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Responses of Agronomically Important Tropical Crops to the Application of Brassinosteroid

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Abstract

Brassinosteroids (Br) have been shown to favor the growth and reproduction of crops under adverse environmental conditions, which negatively affect their growth and production. In order to solve some of the problems in the field with various perennial crops, the application of a homobrassinolide (HBr) (CIDEF-4) has been investigated under *in vitro* and *ex vitro* conditions to evaluate growth at different concentrations in *Musa* spp. L. and *Saccharum officinarum* L. and in the field with foliar applications in *Theobroma cacao* L., *Mangifera indica* L. and *Coffea arabica* L. to evaluate yield and quality of fruits. Morphological and physiological yield components were recorded. The results indicate in the *in vitro* evaluations, increased regrowth height and *ex vitro* differences in growth are improved by increasing the number of applications. In cocoa and coffee plants, flowering and yield are influenced. The high concentrations applied did not necessarily increase the crop yield or the combination with potassium nitrate. In mango, the quality of the fruits was better when applying the HBr alone or in combination with nitrate in fruit firmness, and total soluble solids improved.

Keywords: brassinosteroids, growth, quality fruits, yield, biomass

1. Introduction

The brassinosteroides (Br) are the most active growth-promoting extracts isolated from *Brassica napus* pollen [1]. These are steroidal phytohormones essential for the growth, development and adaptation of plants to biotic and abiotic stress [2–5]. Regulatory activity seems to be due to the influence of these hormones on metabolic processes related to photosynthesis, nucleic acid and protein biosynthesis [6, 7]. It is transported from sites of synthesis to sites of distant action and operates at very low concentrations [8–10]. These compounds have a wide range of activity in protecting plant metabolism when these are under stress conditions and consequently increase their yield [11], through de increase la activity within the antioxidant route [12, 13], promote membrane polarization and influence tolerance to different stressful environmental conditions [14–17]. These are low and high temperatures [18, 19] and tolerance to water stress [20] among others. At the cellular level, they are expressed in elongation, cell differentiation and genetic modulation [7, 21].

In agricultural activities, it has been shown that various abiotic stressors for crops can be mitigated through the application of hormones, such as brassinosteroids [6]. This technology is relevant when considering that agricultural activities each year face environmental factors, such as drought and erratic rainfall [22–24], which has been the main cause of food insecurity that induces decreased yields [25]. Nowadays some Brassinosteroids homologs (HBr) which are similar to natural brassinosteroids, have been shown to be useful in agriculture [26]. They have been used as a strategy to increase growth and yield in various crops in the field and greenhouse through their exogenous application, and these have shown enhancement of growth and reproduction under adverse conditions [11, 16, 27–29]. In addition, HBRs play a fundamental role in flower and fruit development, leaf senescence and abscission [30, 31]. They are also proposed in the reproduction of plants *in vitro* to be added to the culture medium in different explants and organs [32] as substitutes for auxins and cytokinins [33, 34]. Under these conditions, they can stimulate aerial and radical plant growth [35, 36], as noted in rooting, number of leaves and plant height in various orchids when added HBr (Cidef-4) to the medium Yasuda [37] and in *Cedrela odorata* L. the number of shoots and leaves is increased by adding the same (HBr) to the Murashige and Skoog medium (MS) [38, 39].

In the greenhouse, biomass, stevioside, rebaudioside and steviol content are increased in the *Stevia* crop compared to the control [40]. In tomato, it is associated with the content of lycopene and carbohydrates [41] and in transgenic tomatoes, early flowering and a slight increase in fruit yield and number of fruits per plant were obtained [20, 42], fruits and oilseeds [43–45] in addition to enhancing crop quality [46].

2. Response of agricultural crops to the application of brassinosteroids through the increased lab activity

2.1 *In vitro* reproduction of *Musa* spp. “big dwarf banana” with brassinosteroid and other growth regulators

Banana (*Musa* spp.) is widely consumed by the world population and in tropical regions. It is a daily food in developing countries and its *in vitro* reproduction often has a low multiplication coefficient, high percentage of phenolization in explants and low survival [47, 48]. In other cases, abnormal shoots are reported in plants with some growth regulators, which are traditionally incorporated into the growing medium [33].

