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GROWTH OF 'PRATA-ANA' BANANA'S MICROSHOOTS CLONE GORUTUBA FROM SYNTHETIC SEEDS: SUBSTRATES AND BAP CONCENTRATION[1]

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ABSTRACT - The banana crop stands out as an activity of great social and economic importance in Brazil, which occupies the fifth place in world production. Synthetic seed production is becoming promising for a micropropagation and *in vitro* conservation. The aim of the study was to analyze the conversion and growth of 'Prata-anã' banana's microshoots clone Gorutuba from synthetic seed in MS medium and vermiculite, different substrates and concentrations of BAP (6-benzylaminopurine) associated with ANA (acetic naphthalene acid) in the constitution of its capsule were tested. The microshoots were immersed in the sodium alginate matrix (3%) and dripped in a solution of $CaCl_2.2H_2O$ (100 mM) for complexation and then in KNO_3 solution (100 mM) to decomplex. The experimental design was completely randomized in a 2 x 5 factorial design (substrate x BAP concentrations), containing different substrates (MS culture medium and vermiculite) and BAP concentrations (2.22, 4.44, 6.66, 8.88 and 13.32 μmol L^{-1}) associated with NAA (naphthalene acetic acid) 0.54 μmol L^{-1}, totaling 10 treatments, with 4 replicates, and that each replicate containing 5 seeds. The evaluations of conversion, number of leaves, leaf length, leaf height, number of roots, root length and oxidation were performed at 30 and 60 days.The use of the MS medium provided better growth results in relation to vermiculite as substrate, in which the different BAP concentrations did not differ from each other. It was found that, in MS culture medium, BAP concentrations above 8.88 μmol L^{-1} in the capsule composition are not indicated for microshoots growth.
Index terms: encapsulation, micropropagation, *Musa* sp., growth regulators.

CRESCIMENTO DE MICROBROTOS DE BANANEIRA 'PRATA-ANÃ' CLONE GORUTUBA A PARTIR DE SEMENTES SINTÉTICAS: SUBSTRATOS E CONCENTRAÇÃO DE BAP

RESUMO-A bananicultura destaca-se como atividade de grande importância social e econômica no Brasil, o qual ocupa o quinto lugar na produção mundial. A produção de semente sintética vem tornando-se promissora para a micropropagação e conservação *in vitro*. O objetivo deste trabalho foi analisar a conversão e o crescimento de microbrotos de bananeira 'Prata-anã' clone Gorutuba a partir de semente sintética em meio de cultura MS e vermiculita, testando-se diferentes substratos e concentrações de BAP (6-benzilaminopurina), associado a ANA (ácido naftaleno acético), na constituição de sua cápsula. Os microbrotos foram mergulhados à matriz de alginato de sódio (3%) e gotejados em solução de CaCl2. 2H2O (100 mM) para complexação e depois em solução de KNO3 (100 mM) a descomplexação. O delineamento experimental foi inteiramente casualizado, em esquema fatorial 2 x 5 (substrato x concentrações de BAP), contendo diferentes substratos (meio de cultura MS e vermiculita) e concentrações de BAP (2,22; 4,44; 6,66; 8,88 e 13,32 μmol L^{-1}), associado a ANA (ácido naftalenoacético) 0,54 μmol L^{-1}, totalizando 10 tratamentos, com 4 repetições, sendo que cada repetição conteve 5 sementes. As avaliações de conversão, número de folhas, comprimento de folhas, altura, número de raízes, comprimento de raízes e oxidação foram realizadas aos 30 e 60 dias. A utilização do meio de cultura MS proporcionou melhores resultados de crescimento em relação à vermiculita como substrato, na qual as diferentes concentrações de BAP não se diferiram. E pôde-se constatar que, em meio de cultura MS, concentrações de BAP acima 8,88 μmol L-1, na composição da cápsula, não são indicadas para o crescimento dos microbrotos.
Termos para indexação: encapsulamento, micropropagação, *Musa* sp., fitorreguladores.

