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ORIGINAL ARTICLE

# Profile of *in vitro* grown aloe plants using different concentrations of 6-benzylaminopurine

## *Perfil metabólico de babosa cultivada in vitro sob diferentes concentrações de 6-benzilaminopurina*

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**ABSTRACT:** Aloe plants have a great therapeutic potential, which is attributed to the several bioactive compounds found in their composition. Therefore, the objective of this work was to evaluate the profile of secondary metabolites and the morphophysiological characteristics of Aloe plants collected at different periods and submitted to different growing conditions. So, axillary buds were collected at two different periods and grown *in vitro* in MS culture medium containing naphthalene acetic acid (NAA) and 6-benzylaminopurine (BAP) under controlled conditions for 30 days. Then, the morphophysiological characterization and the analysis of the profile of metabolites produced by the plants were performed (HS-SPME / GC-MS techniques). The results showed that the BAP concentrations did not influence the morphophysiological characteristics of the plants. Almost all plants showed a 100% rooting percentage (RP) when grown *in vitro*, both with and without BAP in the nutrient medium. Only the plants subjected to BAP concentration of 3 mg $L^{-1}$ showed a lower RP. The rise in BAP concentrations also promotes a reduction in the total area of volatile organic compounds (VOCs) in plants. Some compounds (hexanal; 2-hexenal; 1-hexanol; 1-nonanol; 2-decanone; beta-ionone) showed a significant reduction (> 0.10%) in their concentration as the concentration of BAP was incremented in the medium cultivation. Nevertheless, the *in vitro* cultivated plants showed a greater total area and concentration of VOCs than the *in vivo* cultivated plants.

**RESUMO:** *A babosa possui grande potencial terapêutico, o qual é atribuído aos diversos compostos bioativos presentes em sua composição. Logo, o objetivo deste trabalho foi avaliar o perfil dos metabólitos secundários e as características morfofisiológicas em plantas de babosa coletadas em épocas distintas e submetidas a diferentes condições de cultivo. Para isso, gemas axilares foram coletas em 2 épocas distintas e cultivadas in vitro em meio de cultura MS contendo ácido naftalenoacético (ANA) e 6-benzilaminopurina (BAP) sob condições controladas durante 30 dias. Em seguida, foi realizada a caracterização morfofisiológica e a análise do perfil de metabolitos produzidos pelas plantas (técnicas HS-SPME/GC-MS). Constatou-se por meio dos resultados que as concentrações de BAP não influenciaram nas características morfofisiológicas das plantas. Quase todas as plantas apresentaram 100% de porcentagem de enraizamento (PR) quando cultivadas in vitro, tanto na ausência quanto na presença de BAP no meio nutritivo. Apenas as plantas submetidas a concentração de BAP de 3 mg $L^{-1}$ apresentaram PR inferior. O aumento das concentrações de BAP também promoveu a redução da área total dos compostos orgânicos voláteis (COV's) das plantas. Alguns compostos (hexanal; 2-hexenal; 1-hexanol; 1-nonanol; 2-decanona; beta-ionona) apresentaram diminuição significativa (>0,10%) da sua concentração à medida em que houve aumento da concentração de BAP ao meio de cultivo. Contudo, as plantas cultivadas in vitro apresentaram maior área total e concentração dos COV's do que as plantas cultivadas in vivo.*

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# 1 Introduction

Traditionally known as aloe, *Aloe vera* L. plants have been used in traditional medicine since ancient times due to their chemical composition and therapeutic properties. Several studies describe their antioxidant, antimicrobial, anti-inflammatory, immunomodulatory, and antineoplastic properties (Lorenzi & Matos, 2008; Andrade Junior *et al.*, 2020; Carvalho *et al.*, 2020).

Aloe has succulent green leaves in a lanceolate shape, with thorns at its ends, where a translucent gelatinous substance, called gel, is also found. This substance occupies most of the volume of its leaves (Parente *et al.*, 2013). The gel consists mainly of water (98.5%) and bioactive components. In addition to the polysaccharides that are found in greater abundance, chemical compounds such as soluble sugars, flavonoids, flavanols, enzymes, minerals, essential and non-essential amino acids, sterols, anthraquinones, glycoproteins, saponins, and vitamins have also been reported in its matrix (Surjushe *et al.*, 2008; El-Shemy *et al.*, 2010; Minjares-Fuentes & Femenia, 2019).