In this regard, non-traditional growth regulators, such as HBr, can favor multiplication processes [18, 49] and improve normal plant growth and development. However, by subjecting the explant to an artificial environment for its reproduction. It is exposed to stressful conditions, which can be attenuated by HBr and improve its reproduction, as happened with the FHIA-21 banana, which favored the formation of roots *in vitro* [50].

The *in vitro* response of *Musa* spp. cv great dwarf shows morphological changes when establishing the apical meristem in MS medium [38] and three growth regulators, 6-bencilaminopurine (BAP), indoleacetic acid (IAA) and homobrasinolide (HBr-Cidef-4; Natura del desierto SA de CV, México) in two concentrations 2 and 4 mg^{L-1}, during 80 days of evaluation with changes of medium every 20 days.

The HBr, in its two concentrations, presents a response similar in the number of leaves and roots to those induced with BAP and IAA and statistically different from the control ($P \leq 0.05$) (Table 1).

The lowest average shoot height was with BAP at the lowest concentration. On the other hand, by increasing the concentration, the height was almost doubled. An

Treatment (mgL ⁻¹)	Height (cm.regrowth ⁻¹)	Number of leaves.regrowth ⁻¹	Number of roots
BAP 2.0	3.59 e	11.25 a	5.12 b
BAP 4.0	6.07 bc	11.12 a	7.37 a
AIA 2.0	4.24 de	10.75 a	8.12 a
AIA 4.0	6.07 bc	9.87 ab	6.50 ab
HBr 2.0	7.48 a	10.75 a	7.75 a
HBr 4.0	7.00 ab	10.87 a	7.50 a
Control	5.35 bc	7.62 b	6.50 ab
CV (%)	13.4	16.4	18.4

*Values with different letters within each factor and column are statistically different ($p \leq 0.05$). BAP(6-benzylaminopurine), HBr (Brassinosteroid Cidef-4), IAA (Indolacetic Acid).

Table 1.

Morphological components in apical meristem explant of *Musa spp. cv gran enano* in interaction with different concentrations of growth regulators during the *in vitro* multiplication stage.

increase in regrowth height from 0.5 mgL of BAP has been reported [51]. This cytokinin has been widely used in the *in vitro* regeneration of various plants [52] and induces the formation of axillary shoots and adventitious roots of the meristematic explant in banana [53] and their efficacy in different banana cultivars has been demonstrated in foliar growth [54]. Auxins in *in vitro* culture induce root formation and shoot growth [54, 55]. In the case of HBr, the response was different, that is, the inclusion of the two concentrations in the medium induced a similar increase in the height of the explant and was superior to the other treatments ($P \leq 0.05$) [56]. Stems were thicker with HBr and thin with narrow leaves in the control. In *Solanum tuberosum* L. the height of the explants was also increased after 30 days of age, by adding 1 mgL⁻¹ of HBr Biobrás-6 to the medium.

Likewise, pronounced effect on the stimulation of proliferation and shoot length when HBr (0.2 μ M) was added to the MS medium compared to the banana control [57]. Robres-Torres [58] add that the application of brassinolide plus benzyladenine (BA) in *Rubus idaeus* L. did not express dependence in the combination of both regulators in the expression of regrowth, but when applying HBr alone, regrowth elongation was increased.

The number of leaves is higher when including growth regulators compared to the control and they were statistically different ($p \leq 0.5$). The inclusion of HBr increased the leaf area of *Lactuca sativa* L. lettuce with the foliar spraying of HBr Biobras-16 [59] and with the regeneration of adventitious shoots in cauliflower hypocotyls and *Spartina* [60].

When comparing the induction of the number of roots between the three growth regulators, a differential response was found according to the concentration used and without difference with the application of the concentrations in HBr (Cidef-4).

Applying HBr to plants increases the number of roots. Similar result cites [61] when doing repeated foliar sprays with BB-16 (0.05 mgL⁻¹) in *Cattleya leuddemanniana* and *Guarianthe skinneri* (Bateman), Dressler and W. E. Higgins. This same result was obtained with con different orchids, like *Oncidium sphacelatum* Lindl., *Trichocentrum andreanum* (Cogn.) R. Jiménez and Carnevali, *Epidendrum stamfordianum* Bateman, *Guarianthe skinneri* (Bateman), Dressler and W. E. Higgins, *Guarianthe aurantiaca* (Bateman ex Lindl.) Dressler and W. E. Higgins and *Brassavola nodosa* (L.) Lindley, in Chiapas, México [37].