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INTRODUCTION

Banana cultivation stands out as an activity of great economic and social importance in Brazil, which is considered a major banana producer, ranking fifth in world production (FAO, 2012). In the national production, the main banana producers are the states of São Paulo, Bahia, Minas Gerais, Santa Catarina and Pará (IBGE, 2014). In the north of Minas Gerais, the municipalities of Janaúba and Jaíba stand out in the national fruticulture as the great banana producers and having as highlight the 'Prata-anã' cultivar, ranging from small to large producer (DONATO et al., 2009).

The 'Prata Anã' banana is a vigorous rustic plant with good weed competition and high resistance to 'banana tree borer' and nematodes. However, this cultivar is affected by diseases such as Yellow Sigatoka and Panamá Disease (LICHTEBERG & ZAFARI, 2003).

According to Rodrigues (2012), a somaclonal variant known as 'Prata-Anã' clone Gorutuba has been used for the expansion of new areas. This clone is resistant to some isolates of *Fusarium oxysporum* f.sp. cubense, a fungus that causes Panama disease (RODRIGUES, 2010). In regions where there is a great infestation of the disease, this type of plant is an excellent alternative for cultivation.

In this context, the use of biotechnological techniques for the expansion of the crop and the production of seedlings free of this pathogen is important. The micropropagation is a vegetative form of propagation of small segments of plants (shoot apices, buds, meristems, leaf fragments or roots, among others), and is carried out in specific containers containing suitable nutrient medium (ULISSES et al., 2010). This technique has been used for the production of seedlings of several plant species, including banana tree, which guarantees large production of plants free of pests and diseases in a short period of time (MORAIS-LINO et al., 2008).

The use of biotechnological techniques, such as the production of synthetic seeds, has been highlighted as an important technique for the micropropagation and *in vitro* conservation of several species due to the possibility of being stored without losing viability (RAI et al., 2009).

In addition to such synthetic seeds having the ability to imitate a seed by the use of coating material and artificial endosperm for the nutrition of the explant, these capsules may be enriched with elements commonly used during the production of artificial seeds such as macro and micronutrients, vitamins, fungicides and sucrose (SAIPRASAD, 2001; SANDOVAL & GUERRA, 2002; MARTIN, 2003). The phytoregulators are also commonly used, such as cytokinins, which are indispensable for cell division and multiplication, apical dominance breakage, induction and proliferation of axillary buds and differentiation of adventitious buds (BIST et al., 2011); and auxins, which are fundamental in the induction of cell division and in the induction of roots, and are also used in the multiplication phases to favor growth (BORGES et al., 2012).

Therefore, as with synthetic seed, micropropagation is also a modality within tissue culture, but the main differential advantages that are objected when using the synthetic seed technique are associated with the elimination of the final phase of micropropagation (that is, rooting and acclimatization) (SINGH et al., 2006).

Thus, the objective of this study was to analyze the conversion and growth of microshoots of 'Prata-anã' banana clone Gorutuba, encapsulated in alginate under the technology of synthetic seed, using as substrate the culture medium MS and vermiculite, being tested different concentrations of BAP (6-benzylaminopurine) associated with NAA (naphthalene acetic acid) in the constitution of its capsule.

MATERIAL AND METHODS

The present study was conducted at the Laboratory of Plant Biotechnology of the Agricultural Research Company of Minas Gerais - EPAMIG, EPAMIG North - Gorutuba Experimental Field, Nova Porteirinha - MG.