Several techniques have been used in order to increase the productivity of plant compounds of interest, including strain selection, metabolic engineering, type and concentration of growth regulators, optimization of cultivation conditions, and the use of elicitors (Souza *et al.*, 2018; Danial *et al.*, 2019; Camargo *et al.*, 2020).

Due to the pharmacological value and the increasing demand of the pharmaceutical industry for plants with genetic characteristics identical to the stock plants, with high phytosanitary and physiological quality, as well as an enhanced capacity for synthesis of secondary metabolites, through genetic improvement, the development of cropping systems that allow rational and sustainable exploitation are essential (Lima *et al.*, 2007). Thus, the *in vitro* propagation of aloe represents a plausible alternative for the commercial propagation of this species.

In this context, the objective of this work was to evaluate the profile of secondary metabolites and the morphophysiological characteristics of aloe plants collected at different periods and subjected to different growing conditions.

# 2 Material and methods

The experiments were conducted at the Laboratory of Biotechnology Applied to Health at the Maria Milza University Center (UNIMAM), located alongside the BR- 101 in the municipality of Governador Mangabeira, state of Bahia, Brazil. The evaluation of volatile compounds produced by plants from *in vitro* cultivation and through conventional cultivation methods took place at the Chemical Ecology Laboratory of the Agricultural Technological Center of the State of Bahia - CETAB in Salvador – Bahia, Brazil.

For the establishment of the *in vitro* plants, axillary buds from aloe plants grown under field conditions were used. The selected stock plants were collected in December 2018 (first experiment) and August 2019 (second experiment), in the same collection site.

After disinfestation, the buds were inoculated in test tubes containing the MS culture medium (Murashige & Skoog, 1962), supplemented with 0.2 mg $L^{-1}$ NAA, 3.0 mg $L^{-1}$BAP, 30 g $L^{-1}$ sucrose, 1 g $L^{-1}$ activated charcoal, and solidified with 7 g $L^{-1}$ agar. The pH of the culture medium was adjusted to 5.8 before autoclaving. The explants were cultivated *in vitro* under controlled conditions of temperature (25º ± 1ºC), photoperiod (16 h), and light intensity (30 μmol.$m^{-2}$. $s^{-1}$) for 30 days. Shoots of axillary buds were cultivated for two more subcultures at intervals of 30 days each, in glass flasks containing 20 mL of the culture medium with the same composition as described above, however, without the addition of regulators.

Then, the shoots from the stock plants collected in December 2018 (first experiment) and August 2019 (second experiment), with approximately 1.5 cm in size, were inoculated in glass flasks containing 20 mL of MS culture medium, supplemented with 0.2 mg $L^{-1}$ NAA, BAP at concentrations of 0.0; 1.0; 2.0 and 3.0 mg $L^{-1}$, 30 g $L^{-1}$ sucrose, 1 g $L^{-1}$ activated charcoal and solidified with 7 g $L^{-1}$ agar, with pH adjusted to 5.8. The explants were cultivated *in vitro* under controlled conditions, similar to the establishment step.

The experimental design used in this work was completely randomized, with four treatments, represented by different concentrations of BAP. Five replications were used and the experimental unit was represented by a flask containing three explants approximately 1.5 cm in size. After 30 days of cultivation, the plants were evaluated for their morphophysiological characteristics and profile of the produced volatile compounds.

The following characteristics were evaluated: plant height (PH), in cm; number of shoots (NS); number of green leaves (NGL); percentage of senescent leaves (PSL) and rooting percentage (RP).

The data resulting from the morphophysiological evaluation were subjected to analysis of variance (ANOVA) and the treatment means were compared by the test of Tukey at 5% probability, as it was not possible to adjust significant polynomial regression models and with high $R^2$ for BAP concentrations. Analyses were performed using the R statistical program using the ExpDes.pt package (Ferreira *et al.*, 2018).