2.2 Influence of brassinosteroid in the acclimation of banana clone *Musa* spp. and *Saccharum officinarum*

The production of micropropagated plantlets is used nowadays to increase new cultivars [62], but there are certain drawbacks in its *ex vitro* phase that limit its widespread use, especially in resisting the stress of transplantation [47, 48]. Exogenous application of HBr has been shown in some plants to improve *ex vitro* survival [18].

The application of HBr in its different frequencies of HBr (Cidef-4) with 4 mgL^{-1} , (6 every 14 days, 3 every 28 days and 2 every 42 days), were efficient in inducing the growth of *Musa* spp. However, the more frequent applications induced greater height.

In the case of plant height with the application of HBr every 14 days, it presents a statistical difference ($p \leq 0.05$) with the rest of the treatments during the four samplings and in all the application frequencies it was superior to the control (Table 2). The results suggest an interaction between the concentration and its frequency of application, and Mandava [63] adds that the brassinosteroid is more sensitive when the young

Time (days)	Treatments	Height (cm)	Stem thickness (cm)	Number of leaves
14	1. Control	5.53 b*	0.53 a	5.5 ab
	2. HBr every 14 days	6.41 a	0.50 a	5.5 b
	3. HBr every 28 days	5.95 ab	0.45 a	4.8 b
	4. HBr every 42 days	5.60 b	0.51 a	6.0 a
	**CV (%)	8.59	10.64	11.46
42	1. Control	10.33 b	0.63 b	6.5 a
	2. HBr every 14 days	12.63 a	0.65 b	6.5 a
	3. HBr every 28 days	11.06 ab	0.61 b	6.1 a
	4. HBr every 42 days	11.25 ab	0.78 a	7.0 a
	CV (%)	10.62	7.07	8.25
70	1. Control	10.78 b	0.75 b	7.8 a
	2. HBr every 14 days	16.33 a	0.90 a	8.0 a
	3. HBr every 28 days	13.71 b	0.80 ab	7.6 a
	4. HBr every 42 days	13.83 b	0.86 ab	8.5 a
	CV (%)	7.72	8.87	6.65
84	1. Control	15.21 b	0.81 a	8.3 a
	2. HBr every 14 days	19.17 a	1.01 a	8.6 a
	3. HBr every 28 days	16.33 ab	0.91 a	8.1 a
	4. HBr every 42 days	16.08 ab	1.03 a	8.8 a
	CV (%)	14.6	17.0	9.2

*Values with different letters within each factor and column are statistically different ($p \leq 0.05$). HBr (Brassinosteroid 4 mgL^{-1} de Cidef-4).

**CV = Coefficient of variation. The number of applications. The first 14 days only 1 application in treatment 2. Application at 42 days 3 applications in treatment 2, and 1 application treatment 3 and 4. Evaluation at 70 days, 5 applications treatment 2, 3 application treatment 3 and 1 application treatment 4. Evaluation at 84 days, 6 applications treatment 2, 3 application treatment 3 and 2 application treatment 4.

Table 2. Morphological components of *Musa* spp. cv “great dwarf” in interaction with different frequencies of HBr application (Cidef-4).

tissues present higher content of auxins and the expression of the growth of the plants treated with exogenous HBr can be related to the endogenous plant hormones [18].

Similar results, but with a lower proportion of plant height induction (2%) have been reported by Jeyakumar et al. [64] in “robusta” banana when applying a foliar HBr at a concentration of 0.2 mg kg⁻¹ at four and 6 months after having been planted. On the other hand, Izquierdo et al. [33] mention an increase in plant height in banana clone FHI-18 applying by immersion and foliarly before transplantation the HBr BB-6. However, Herrera et al. [65] indicate an increase in the height of plants in banana cultivation with the application of Br applied every 28 days, compared to the application of every 14 days, which, in our case, was the most frequent application and was expressed in the greater height of the regrowth. It is probable that the response is associated with the rapid degradation of Br, as indicated by Janeczko et al. [44], and the more frequent application, may favor its action for a longer time. Application of BR by spraying at specific developmental stages can enhance crop yield [66].