The microshoots were obtained from seedlings pre-established during 120 days of *in vitro* culture. These plants originate from seedlings of banana tree little horny type of the cultivar 'Prata-Anã' clone Gorutuba, from the EPAMIG Experimental Field, in which the outer sheaths were removed, which posteriorly had their size reduced, the disinfestation was carried out in the flow chamber with 70% alcohol for 5 minutes and in 2% sodium hypochlorite for 30 minutes, followed by the triple wash in autoclaved distilled water. In a laminar flow chamber, the explants underwent a second mechanical cleaning step. With the aid of a scalpel, the explants were reduced to 3cm in length and established in 200 ml vials containing 30 ml of solid culture medium comprising: mineral salts and vitamins of the MS culture medium (MURASHIGE and SKOOG, 1962) supplemented with 31.08 μmol L^{-1} of BAP, 30 g L^{-1} sucrose, 0.1 g of inositol, pH adjusted to 5.8 ± 0.1, previously the addition of the agar gelling agent at

7 g L^{-1}, and autoclaved at 121°C and 1.5 atm for 20 minutes for sterilization. The explants were incubated in a growth room at controlled temperature (25 ± 2°C) and under a photoperiod of 16 hours of light (30 W/m²). The subculture of the explants was performed every 30 days.

These explants were subcultured obtaining, on average, 4 shoots per explant. For the encapsulation, 4 cm microshoots were immersed into the sodium alginate matrix (3%) in MS culture medium, using one microshoot per capsule and removing them with the help of an automatic pipette set to 700 µL. In the sequence, the encapsulating units were dripped into $CaCl_2$ solution.$2H_2O$ (100 mM) in which they remained for 20 minutes for complexation. The synthetic seeds individually formed, were subjected to triple washing in distilled and sterilized water and, then, immersed in KNO_3 solution (100 mM) for 15 minutes for decomplexing, and in the sequence, submitted to triple washing again in distilled and sterilized water. In the microtubule encapsulation, different concentrations of BAP (2.22, 4.44, 6.66, 8.88, 13.32 µmol L^{-1}) associated with NAA (0.54 µmol L^{-1}) were used. For the *in vitro* conversion of the synthetic seeds, these were established in 200 ml glass vials using solid MS culture medium (30 ml/vial) at the same concentrations previously cited. Such medium had its pH adjusted to 5.8 ± 0.1 and autoclaved at 121 °C and 1.5 atm for 20 minutes for sterilization. The synthetic seeds were also established, *in vitro*, in autoclaved vermiculite (20 g/vial), at 121ºC and 1.5 atm for 20 minutes, in order to evaluate if there could be growth in vermiculite even though they had less nutrients, since the nutrients for the microshoots were only from the capsule. These treatments, which had vermiculite as a substrate, were periodically irrigated with autoclaved distilled water using an automatic pipette adjusted to 700 µL, so that moisture in the vials was always favorable for the conversion and growth of the synthetic seed, and that it did not dehydrate. In both MS and vermiculite medium, all different concentrations of BAP were used.

The experimental design used was completely randomized in a 2 x 5 factorial scheme (substrate x BAP concentrations) containing different doses of BAP (6-benzylaminopurine) 2.22; 4.44; 6.66; 8.88; 13.32 µmol L^{-1} associated with NAA (naphthalene acetic acid) 0.54 µmol L^{-1}, totalizing 10 treatments, with four replicates, each replicate containing five synthetic seeds. The evaluations were carried out in the period of 30 and 60 days, in which were evaluated the conversion, number of leaves, length of leaves and height at 30 days; and number of leaves, leaf length, height, number of roots, roots length and oxidation at 60 days.

The data were submitted to analysis of variance ($F < 0.05$) by the statistical program Sisvar (FERREIRA, 2008) and, when significant, it was performed the regression analysis for doses and Tukey test in order to compare the use effect of the culture medium and vermiculite.

RESULTS AND DISCUSSION

- **Analysis at 30 days**

According to the analysis of variance shown in Table 1, it was verified that there was interaction of substrate effects and concentrations of BAP (SxC) for the conversion variables (CONV), leaf number (LN), leaf length (LL) and height (H).