The evaluation of the chemical composition of Aloe vera plants was carried out using the solid-phase microextraction technique (SPME), in which 0.2 g of leaves of the selected plant were weighed, both in the *in vitro* grown plants and in the plants grown in the conventional cultivation method. Plants from conventional cultivation were collected at the same periods and in the same place as the plants that were used for the *in vitro* cultivation experiment, maintaining the genetic pattern in both assays.

Volatile compounds (VOCs) were extracted using the solid-phase microextraction technique (HS-SPME) under the following conditions: CAR/PDMS/DVB extraction fiber; 0.2g of a leaf; extraction temperature of 80 ºC and

30 min of extraction.

The fiber was introduced into the injector of a gas chromatograph coupled to a mass spectrometer (GC-MS, Model QP2010 Plus, Shimadzu). Separation of the VOCs were performed on a DB-1MS column (30 m x 0.25 mm x 0.25 μm). Injections were performed in splitless mode, using helium-carrier gas with a flow of 0.7 mL/min and constant linear speed of 30.2 cm/s, injector temperature, and transfer line temperature of 260°C. The total running time was 46.0 min. Volatile compounds were identified through mass spectrometry and the Kovats Index.

## 3 Results and Discussion

The results obtained in this work showed that the addition of BAP in the *in vitro* cultivation of aloe plants from the collection carried out in December 2018 (first experiment) promoted a significant response only in the characteristic number of shoots (NS), showing the cytokinin activity in stimulating shoot formation (Table 1).

Although no influence was observed between the concentrations of BAP for PH and NGL, it is observed that the use of 1 mg $L^{-1}$ of BAP provided higher NS when compared to higher concentrations of BAP in the nutrient medium (Table 1), which corroborates the results obtained by Camargo *et al.* (2020) in a work carried out with strawberry. The authors observed a significant expression of NS in MS culture medium with 1.0 mg $L^{-1}$ BAP, which promoted a greater number of shoots, explaining the use of cytokinins in micropropagation, with the purpose of breaking apical dormancy, enhancing one of the objectives of the *in vitro* propagation.

**Table 1.** Mean values of plant height (PH), number of shoots (NS), and number of green leaves (NGL) of aloe plants cultivated *in vitro* using different BAP concentrations.

**Tabela 1.** Valores médios de altura de planta (AP), número de brotos (NB) e número de folhas verdes (NFV) de plantas de babosa cultivadas *in vitro* sob diferentes concentrações de BAP.

| BAP concentration (mg $L^{-1}$) | PH | NS | NGL |
|---|---|---|---|
| 0 | 4.11 a | 0.60 ab | 2.27 a |
| 1 | 3.20 a | 1.73 a | 2.60 a |
| 2 | 3.24 a | 0.53 ab | 2.53 a |
| 3 | 3.49 a | 0.33 b | 2.73 a |

Means followed by the same letters are not different from each other by the test of Tukey at 5% probability.

Médias seguidas pelas mesmas letras não diferem estatisticamente entre si pelo teste Tukey a 5% de probabilidade.

In evaluating the effect of BAP and NAA phytoregulators in aloe plants, Danial *et al.* (2019) also observed lower PH values as the concentrations of growth regulators were incremented. In relation to NGL, the authors mention a variation from 3.17 to 7.17, with greater expression in the concentration of 1.0 mg $L^{-1}$ of BAP, while in this work, a variation from 2.27 to 2. 73 occurred with greater expression in the concentration of 3.0 mg $L^{-1}$ of BAP. This difference can be attributed to the different genetic patterns among the stock plants used in the experiments.

The percentage of senescent leaves (PSL) shown by the plants was low, regardless of the concentration of BAP used in the experiment (Table 2). In relation to the rooting percentage (RP), the values are very satisfactory, with RP greater than 99% at all BAP concentrations used in the experiment (Table 3). In agreement with the experiment carried out by Molsaghi *et al.* (2014), in which they used Aloe vera plants in a culture medium enriched with BAP and observed high rooting rates (close to 100%).

**Table 2.** Percentage of senescent leaves (PSL) and rooting (RP) in aloe plants grown *in vitro* using different MS culture medium different from BAP.