In our case, the thickness of the stem and the number of leaves are the variables with the lowest morphological expression in the different treatments evaluated and they only present statistical differences in two or three of the samplings carried out. On the other hand, Izquierdo et al. [33] reported an increase in the number of leaves and diameter of the pseudostem in banana with the application of HBr, as in the clone FHIA-18, when applying the HBr BB-6.

In potato plants (*S. tuberosum* L.) they found an increase in stem length, fresh biomass and a greater number of minitubers per plant [50].

In the case of the number of leaves, [67] mention a significant increase in the number of leaves of shoots of *Vriesea* plants (Bromeliaceae) when HBr (MH5) was applied, improving their quality indicators after 49 days of acclimatization. In our case, no differences were expressed between the frequencies of HBr application in this variable.

In the physiological components, the application of HBr alone every 42 days increased the dry biomass of the shoot and was statistically different ($p \leq .05$) to the rest of the treatments, but in the root system the biomass allocation was similar in all the treatments and no statistical difference (**Table 3**).

It has been shown that in the mustard plant *Brassica juncea* (L.) Czern, the yield components were increased with the application of 28-homobrassinolide [43]. Brassinosteroids likely play a crucial role in modulating plant growth and development, which affect crop architecture and yield [7].

In *S. officinarum*, plants var. CP 72–2086 were used. The height of the plant with the frequencies and concentrations evaluated of HBr (Cidef-4) was statistically higher than the control ($P \leq 0.05$) (**Table 4**).

Treatments	(g.plant ⁻¹)	
	Shoot	Root
Control	2.50 b	0.62 a
HBr every 14 days	2.65 b	0.66 a
HBr every 28 days	2.19 b	0.60 a
HBr every 42 days	3.39 a	0.66 a
**CV (%)	10.63	7.01

Table 3. Comparisons of means of physiological variables of the giant dwarf clone of *Musa* sp. in the process of acclimatization with different frequencies of application of an HBr at 84 days after sowing.

Treatments	Height (cm)	Stem thickness (cm)	Number of leaves
Control	90.8 b	6.3 b	11.5 abc
1% HBr every 14 days (5 applications)	112.6 a	7.5 ab	15.1 a
2% HBr every 14 days	117.6 a	7.8 a	10.0 c
3% HBr every 14 days	113.6 a	7.7 a	12.1 abc
1% HBr every 28 days (2 applications)	109.8 a	7.6 a	11.5 abc
2% HBr every 28 days	110.8 a	7.9 a	10.6 bc
3% HBr every 28 days	118.6 a	7.3 ab	11.5 abc
1% HBr every 42 days (1 application)	114.0 a	7.3 ab	12.3 abc
2% HBr every 42 days	112.0 a	7.2 ab	14.1 ab
3% HBr every 42 days	117.8 a	7.3 ab	11.3 abc
**CV (%)	8.59	9.3	17.3

Table 4.

Morphological components of the yield in Saccharum officinarum in interaction with frequencies and concentrations of HBr (Cidef-4).

In general, the application frequencies in interaction with the concentrations shows a slight increase in plant size as the concentration increased from 1 to 3%. The most frequent applications of 14 and 28 days, also present a certain tendency in the increase of the height of the plant.

HBr (Brassinosteroid Cidef-4), % CV (coefficient of variation). Letters that are not the same indicate statistical difference (Tukey, $P \leq 0.05\%$). Seventy day Data.

In the cane variety (C0. 86,032), brassinolide induced a positive effect on *ex vitro* plants of stem elongation, leaf formation and adventitious shoots during 3 weeks, in addition, survival increased as the concentration of brassinolide increased [68]. The above effect has been demonstrated by inducing elongation and cell division, resulting in growth, thickening and curvature in oat coleoptiles [63].

The applications of exogenous brassinosteroids induce diverse physiological responses, in addition to cell expansion, vascular differentiation, reproductive development, seed germination, flowering, and fruiting [69, 70].