For the conversion of synthetic seeds to *in vitro* plants (CONV), it was verified that there was an increase in the number of microshoots converted to plants as the concentration of BAP was increased for the treatments that used the MS medium, and the concentration of 8.22 µmol L^{-1} presented the maximum value of conversion percentage, 55.08%. From this concentration, it is observed a decrease in the percentage of conversion caused by the increase in the concentration of BAP, which may have acted in an inhibitory way. When vermiculite was used as substrate, the different concentrations of BAP did not differentiate in the conversion of synthetic seed to plant, presenting 57.0% in all concentrations (Figure 1A). It is believed that irrigation was fundamental for the conversion of the microshoot in the vermiculite, since the conversion percentage was higher in this substrate than in culture medium. Although the vermiculite did not provide moisture and nutrients for the microshoots, the water supplemented once a week proved to be efficient for the control of microshoots dehydration. It is also evidenced that endogenous hormones from the microshoot itself were effective in converting the synthetic seed into a new plant, showing that the absence of nutrients from the MS culture medium did not interfere with this characteristic. The culture medium, for being semi-solid, provides the amount of water in a slow way and, in spite of providing the macro and micro nutrients necessary for the microshoot development, the immediate availability of the water may have been a differential in the conversion of 'Prata-anã' banana's microshoots clone Gorutuba.

According to Adriani et al. (2000), *in vitro*

plant conversion is the most important aspect of synthetic seed technology and, despite of this, is one of the limiting factors of the practical use of this tool. Ganapathi et al. (2002) obtained 100% conversion of synthetic seeds to *Musa* sp. cultivar 'Basrai' using 5 mg L^{-1} of BAP in MS medium, whereas Sandoval-Yugar et al. (2009) obtained from 70 to 80% of conversion in banana tree cultivar 'Grand Naine'; in the 'Nanicão' cultivar, Matsumoto et al. (1995) obtained 75% conversion; and in 'Rasthali' cultivar, Ganapathi et al. (2001) observed a 66% conversion of synthetic seeds, evidencing that the conversion of synthetic seed is dependent on the genotype or cultivar used (HASSANEIN et al., 2011), since in our study only 55.08% of conversion was obtained in semi-solid medium for 'prata-anã' banana tree clone Gorutuba.

According to Sandoval-Yugar et al. (2009), in the case of banana (*Musa* sp), the use of synthetic seed techniques can improve the quality of plants *in vitro* and decrease production costs.

For the leaf number characteristic (LN), an increasing effect was observed as a function of the different concentrations of BAP when the MS culture medium was used up to the concentration of 9.34 μmol L^{-1} in which it was possible to verify the maximum of 1.48 leaves, then decreasing due to a possible inhibitory effect caused with the increase of the cytokinin concentration. The amount of endogenous cytokinin up to the above concentration should have been balanced with the provided in culture medium which, from its elevation, may have caused a phytotoxic effect, thereby reducing the number of leaves produced per plant. When vermiculite was used as a substrate, BAP concentrations did not significantly influence the number of leaves, obtaining approximately 1.38 leaves in the evaluation period (Figure 1B). The vermiculite presents only like a support for the development of the synthetic seeds. In this case, there was no nutrient supplementation via irrigation, which was performed only with water. In view of this fact, it is noticed that an external supplementation with the macro and micronutrients of the MS is favorable for the formation of leaves in the *in vitro* plants of 'Prata-anã' banana tree clone Gorutuba.

The number of leaves is an important characteristic and possibly leaves with higher number of leaves have higher rates of vegetative plant propagation in the field, since the leaves are the structures responsible for the capture of solar energy and the production of organic matter through photosynthesis (SOUSA, 1994). Therefore, a well developed foliar system allied to the root system allows better *in vitro* development of the plant as well as its *ex vitro* vegetative plant propagation.

For leaf length (LL), an increase in length was observed according to the increase in concentrations up to 8.19 μmol L^{-1} of BAP, with a maximum value of 10.38 mm, when the MS medium was used; from this concentration, there was a decrease. When using vermiculite as a substrate, there were no significant differences in leaf length, with 2.95 mm in all concentrations used (Figure 1C).

