of vegetative growth, flower differentiation, and flowering. The other two models described the stages of fruit filling and harvest. All three models were established independently using the same design, factors and blocks to adjust the multivariate models. In addition, since there was no interaction between the factors, the statistical analysis included tests of variances between the control and other treatments to elucidate the effects of the treatments.

A descriptive interpretation was performed using the averages per treatment and phenological stage

for each variable; these data are presented in the tables. SAS 9.3 (SAS Institute, Inc.), R, and Statgraphics Centurio XV (Statpoint Technologies, Inc.) were used for the data analysis.

RESULTS AND DISCUSSION

Micronutrient accumulation in plants

The multivariate analysis showed a significant interaction between the productive cycle and fertilizer treatment ($P < 0.0001$); in addition, statistical differences were seen between the treatments ($P < 0.0001$). The micronutrients, except Cu, tended to increase in accumulation in whole plants at harvest (Tab. 1) as the doses of N fertilizers increased.

Table 1. Effect of N doses on micronutrient accumulation in banana cv. Williams at harvest in two production cycles.

N dose (kg ha ⁻¹)	Micronutrient accumulation (kg ha ⁻¹)				
	First cycle				
	Cu	Fe	Mn	Zn	B
Absolute control	1.0	8.1	2.8	3.4	2.0
0	0.2	5.7	2.4	1.6	1.5
161	0.2	6.5	2.7	2.1	1.6
321.8	0.2	7.9	2.3	2.4	2.2
483	0.2	7.4	3.2	2.3	1.7
Second cycle					
Absolute control	2.3	4.4	2.7	2.6	0.6
0	4.3	4.8	2.0	2.1	0.5
161	3.4	4.4	3.1	3.4	0.8
321.8	3.3	5.3	3.0	3.3	0.9
483	3.6	5.3	3.2	3.0	0.6

This effect was not clear for the first cycle (Tab. 1), where the absolute control surpassed the other treatments in the accumulation of Fe, Zn, Cu, and B. This response could be associated with micronutrient retranslocation from the mother plant to the ratoon. The response for Fe and Cu could also be due to their high contents in Uraba soils. The higher N doses (321.8 and 483 kg ha⁻¹) provided the highest accumulation of micronutrients, except for Cu, as compared to the control (0 kg ha⁻¹ N). Copper had the highest accumulation in the absolute control, possibly because of the high initial content of Cu in the

soils (ICA, 1992) and the absence of fertilizer applications in this treatment. According to Soto (2001), applications of phosphorus fertilizers can reduce Cu availability for plants because of the formation of Cu phosphates with low solubility in the soil, which may have happened in the other treatments, as compared to the absolute control.

For the second cycle, the accumulation of micronutrients increased as the N doses increased (Tab. 1), with a high accumulation of Zn and B at 321.8 kg N ha⁻¹ and the highest Fe and Mn contents seen with 483 kg ha⁻¹ of N. Regardless of the micronutrient accumulation in the absolute control in the first cycle, the doses 321.8 and 483 kg ha⁻¹ N generated the highest accumulation for most micronutrients in the banana plants (Tab. 1). A similar result was found by Nyombi *et al.* (2010), who applied 400 kg ha⁻¹ N, 50 kg ha⁻¹ P, and 600 kg ha⁻¹ K and observed increased concentrations of mineral nutrients in banana plants.

The averaged sequence of micronutrient extraction by plants differed between the production cycles: Fe>Mn≥Zn>B>Cu in the first cycle and Fe>Cu>Mn≥Zn>B in the second one. The Cu accumulation changed substantially between the cycles. According to Havlin *et al.* (2013), at low water contents in the soil resulting from low precipitation, the concentration of Cu in the soil solution declines, and its adsorption by soil particles increases, which makes it more difficult for plants to absorb Cu than other nutrients. The conditions of the second cycle (high precipitation rate) were more favorable for Cu availability in the soil. Therefore, the differences in Cu accumulation between the cycles could be attributed to less Cu accumulation in plants under conditions of low precipitation (the first cycle) and to a higher accumulation of Cu in plants under more favorable climatic conditions (the second cycle).