**Tabela 2.** Porcentagem de folhas senescentes (PFS) e enraizamento (PR) em babosa cultivada *in vitro* em meio de cultura MS contendo diferentes concentrações de BAP.

| BAP concentration (mg $L^{-1}$) | Senescent leaves (%) | Rooting (%) |
|---|---|---|
| 0 | 6.66 * | 99.87 |
| 1 | 0.00 | 100.00 |
| 2 | 0.00 | 99.93 |
| 3 | 0.00 | 99.87 |

* The plants showed one senescent leaf.

* As plantas apresentaram uma folha senescente.

In the first experiment, 17 volatile organic compounds were identified. Out of which, 13 showed a higher percentage in *in vitro* grown plants than in *in vivo* grown plants (Table 3). The compounds that showed greater expression in plants cultivated *in vivo* were benzaldehyde (10.17%); 2-hexenal (6.18%), nonanal (4.78%), hexanal (4.16%), 1-octanol (2.45%), 1-pentanol (2.43%), 1-hexadecanol (2.34%) and 2-ethyl-1-hexanol (1.41%), which may reflect the absence of the phytoregulator. It was also possible to observe that the compounds 1-hexanol and beta-linalool were only present in *in vitro* grown plants, both in the presence and absence of BAP (Table 3).

The other compounds (hexanal; 2-hexanal; 1-hexanol; benzene-acetaldehyde; 2-ethyl-2-hexanol; 2-octenal; 1-octanol; beta-linalool; nonanal; 1-nonanol; decanal; 2-decanone; beta-ionone) showed higher concentration in *in vitro* grown plants (Table 4). However, it was observed that some compounds (benzene acetaldehyde; 1-nonanol; 2-decanone; beta-ionone, and 2,4-dimethyl-pentanal) exhibited a decrease (>0.10%) in their concentration when compared to the expression in plants grown *in vivo*, that is, without the addition of BAP.

**Table 3.** Volatile compounds analyzed through HS-SPME-CG-MS in aloe plants cultivated *in vitro* in MS culture medium containing different concentrations of BAP (0.0, 1.0, and 2.0 mg $L^{-1}$) and plants cultivated *in vivo*.
**Tabela 3.** Compostos voláteis, analisados por HS-SPME-CG-MS em plantas de babosa cultivadas *in vitro* em meio de cultura MS contendo diferentes concentrações de BAP (0,0; 1,0 e 2,0 mg $L^{-1}$) e plantas cultivadas *in vivo*.

| RT/min | Compound | 0.0 | 1.0 | 2.0 | *In vivo stock plant* |
|---|---|---|---|---|---|
| 5.032 | 1-pentanol | 0.29 | 0.70 | 0.95 | 2.43 |
| 5.834 | Hexanal | 4.25 | 5.66 | 6.46 | 4.16 |
| 7.449 | 2-hexenal | 20.84 | 24.09 | 30.56 | 6.18 |
| 8.021 | 1-hexanol | 8.55 | 9.28 | 9.09 | 0.00 |
| 11.333 | Benzaldehyde | 3.35 | 3.86 | 3.70 | 10.17 |
| 12.125 | benzene acetaldehyde | 1.10 | 1.01 | 0.94 | 0.72 |
| 14.087 | 2-ethyl-1-hexanol | 2.24 | 7.07 | 1.78 | 1.41 |
| 15.220 | 2-octenal | 0.79 | 0,84 | 0.64 | 0.43 |
| 15.742 | 1-octanol | 4.56 | 5,74 | 6.17 | 2.45 |
| 16.865 | beta-linalool | 0.27 | 0,07 | 0.33 | 0.00 |
| 17.046 | Nonanal | 9.22 | 10,98 | 9.76 | 4.78 |
| 19.730 | 1-nonanol | 4.82 | 2,19 | 1.42 | 0.84 |
| 20.181 | Decanal | 0.49 | 0,22 | 0.96 | 0.11 |
| 29.539 | 2-decanone | 0.27 | 0,18 | 0.14 | 0.08 |
| 32.955 | beta-ionone | 1.41 | 0,79 | 0.47 | 0.67 |
| 46.571 | 2,4-dimethyl-pentanal | 0.30 | 0,08 | 0.03 | 0.93 |
| 47.859 | 1-hexadecanol | 0.00 | 0,98 | 0.69 | 2.34 |

The analysis of the total area of volatile organic compounds (VOCs) in the *in vitro* grown aloe plants promoted a significant increase in the production of VOCs when compared to *in vivo* cultivation, but the use of the BAP phytoregulator did not provide a significant increase in the production of volatile secondary metabolites, as well as the increase in the concentrations of BAP proposed in this study showed an antagonistic action to the production of VOCs, as observed in Figure 1.