Similar results were found by Villa et al. (2007) working with mulberry and grapevine, in which the length of the aerial part reached the maximum value with the use of 2.0 mg.L^{-1} of BAP and with 3.0 g.L^{-1} of activated charcoal and, from this point, the BAP growth regulator and/or activated charcoal began to inhibit the development of plants *in vitro*, showing a decrease in length. However, Ferdous et al. (2015) observed in banana tree cultivar Amritasagar that the dose of 5.0 mg.L^{-1} of BAP was the most responsive in plant multiplication *in vitro*.

Analyzing the number of leaves and relating to their size, it was observed that in this study the leaves produced had a relatively large size compared to the amount of them. This can be attributed to the fact that the BAP growth regulator stimulates the formation of a greater number of shoots, but these shoots presented a reduced size, with a smaller number of nodal segments and leaves being evidenced (ASMAR et al., 2012).

Regarding the height of the plants *in vitro* (H), it could be verified that there was the same increasing effect when using the MS medium, according to the increase of the BAP concentration up to 9.73 μmol L^{-1}, in which the maximum height of the plants was 22.97 mm; thereafter, there was an opposite effect, that is BAP inhibited plant growth *in vitro*. When using vermiculite as substrate, there were no significant differences for height, presenting 6.86 mm in the different concentrations of BAP (Figure 1D).

Ferdous et al. (2015) found greater number of leaves for banana tree cultivars Sabri and Amritasagar in the dose of 5.0 mg.L^{-1} of BAP; dose ratio higher than that used in our experiment, again noting that the dose of cytokinin is dependent on the genotype used.

This result shows the importance that the MS medium has for the development of microshoots in 'Prata-anã' banana tree clone Gorutuba. Their salts are fundamental to providing plant growth and maintenance *in vitro*. Despite the conversion of seeds to plants using vermiculite as a substrate, it was verified that they alone can not maintain the growth as a whole, evidencing low plant height *in vitro*.

Analyzing the results obtained in this study, it was observed that, at 30 days, when the BAP concentrations were increased, until the concentration of approximately 9.73 μmol L^{-1}, the characteristics analyzed were, in general, responsive; from that point, the BAP concentration may have caused phytotoxicity to the plant *in vitro*, inhibiting the conversion and its growth. This may have occurred because the microshoot already obtained the level of endogenous cytokinins required for development, making exogenous supplementation detrimental.

- **Analysis at 60 days**

According to the analysis of variance for the 60 days of evaluation, it was verified that there was interaction of substrate and concentration (SxC) effects only with the number of roots (NR). For the substrate (S), leaf number (LN), leaf length (LL), height (H) and number of roots (NR) were significant (Table 2). Analyzing the concentrations of BAP (C), it was verified that the height (H) and number of roots (NR) characteristics were significant.

In accordance with Table 3, it can be verified that, regardless of the BAP concentration used, the MS medium provided higher number of leaves (LN), leaf length (LL) and height (H) in relation to the use of vermiculite as substrate at 60 days of evaluation.

This may have been due to the fact that the culture medium used for the culture of plant cells, tissues and organs provide substances essential for tissue growth and largely control the *in vitro* development pattern (CALDAS et al., 1998). However, it is worth mentioning that the addition of phytoregulators is necessary in the supplementation of the mediums to supply the possible deficiencies of the endogenous levels of hormones in the explants used to establish the culture. Thus, the combination of an auxin and a cytokinin is used (SKOOG & MILLER, 1957).

According to Caldas et al. (1998) and Skoog and Miller (1957), it is possible to note that the composition of the MS medium is decisive in the growth of the seedlings because it provides the necessary nutrients and supplements it when the seed capsule no longer has nutrients or when it undo due to the growth of the seedling.