The micronutrient accumulation in the plants in both cycles differed from that found by Rodríguez *et al.* (2004), Selvamani and Manivannan (2009), and Medina (2010). The differences, in most cases, were for the ratio of Mn/Fe accumulation. These elements are typically accumulated in *Musa* plants with high contents, as compared to other micronutrients (Soto, 2001; Jeyabaskaran *et al.*, 2018). There were notable differences from Medina (2010), who reported the accumulation sequence Mn>Fe>Zn>B>Cu. This author worked with banana cv. Williams in a Fluvaquentic Eutrudepts soil, the same soil subgroup as two of the soils used in the present study (Soil Survey Staff, 2014) but with different fertilization practices (without micronutrient applications) and more favorable climatic conditions for plant cultivation, which might explain the differences in the micronutrient accumulations.

Table 2 compares the micronutrient accumulation in cv. Williams with data for banana Robusta (Walmsley and Twiford, 1976) and Cavendish (Lahav and Turner, 1992). Although these three cultivars belong to the same subgroup, Cavendish, the plants differed in the contents of micronutrients. Walmsley and Twiford (1976) reported a lower accumulation of micronutrients than Lahav and Turner (1992) and the present research, with the exception of Mn. The lowest Mn content reported by Walmsley and Twiford (1976) fit the typical interval for cv. Williams, while the highest Mn content was greater than the accumulation in cv. Williams (Tab. 2). These data indicate that micronutrient accumulation in banana plants could vary depending on the cultivar, edaphoclimatic conditions, and agronomic practices, such as planting density and fertilization.

The highest doses of N fertilizers generated the highest production (Torres, 2016) and the highest levels of

Table 2. Micronutrient accumulation in banana cv. Williams for two production cycles in Uraba vs. micronutrient accumulation in bananas Robusta and Cavendish.

Micronutrient	cv. Williams	Plants Robusta (Walmsley and Twiford, 1976)	Plants Cavendish (Lahav and Turner, 1992)
	kg ha ⁻¹ year ¹		
Fe	2.70-49.0	1.0-1.1	5.9
Mn	1.20-1.99	1.3-8.3	12.5
Cu	0.13-2.70	0.053-0.073	0.37
Zn	1.0-2.12	0.16-0.19	4.7
B	0.34-1.37	0.17-0.34	1.27

micronutrient accumulation (Tab. 1). The treatments with 321.8 and 483 kg ha⁻¹ N had better results. Soto (2001) and Robinson and Galán (2012) stated that, when increasing N doses, the absorption and accumulation of other mineral nutrients also increased, with consequent increases in production.

Nutrient accumulation by organ and phenological stage

Tables 3 illustrate the levels of micronutrient extraction by plant organ. The micronutrient contents in the roots were not included because these were negligible as compared to the other organs. Furthermore, the micronutrient concentration in roots might not always reflect plant requirements since metal micronutrients tend to precipitate on root surfaces and contribute to the formation of Fe plaque, especially

when affected by changing redox regimes in tropical soils (Pi *et al.*, 2010; Osorio, 2014).

Corm

Only the model for the harvest stage revealed the presence of significant differences ($P < 0.0082$) between the fertilizer treatments for B accumulation in the corm. No statistically significant effect from the N doses was found on the accumulation of other micronutrients in the corm during plant development. The descriptive analysis by development stage showed that, while the plants grew (Tab. 3), the accumulation of micronutrients in the corm varied. The doses of 321.8 and 483 kg ha⁻¹ N resulted in the highest accumulation of most micronutrients at each phenological stage, with the exception of Cu and Mn, which had a reduced corm content with the dose of 321.8 kg N ha⁻¹ at flowering (Tab. 3).

Table 3. Effect of N doses on average content of micronutrients in corm of banana cv. Williams.