2.3 Flowering and fruiting response of *Theobroma cacao* L

Cocoa was used as food, medicine and tribute in Mesoamerican territory [71, 72]. It is a crop with wide flowering and low fruiting percentage. This is attributed to environmental and hormonal factors.

The application of potassium nitrate alone does not show a positive effect in increasing the flowering and fruiting variables in *T. cacao* L. A similar response is cited by García et al. [73] when applying 2% KNO_3 in *Acca sellowiana* [O. Berg] Burret and the lack of response assign it to necrotizing effects on flower buds and fruits. The differences occurred with the application of HBr alone and in combination with different concentrations of potassium nitrate.

The increase in flowers was expressed with the application of HBr alone and was statistically different from the other treatments ($P \leq 0.05$). Combinations with KNO_3 present a number of flowers similar to the control (Table 5).

T. cacao L sowing 3 × 3 with a shade of *Inga micheliana* Harms., *Pouteria sapota* (Jacq.) and *Tabebuia rosea* (Bertol.) A. DC. The values are averages of 5 trees and 4 branches of 0.5 m per tree after the trunk toward each cardinal point.

Treatments	Number tree ⁻¹			
	Flowers	Opennings Flowers	Cherelles	Fruits
1) Control	6.5 c**	17.2 b	4.32 bc	1.96 bc
2) 2% de ***KNO ₃	6.3 c	18.1 b	3.50 cd	1.86 bc
3) 1.5% HBr	9.1 a	26.5 a	4.66 b	1.66 bc
4) 1.5% HBr + 2% KNO ₃	5.7 c	18.10 b	3.04 d	1.32 c
5) 1.5% HBr + 4% KNO ₃	7.8 b	16.16 b	4.80 b	2.42 ab
6) 1.5% HBr + 6% KNO ₃	8.3 ab	17.46 b	7.72 a	2.98 a
****CV %	7.9	7.6	10.2	22.8

** The letters that are not equal between the columns indicate significant statistical difference ($p \leq 0.05$). ***KNO₃ (13N-2P-44K at the rate of 200 g in water 10 l). HBr was applied only at 1.5% in September and in combination with KNO₃ a, 2, 4 and 6% in September, October and November. The values are averages of 15 weeks of evaluation in treatments 1, 2, 3, and 4 that were applied in September. Treatment 5 applied in October with nine samplings and treatment 6 applied in November with 4 samplings. **** Coefficient of variation (%).

Table 5.
 Flowering and fruiting of *Theobroma cacao* L. regional criollo type in response to the applications of potassium nitrate and homobrassinolide.

The number of flowers increased 40% with the application of HBr and 87% when combined with KNO₃ at 2% in relation to the control. It is important to indicate that the application of HBr was in September, the time when the second most important flowering flow begins in *T. cacao* L in this region, and although no detrimental effects were observed in floral structures, the emission was decreased. On the other hand, they increased 27% in relation to the control when combining HBr plus 6% KNO₃, which was applied in November. At this date the night temperature decreases and fruiting is favored.

On the other hand, the conversion of flowers to small fruits was higher with the application of KNO₃ and HBr in November and this process continued until the formation of fruits, which were statistically different from the rest of the treatments. ($P \leq 0.05$). However, the highest number of fruits found is more related to the date of application of the treatment at the end of the year, that is, October and November. This suggests that the development of the fruit requires the protection of HBr to achieve the conversion of Cherrille to fruit, which seems to be a critical period. In addition to the above, fruiting demands more photosynthates and HBr has been shown to stimulate CO₂ assimilation [74, 75]. In other crops, the application of HBr has been effective to promote the increase in yield in annual crops [10, 76] and perennials as in *Passiflora edulis* f. *flavicarpa* applied after flowering has appeared [77].

2.4 Application of a brassinosteroid affects quality in 'ataúlfo' mango

Currently, there is a decrease in mango yield in the south-southeast of Mexico due to various causes attributed to environmental and management factors. Current results in plantations are reflected in high floral abortion rates, premature fruitlet drop and the presence of parthenocarpic fruits or "nubbins. It is considered that HBr can promote fruiting [8, 16, 29] by demonstrating the beneficial effects of HBr in different crops such as vegetables, legumes, cereals, fruits and oilseeds [44, 45, 74], in addition to improving the quality of crops [27, 44, 46, 78] and the yield [46].