Regarding the number of roots (NR), it was verified that, when the MS medium was used, there was an increase according to the increase of the BAP concentration, until the concentration of 8.49 μmol L-1, in which observed the maximum of roots, 1.90 roots; from this concentration, an opposite effect was verified, that is, the increase of the BAP concentration inhibited the emission of roots. When vermiculite was used as substrate, the different BAP concentrations did not differ significantly, presenting 0.60 roots at all concentrations (Figure 2A). This data allows inferring that NAA supplementation in the capsule may have been efficient for root formation in the *in vitro* 'Prata-anã' banana plants clone Gorutuba. However, those that grew in vermiculite had no source of surplus nutrients, which may have impaired rooting, contrary to the plants cultivated in MS medium, since the nutrients provided could help induce the endogenous auxin metabolism, providing a higher emission of roots of plants *in vitro*. The cytokinin/auxin interaction may also have been a preponderant factor for root development, since low concentrations of cytokinin, together with higher concentrations of auxin, induce rhizogenesis, so concentrations above 8.88 μmol L^{-1} of BAP may have caused a phytotoxic effect for this characteristic.

Iqbal et al. (2013) observed rooting of the banana tree (*Musa sapientum* L) only at the concentration of 2 mg.L^{-1} IAA. This hormone has not been tested in our study, evidencing that different auxins promote different root development in banana tree, depending also on the genotype used.

Similar results were obtained by Diniz et al. (2003), who observed a reduction in the number of "macela" explants with roots in medium with high concentrations of BAP. This fact confirms the theory that high doses of cytokinins inhibit or delay root formation (BEN-JAACOV et al., 1991).

For the height characteristic (H) shown in Figure 2B, there was a quadratic effect where height increased according to the increase in BAP concentration to the concentration of 8.71 μmol L^{-1}, with a maximum height of 32.70 mm; from this concentration, there was a fall in plant height.

Oliveira et al. (2001), studying banana tree cultivar FHIA-01 (*Musa* sp., AAAB Group), in which the height of the seedlings during multiplication was inversely proportional to the BAP concentration, observed that the seedlings obtained in the BAP concentrations of 5.0 mg.L^{-1} and especially of 7.5 mg.L^{-1} presented a reduced size, always inferior to 1.5 cm. The inhibitory effect of BAP on shoot height was also reported in the *in vitro* of the "macela" (DINIZ et al., 2003), of vine (LUCAS et al., 2006), of Prunaceas (LEONTIEV-ORLOV et al., 2000) and peach trees (ROGALSKI, 2002).

These results may be related to the fact that this growth regulator is not responsible for promoting the lengthening of shoots. According to Oliveira et al. (2001), these small seedlings are not desirable in the micropropagation process because they need to

undergo an elongation before rooting, so that there are no high losses in acclimatization

As at 30 days, the 60-day analysis showed that the increase in BAP concentrations positively influenced the characteristics evaluated up to a certain concentration, from which there was an opposite effect, which can be explained by the endogenous level of cytokinin in the microshoots, causing excessive high concentrations to the plants *in vitro*.

With the results obtained, it can be considered that concentrations above 8.88 μmol L^{-1} of BAP are not indicated, with lower concentrations presenting better results and, consequently, savings in materials and reagents. The substrate type also influences *in vitro* plant growth, in which it can be seen that MS medium provides more nutrients and better conditions to plants *in vitro* than vermiculite, in which the different concentrations of BAP in the seed capsule do not differentiate in the results obtained. New studies needs to be done by testing new types of microshoots growth substrates, as well as exogenous nutrient supplementation for their development, together with the study with the MS medium that is already standardizing for the development of the Synthetic seed technique for 'Prata-anã Banana Plant clone Gorutuba.

TABLE 1– Summary of variance analysis for the variables: conversion (CONV), number of leaves (LN), leaf length (LL) and height (H) of *in vitro* 'Prata-anã' banana plants clone Gorutuba at 30 days of cultivation.