Stage of development	N dose (kg ha ⁻¹)	Cu	Fe	Mn	Zn	B
		mg kg ⁻¹ corm dry weight				
Vegetative	Absolute control	128.9	117.4	98.6	88.2	21.6
	0	102.5	174.4	78.3	71.1	31.8
	161	139.8	132.5	102.0	104.9	39.4
	321.8	93.5	126.2	78.8	89.2	31.9
	483	156.1	189.5	104.0	89.1	24.0
Flower differentiation	Absolute control	144.7	133.2	115.7	128.2	36.0
	0	132.7	167.1	62.2	81.8	25.1
	161	164.6	123.9	63.0	72.9	32.1
	321.8	180.0	128.3	98.2	94.9	21.8
	483	147.9	150.8	86.7	124.7	22.6
Flowering	Absolute control	125.7	251.9	43.4	63.0	35.2
	0	65.0	187.0	45.4	52.7	27.8
	161	79.5	261.8	52.1	58.2	30.7
	321.8	67.0	174.3	41.0	64.5	33.1
	483	155.1	120.5	68.8	74.0	45.8
Fruit filling	Absolute control	141.7	101.2	86.9	110.8	14.6
	0	95.8	94.4	51.2	59.2	20.5
	161	86.0	99.9	44.5	67.0	25.5
	321.8	89.8	167.7	51.4	66.4	19.4
	483	128.2	117.0	66.3	71.3	11.5
Harvest	Absolute control	94.7	350.6	103.0	163.4	24.8
	0	206.2	134.8	83.4	74.7	24.3
	161	192.1	213.0	66.4	71.5	34.7
	321.8	98.5	165.9	92.1	88.4	67.8
	483	143.7	195.8	105.7	99.6	38.5

Yang *et al.* (2013) used fertilizer rates of 385 kg N ha⁻¹ and found the lowest accumulation of nutrients in the corm at harvest. However, in the present experiment, the control, 0 kg ha⁻¹ N, had a higher accumulation of some nutrients in the corm in all stages of development (Tab. 3). This result could be attributed to nutrient recycling between the mother plant and the ratoon. Additionally, Fe and B might see less retranslocation from the corm, which is one of the main reserve organs (Galvis *et al.*, 2013), since these elements has low mobility in the phloem (Marschner, 2012).

The micronutrient accumulation in the corm followed the order Fe>Cu>Zn>Mn>B, which differed from that found by Walmsley and Twyford (1976) (Mn>Fe>Zn>B>Cu) and Martínez (2006) (Mn>Fe>B>Cu>Zn). The cv. Williams corm contained 94.4-350 mg Fe kg⁻¹ and 52.7-163.4 mg Zn kg⁻¹

(Tab. 3), while Martínez (2006) found wider ranges of 73-880 mg Fe kg⁻¹ and 13-182 mg Zn kg⁻¹. Walmsley and Twyford (1976) reported an accumulation of Fe (85-153 mg kg⁻¹) comparable to the present research, while the lowest level of Zn (19 mg kg⁻¹) (Walmsley and Twyford, 1976) was lower than found in cv. Williams. These results could be attributed to differences in soil conditions, crop management, cultivar, and fertilizer doses.

Pseudostem

The models revealed differences between the fertilizer treatments at fruit filling ($P<0.00002$) for the accumulation of Zn and at harvest ($P<0.0030$) for the accumulation of Cu and B. In general, the contents of Fe, Mn, and Zn in the pseudostem tended to increase during plant development (Tab. 4), similar to that reported by Jeyabaskaran *et al.* (2018). Increasing rates of

Table 4. Effect of N doses on average content of micronutrients in pseudostem of banana cv. Williams.

Stage of development	Nitrogen dose (kg ha ⁻¹)	Cu	Fe	Mn	Zn	B
		mg kg ⁻¹ pseudostem dry weight				
Vegetative	Absolute control	71.3	532.1	165.7	68.8	35.9
	0	80.2	804.5	140.4	85.8	38.8
	161	114.8	418.9	174.9	69.7	48.7
	321.8	69.8	447.4	183.0	70.4	41.7
	483	122.8	445.2	169.7	68.6	44.4
Flower differentiation	Absolute control	133.4	229.6	110.4	71.5	39.2
	0	81.6	152.8	103.2	59.5	35.9
	161	198.1	215.3	109.2	59.1	43.3
	321.8	170.5	240.4	111.5	60.1	27.4
	483	172.4	228.8	132.0	65.2	44.3
Flowering	Absolute control	192.4	435.3	190.5	142.0	59.9
	0	212.8	326.3	252.7	151.6	46.2
	161	282.5	435.3	234.0	136.2	66.0
	321.8	174.7	406.4	244.9	145.1	57.0
	483	326.8	395.3	196.6	147.5	59.0
Fruit filling	Absolute control	20.3	374.3	254.5	180.0	123.1
	0	14.1	319.0	230.5	217.7	148.3
	161	16.1	396.5	232.7	195.7	104.7
	321.8	14.9	806.0	224.8	182.5	117.8
	483	16.7	383.5	248.4	202.8	119.1
Harvest	Absolute control	88.7	665.4	223.8	301.3	123.4
	0	19.8	691.7	230.6	241.4	117.0
	161	15.8	522.9	260.3	293.3	121.1
	321.8	16.6	640.4	202.3	277.7	137.4
	483	18.7	711.6	271.3	295.2	106.6

N, especially the doses 321.8 and 483 kg ha⁻¹ N, had a positive effect on micronutrient accumulation in the pseudostem (Tab. 4). These results were similar to those of Castillo *et al.* (2011), who obtained the highest contents of mineral nutrients in the pseudostem with doses of 375 kg ha⁻¹ N.