Mangoes from all treatments were harvested and the values of each variable were recorded 12 days after harvest (Table 6).

Time (Days)	Treatments	Firmness (N.frut-1)	Total sugars. fruit-1	Total soluble solids (°Brix.fruit-1)
12	**KNO ₃	0.61 b*	0.18 b	18.05 b
	***HBr(2 g) + KNO ₃	0.64 ab	0.27 a	19.80 a
	HBr (4 g) + KNO ₃	0.57 b	0.19 b	17.92 b
	HBr (6 g) + KNO ₃	0.70 a	0.25 b	20.07 a
	CV %	5.9	7.0	3.5

*Values with the same letter within each factor and column are equal according to Tukey test $P \leq 0.05$. CV = Coefficient of variation, (1) **Control (2% Potassium nitrate KNO₃), (2) *** 2 g of HBr Cidef-4/hectare applied on October 16; (3) 4 g of HBr Cidef-4/hectare applied in two equal parts on October 16th and 30th; (4) 6 g of HBr Cidef-4 applied in three equal parts on October 16th and 30th and on November 14th.

Table 6.

Quality variables during 'Ataulfo' mango ripening sprayed at preflowering and preharvest with HBr Cidef-4 stored at room temperature ($22 \pm 2^\circ\text{C}$).

The spraying of HBr significantly affected ($P \leq 0.05$) the firmness of the mangoes. The applications of the fruits induced greater firmness in the treatments with 2 g ha^{-1} and 6 g ha^{-1} . The greatest difference was registered in the treatment with three applications of HBr. In other crops, such as papayas, they cite an increase in the firmness of the fruits when increasing the application of 24-epibrassinolide (epiBR) to $1 \mu\text{M}$ [79].

Total sugars increased by combining HBr plus KNO₃ (Table 6) as the amount of (HBr) increased. In this regard, the highest value was found in the fruits that were sprayed with 2 and 6 g of HBr and it was statistically different from the rest of the treatments ($P \leq 0.05$). In general, the results express a more consistent response in terms of the average sugar content in the treatments applied with potassium nitrate and in the HBr treatments ($2 \text{ and } 6 \text{ g ha}^{-1}$).

The amount of TSS found in the treatment with potassium nitrate was 18.0°Brix and in the treatments where HBr was sprayed three times (2 g ha^{-1}), the value fluctuated between 19.8°Brix . An increase in the content of total soluble solids without the influence of the concentration of HBr was reported in watermelon [80] but a higher content of total soluble solids is confirmed in *P. edulis* compared to the control with the application of HBr [77]. In papayas, the total soluble solids of the fruits were increased with the application of HBr [79].

In-plant tissues, exogenous applications of HBr induce ethylene production [81, 82] and in some fruits such as mango and strawberries, HBr in small amounts may not be critical for ripening [83, 84], but exogenous applications they are capable of inducing ethylene production [41, 83]. In the Kensington Pride mango variety, ethylene production and peak respiration occurred on the fourth day of ripening, and although the amount of HBr in the fruit was traces, the exogenous application of Epi-BL ($45 \text{ and } 60 \text{ ng g}^{-1}$) promoted fruit ripening [83].

2.5 Flowering and yield response of *Coffea arabica* cv catimor

C. arabica L. plantations currently face contrasting changes in the amount of rainfall and the distribution of rainfall that has decreased their yield. It also interacts with internal, hormonal and nutritional factors [85]. The importance of the interaction of the environment in the flowering of fruit trees has been pointed out [86].