VF	DF	CONV	LN/1	LL	H
Substrate(S)	1	1440.0000*	0.0002^{ns}	219.9141**	1090.1448**
Concentration(C)	4	165.0000^{ns}	0.0121^{ns}	12.331450*	74.1738*
SxC	4	1265.0000**	0.0559*	16.875540*	66.9197*
Residue	30	266.6666	0.0188	4.2926	24.0967
CV(%)		**32.02**	**9.96**	**39.08**	**40.65**

**, * e ^{ns}: significant at 0.01, significant at 0.05 and not significant, respectively.
1 For analysis, the data were transformed into$(x + 1)^{0.5}$

TABLE 2 - Summary of variance analysis for the variables: leaf number (LN), leaf length (LL), height (H), number of roots (NR), root length (RL) and oxidation (OXI) of 'Prata-anã' banana tree clone Gorutuba at 60 days of cultivation.

VF	DF	LN/1	LL	H	NR/1	RL	OXI
Substrate(S)	1	0.2532*	448.3641**	1927.3768**	0.7336**	16.3072^{ns}	71.1022^{ns}
Concentration(C)	4	0.0287^{ns}	19.0667^{ns}	89.3452*	0.0586*	48.4470^{ns}	104.4422^{ns}
SxC	4	0.0342^{ns}	15.2097^{ns}	58.9580^{ns}	0.1071**	75.4732^{ns}	604.4522^{ns}
Residue	30	0.0343	7.8312	30.3373	0.0193	98.3666	288.8888
CV(%)		**11.77**	**24.04**	**25.24**	**9.97**	**46.81**	**30.72**

**, * e ^{ns}: significant at 0.01, significant at 0.05 and not significant, respectively.
1 For analysis, the data were transformed into $(x + 1)^{0.5}$

TABLE 3 - Leaves Number (LN), leaf length (LL) and height (H) of *in vitro* 'Prata-anã' banana tree clone Gorutuba at 60 days of culture medium MS and vermiculite.

Culture medium	LN	LL (mm)	H (mm)
Vermiculite	1.26 b	8.29 b	14.88 b
MS medium	1.78 a	14.99 a	28.78 a

*Means followed by the same letter do not differ from each other by the F test at the 5% level of significance.