The sequence of micronutrient accumulation in the pseudostem changed from Fe>Mn>Cu>Zn>B at flowering to Fe>Zn>Mn>B>Cu at harvest. At both phenological stages, the data differed from those of Walmsley and Twyford (1976), Soto (2001), and Martínez (2006). These variations could be explained by interactions of environmental factors, soil, cultivar, and crop management, especially fertilization (Robinson and Galán, 2012). Additionally, the contents of elements in the pseudostem could vary between the cultivars depending on morphological differences

in the distribution and configuration of vascular elements in the pseudostem (Souza *et al.*, 2016).

Leaves

The nutrient accumulation models revealed significant differences between the fertilizer treatments at fruit filling ($P<0.0061$) and harvest ($P<0.0387$). The nutrients participating in the first model included Cu, Fe, and Zn, and the second model included Cu and B. The micronutrients differed in their accumulation in the leaves, with Cu contents that decreased and Mn contents that increased after flower differentiation (Tab. 5). The descriptive analysis showed that the N applications, especially at 321.8 and 483 kg ha⁻¹, favored a higher accumulation of the majority of the micronutrients in the leaves in all development stages, except flower differentiation (Tab. 5).

Table 5. Effect of N doses on average content of micronutrients in leaves of banana cv. Williams.

Stage of development	Nitrogen dose (kg ha ⁻¹)	Cu	Fe	Mn	Zn	B
		mg kg ⁻¹ leaf dry weight				
Vegetative	Absolute control	93.4	241.6	166.2	56.9	50.9
	0	68.0	242.3	164.0	41.5	44.7
	161	44.9	260.3	315.2	48.3	57.8
	321.8	49.0	222.6	105.0	37.4	41.5
	483	102.3	332.3	217.2	67.7	37.6
Flower differentiation	Absolute control	176.4	293.7	95.2	30.9	16.6
	0	159.7	175.9	71.8	38.3	17.2
	161	117.4	319.7	181.1	57.6	28.3
	321.8	88.8	133.1	61.6	36.8	18.5
	483	70.5	156.1	89.3	31.2	24.5
Flowering	Absolute control	66.6	257.5	100.2	36.9	13.2
	0	79.8	273.7	104.0	59.0	16.5
	161	45.9	227.0	111.6	28.0	12.3
	321.8	147.2	288.2	101.1	54.6	16.0
	483	71.6	293.8	105.3	61.1	19.4
Fruit filling	Absolute control	47.9	202.4	258.7	27.5	20.7
	0	26.6	175.5	266.3	16.1	15.4
	161	40.9	243.1	258.5	33.2	25.2
	321.8	33.1	211.9	309.0	21.2	19.7
	483	22.2	160.4	288.2	22.9	22.1
Harvest	Absolute control	37.7	221.6	379.4	201.8	118.8
	0	29.8	223.6	285.8	67.9	43.7
	161	39.1	226.9	376.5	166.0	85.2
	321.8	38.8	263.8	316.0	145.7	75.1
	483	40.1	285.9	404.9	80.8	45.7

The higher doses of N (321.8 and 483 kg ha⁻¹) mostly contributed to the accumulation of B in the leaves, as compared with 0 kg ha⁻¹ N (Tab. 5). The leaf contents of B and Mn could have highly significant, positive associations with the contents of a, b, and total chlorophyll, as observed in 'Prata' bananas (Arantes *et al.*, 2016). This relationship is important since more than 70% of the leaf N could be a part of the chlorophyll molecules (Marschner, 2012).

The order of micronutrient accumulation in the leaves was Mn>Fe>Cu>Zn>B, differing from the Mn>Fe>B>Zn>Cu reported by Walmsley and Twyford (1976), Soto (2001), and Martínez (2006), which could be attributed to the cultivar and practices. The ranges of micronutrient accumulation in the cv. Williams leaves differed from the literature data. For example, the Mn content was 61.6-404.9 mg kg⁻¹ leaf DW (Tab. 5), while other authors reported 772-1945 (Walmsley and Twyford, 1976) and 867 mg kg⁻¹ leaf DW (Soto, 2001).