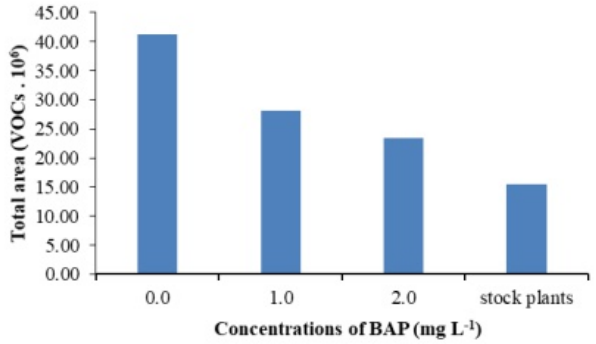


**Figure 1.** Total area of volatile compounds observed in aloe plants cultivated *in vitro* in MS culture medium containing different concentrations of BAP (0.0, 1.0, and 2.0 mg $L^{-1}$) and from *in vivo* grown plants (stock plants).
**Figura 1.** Área total dos compostos voláteis observados nas plantas de babosa cultivadas *in vitro* em meio de cultura MS contendo diferentes concentrações de BAP (0,0; 1,0 e 2,0 mg $L^{-1}$) e das plantas cultivadas *in vivo* (Matriz).

Regarding the pharmacological activity of the compounds found in this work, the properties of benzaldehyde are highlighted, as they are reported in the literature as antimicrobial, anticonvulsant, analgesic, and anti-inflammatory. Another compound that deserves attention is beta-ionone, due to its anticancer, liver, and antimicrobial properties (Gomes-Carneiro *et al.*, 2003).

Phenolic compounds, such as beta-linalool, have antibacterial and antifungal characteristics, showing antibiotic, free radical reducing- and enzymatic inhibiting-capacity (Juiz *et al.*, 2016; Ogunwanda *et al.*, 2018). According to El-shemy *et al.* (2010), phenolic compounds feature prominently in the composition of aloe.

The result of the morphophysiological analysis with the plants from the second experiment, collected in August 2019, revealed a similar result to the first experimental test for the PH and NGL, which did not show significant differences in the concentrations of BAP used in the nutrient medium, differing from the first experiment only in relation to the number of shoots (Table 4).

**Table 4.** Mean values of plant height (PH), number of shoots (NS), and number of green leaves (NGL) of *in vitro*-grown aloe plants in MS culture medium containing different concentrations of BAP.
**Tabela 4.** Valores médios de altura de planta (AP), número de brotos (NB) e número de folhas verdes (NFV) de plantas de babosa cultivadas *in vitro* em meio de cultura MS contendo diferentes concentrações de BAP.

| BAP concentration (mg $L^{-1}$) | PH | NS | NGL |
|---|---|---|---|
| 0 | 3.18 a | 0.50 a | 5.67 a |
| 1 | 3.03 a | 0.50 a | 5.75 a |
| 2 | 2.92 a | 0.25 a | 4.75 a |
| 3 | 3.03 a | 0.42 a | 5.00 a |

Means followed by the same letter are not statistically different from each other by the F test at 5% probability.
Médias seguidas pelas mesmas letras não diferem estatisticamente entre si pelo teste F a 5% de probabilidade.

Pérez-Alonso *et al.*, (2015) also evaluated *in vitro* aloe plants at different concentrations of BAP and observed divergent results from those obtained in the present study. The authors inferred that the absence or high concentrations of the phytoregulator tends to negatively interfere with the morphophysiological characteristics of the seedlings.