The presence of stressors, biotic or abiotic, that affect crop yield under field conditions, can be attenuated by applying growth regulators, or, when the warm-humid temperatures of the tropics do not induce reproductive cell differentiation,

Treatments	Number of flowers	length of lateral branches (cm)	Number of leaves on lateral branches	Fruits number
Control (water)	13.6 b*	54.3 ab	13.6 ab	37.0 c
HBr (0.5%)	19.0 ab	60.6 ab	15.4 a	44.2 ab
HBr (1.0%)	17.6 b	65.9 a	14.3 ab	52.9 a
KNO ₃ (1%)	13.8 b	54.1 ab	12.4 ab	29.0 c
HBr (0.5%) + KNO ₃ (1%)	27.1 a	55.3 ab	14.1 ab	41.1 b
HBr (1%) + KNO ₃ (1%)	17.8 ab	61.9 ab	14.3 ab	40.8 b
CV %	31.0	22.0	21.7	20.5

*The letters that are not equal between the columns indicate significant statistical difference (Tukey $p \leq 0.05$). The application of the four treatments was carried out in February: HBr Cidef-4 al 0.5%, HBr Cidef-4 al 1%, KNO₃ al 1%, HBr Cidef-4 al 0.5% + KNO₃ al 1%, HBr Cidef-4 al 1% + KNO₃ al 1% + and Control (water application). The plantation of *C. arabica* L. variety Catimor with a shade of *Inga micheliana* Harms. In each tree, a branch was identified toward each cardinal point and in it, an area of 50 cm was indicated, for the taking of variables of flowering and fruiting.

Table 7. Comparisons of means of the number of flowers, leaves, fruits and length of four bands in *Coffea arabica* L. var. Catimor in response to the application of HBr in interaction with potassium nitrate.

has been increased by potassium nitrate. Potassium nitrate as a flowering inducer and brassinosteroidal steroidal hormone can favor fruit growth in *Coffea* spp., as in *Mangifera indica* L [87]. The application of HBr (Cidef-4) alone and in combination with 1% KNO₃ was evaluated.

The number of flowers increased with the application of potassium nitrate plus HBr in the lowest concentration and was higher than the rest of the treatments ($P \leq 0.05$). It represented an increase of 100% in relation to the control. The lowest number of flowers was recorded in the control and when only potassium nitrate was applied. The HBr applications registered a mean increase in the number of flowers when it was applied alone, or in interaction with potassium nitrate, however, the highest values were presented when the concentration of 0.5% of HBr was included (Table 7).

The application of 1% KNO₃ alone induced flowering similar to the control. On the other hand, with the application of the two concentrations of brassinosteroid alone, the difference in the number of flowers was 36% in relation to the control.

The growth of the bandola and the number of leaves increased in the treatments where only HBr was applied at the two concentrations, or in combination with potassium nitrate and the lowest values were with the application of potassium nitrate alone and the control.

The application of HBr induces a wide range of responses, including an increase in the cellular expansion of the leaves, increased elongation of the stem [14, 15, 28, 88] that increase the leaf surface, plant biomass and the yield of various crops [8].

The average number of fruits increased 29% more per band with the 0.5 and 1% HBr treatments. The combination of 1% KNO₃ plus 0.5 and 1% of HBr induced similar values to the applications of HBr alone.

In different crops of economic importance, brassinosteroids are characterized by stimulating plant growth, increasing the yield of biomass production and accelerating the ripening of fruits. In addition, they strengthen the resistance of plants to pests and abiotic stressors such as drought and sudden changes in temperature [63].

It was discovered that Br promotes tomato yield through improved autophosphorylation of SlBRI1, and increased plant expansion, leaf area, fruit weight and number of fruit per cluster [10]. Brassinolide (BL), 28-homobrassinolide (28-hBL) and 24-epibrassinolide (24-eBL) treatments stimulated both tomato growth and yield [41].

3. Conclusions

HBr favors the growth of *Musa* spp. *in vitro* through regrowth height and similarly to BAP and AIA regulators in the number of leaves and roots. In *ex vitro* conditions, there were differences in the growth of *Musa* spp. and *S. officinarum* between the application frequencies and in general, the increase in the number of applications favors this process.

The exogenous applications of HBr in *T. cacao* L, *M. indica* L. and *C. arabica* L. show variable results. Plant growth is modified as in *C. arabica* L and flowering is influenced as in *T. cacao* L and *C. arabica* L. and the yield in both crops. The higher concentrations applied did not necessarily increase the crop yield or the combination with potassium nitrate. The quality of the mango fruits showed variations in the concentrations of HBR but it was better alone or in combination with nitrate. Under field conditions, we consider it important to identify the dose and timing of HBr application in the plant organ of interest, be it flowers, foliage or fruits.

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