Leaves are source organs for fruit filling (Soto, 2001). However, high doses of N could promote excessive leaf growth and favor a greater accumulation of N in leaves, i.e. plants allocate more N in leaves than in fruit formation, resulting in low productivity (Deus *et al.*, 2020). The increased accumulation of Mn and B in the cv. Williams leaves at the higher N doses (Tab. 5) could be due to increased rates of leaf growth; thus, the leaves might have demanded greater amounts of micronutrients for growth when stimulated by high N rates.

Bunch

The model obtained at the harvest stage revealed differences between the fertilizer treatments ($P<0.0007$) for the accumulation of Mn and Cu in the bunches. At both development stages (fruit filling and harvest), the higher doses of N favored micronutrient accumulation in the fruits (Tab. 6) and increased fruit yield (Torres, 2016). Increasing the doses of N fertilizers most highly stimulated Zn accumulation in the bunches, as compared to other micronutrients. While the B contents diminished during fruit formation, the Mn contents decreased when increasing N doses at fruit filling, which increased at harvest (Tab. 6).

The fruits accumulated micronutrients in the sequence Fe>Cu>Zn>Mn>B, differing from the sequences Mn>Fe>B>Zn>Cu (Walmsley and Twyford, 1976), Fe>Zn≈Mn>Cu (Forster *et al.*, 2002), and Mn>Fe>Zn>B>Cu (Yang *et al.*, 2013); additionally, the contents of Cu, Zn and B in the cv. Williams fruits (Tab. 6) exceeded those reported for other cultivars (3-10.2, 4-49.2, and 9-23.6 mg kg⁻¹ bunch for Cu, Zn, and B, respectively) (Walmsley and Twyford, 1976; Yang *et al.*, 2013).

Micronutrient accumulation in fruits is important for fruit formation and quality (Robinson and Galán, 2002; Pareek, 2016). Despite the differences in the micronutrient accumulation in the fruits between the N treatments during the two production cycles, these was no effect on the fruit export quality in terms of postharvest fruit duration or peel splitting (data not shown). In the tropics, banana

Table 6. Effect of N doses on average contents of micronutrients in bunches of banana cv. Williams.

Stage of development	Nitrogen dose (kg ha ⁻¹)	Cu	Fe	Mn	Zn	B
		mg kg ⁻¹ bunch dry weight				
Fruit filling	Absolute control	137.7	331.6	126.1	136.0	102.8
	0	232.3	300.6	139.7	156.1	103.7
	161	153.5	343.8	131.4	163.5	67.4
	321.8	166.7	696.8	129.0	168.8	104.4
	483	185.1	313.8	118.4	151.3	89.7
Harvest	Absolute control	116.0	214.3	92.8	101.2	66.5
	0	261.2	262.2	92.9	92.6	71.2
	161	166.9	324.9	137.8	153.6	64.6
	321.8	183.2	211.9	78.1	96.4	53.5
	483	142.8	267.4	133.4	125.9	69.5

fruits could serve as a source of micronutrients, especially Mn, Fe, and Zn, for human diets (Pareek, 2016; Ashokkumar *et al.*, 2018). According to our results, increasing the rates of N applications favored to the accumulation of the majority of the micronutrients in the cv. Williams fruits. More research should be conducted for more than two cycles to assess micronutrient accumulation and distribution in banana plants with ratoons from three or more generations.

CONCLUSIONS

Increasing the doses of N fertilizer in the two production cycles increased the levels of micronutrient accumulation in the cv. Williams banana plants, including the fruits. The better rates of N fertilizers for the highest accumulation of Cu, Fe, Mn, Zn, and B in the plants were 321.8 and 483 kg ha⁻¹ N.

The averaged sequences of micronutrient extraction by whole plants differed between the production cycles: Fe>Mn≥Zn>B>Cu in the first cycle and Fe>Cu>Mn≥Zn>B in the second one. The sequence of micronutrient accumulation in the organs was organ-specific and varied in each organ, depending on the stage of development.

Conflict of interests: The manuscript was prepared and reviewed with the participation of the authors, who declare that there exists no conflict of interest that puts at risk the validity of the presented results.

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