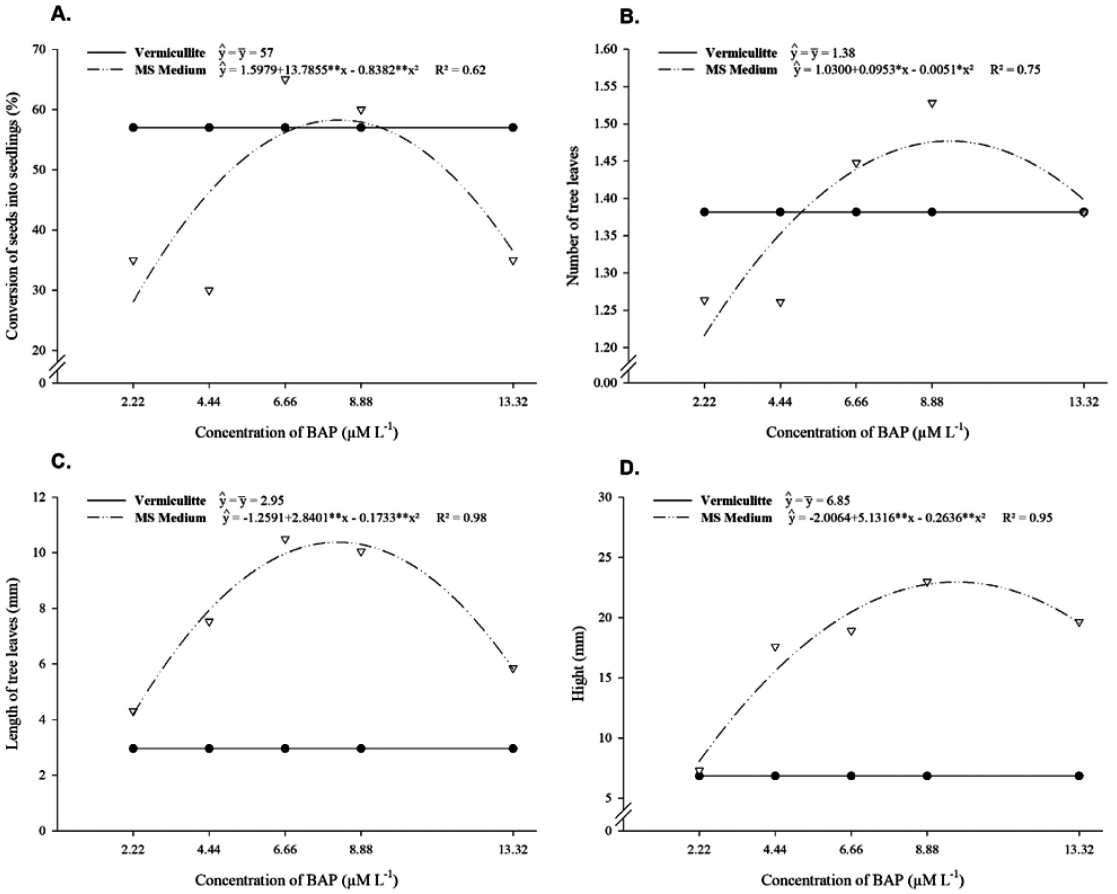

FIGURE 1- A) Conversion of synthetic seeds; B) Number of leaves (LN); C) Leaf length (LL) e D) Height of *in vitro* (H) to *in vitro* 'Prata-anã' banana plants clone Gorutuba (CONV) at 30 days of culture under different BAP doses and in cultures in MS medium and vermiculite.

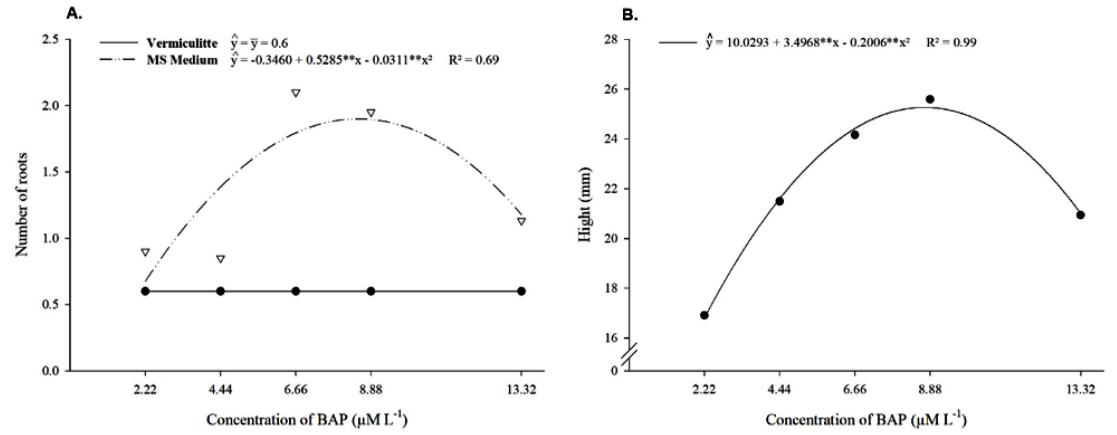

FIGURE 2- A) Number of roots (NR) of *in vitro* 'Prata-anã' banana tree clone Gorutuba at 60 days of culture under different doses of BAP and in cultures in MS medium and vermiculite e B) Height (H) of the *in vitro* 'Prata-anã' banana tree clone Gorutuba at 60 days of culture under different doses of BAP.

CONCLUSION

The MS culture medium is the most suitable substrate for the cultivation of the synthetic seed of 'Prata-anã' banana tree clone Gorutuba compared to vermiculite.

Concentrations of BAP up to 9.73 μmol L^{-1}, in the composition of the capsule, are not indicated for the conversion of microshoots and aerial formation of shoots; in addition, concentrations up to 8.88 μmol L^{-1} of BAP are most indicated in the *in vitro* plant rhizogenesis of 'Prata-anã' synthetic clone Gorutuba.

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