Regarding the characteristic percentage of senescent leaves (PSL), the plants evaluated in this experiment showed higher values than the plants evaluated in the first experiment, at all concentrations of BAP (Table 5), showing the likely influence that the external environment has on the genetic pattern of plants. To establish a protocol for aloe micropropagation, Debiasi *et al.* (2007) observed different morphogenetic patterns of explants, regardless of the use and concentration of plant growth regulators supplemented to the culture medium. According to the authors, the variation in the responses of *A. vera* explants demonstrates the cellular sensitivity to

the action of plant regulators.

**Table 5.** Percentage of senescent leaves (PSL) and rooting (RP) in aloe plants cultivated *in vitro* in MS medium containing different concentrations of BAP.
**Tabela 5.** Porcentagem de folhas senescentes (PFS) e de enraizamento (PR) em plantas de babosa cultivadas *in vitro* em meio MS contendo diferentes concentrações de BAP.

| BAP concentrations (mg $L^{-1}$) | Senescent leaves (%) | Rooting (%) |
|---|---|---|
| 0 | 50.00 * | 100.00 |
| 1 | 33.33 * | 100.00 |
| 2 | 25.00 ** | 100.00 |
| 3 | 66.67 ** | 91.67 |

* The plants showed one senescent leaf. ** The plants showed one or two senescent leaves.
* As plantas apresentaram uma folha senescente. ** As plantas apresentaram uma ou duas folhas senescentes.

As in the first experiment, almost all of the plants showed a 100% rooting percentage (RP) when cultivated *in vitro*, both in the absence and in the presence of BAP in the nutrient medium. Only plants subjected to a BAP concentration of 3 mg $L^{-1}$ had a lower RP. High rooting rates are important for the acclimatization phase of the plants, as well as the addition of auxin (NAA) to the growing medium to stimulate rooting.

In the second experiment, 17 VOCs were also identified, of which 10 showed more expression. in plants grown *in vitro*, and seven showed a higher percentage in the *in vivo* matrix (Table 6).

It could have been observed that as in the first experiment, the compounds 1-hexanol and beta-linalool expressed themselves only in *in vitro* grown plants, with or without the addition of BAP in the nutrient medium (Table 6).

Most of the organic compounds found in this work showed greater expression in *in vitro* grown plants (Table 8). However, it was observed that some compounds (hexanal; 2-hexenal; 1-hexanol; 1-nonanol; 2-decanone; beta-ionone) showed a significant decrease (> 0.10%) in their concentration when BAP was added to the growing medium.

As in the first experiment, two compounds in this experiment should be observed for their pharmacological properties, beta-ionone, and beta-linalool, which showed low expression in the *in vivo* stock plants. The therapeutic activities of these substances are of great importance for the health area, as reported in the literature and described in this work, which explains the *in vitro* propagation of *A. vera* with the purpose to increase the production of these metabolites.

Another compound that deserves attention is benzaldehyde, which showed a high percentage in *in vivo* cultivation. Benzaldehyde has very important pharmacological properties, such as antibacterial, antifungal, and antitumor activities (Jang *et al.*, 2014; Ullah *et al.*, 2015). In addition to the pharmaceutical industry, this compound is used as a synthetic precursor or reagent in the chemical and food industry (Leite, 2020).

**Table 6.** Volatile compounds analyzed through HS-SPME-CG-MS *in vitro*-grown aloe plants in MS culture medium containing different concentrations of BAP (0.0, 1.0, 2.0, and 3.0 mg $L^{-1}$) and *in vivo* grown plants.
**Tabela 6.** Compostos voláteis, analisados por HS-SPME-CG-MS em plantas de babosa cultivadas *in vitro* em meio de cultura MS contendo diferentes concentrações de BAP (0,0; 1,0, 2,0 e 3,0 mg $L^{-1}$) e plantas cultivadas *in vivo*.

| RT/min | Compound | 0.0 | 1.0 | 2.0 | 3.0 | *In vivo* stock plant |
|---|---|---|---|---|---|---|
| 5,032 | 1-pentanol | 0.84 | 1.27 | 1.96 | 2.83 | 3.33 |
| 5,834 | Hexanal | 5.48 | 2.26 | 2.31 | 1.95 | 4.96 |
| 7,449 | 2-hexenal | 39.25 | 21.43 | 11.42 | 5.38 | 3.11 |
| 8,021 | 1-hexanol | 13.20 | 6.76 | 8.32 | 8.87 | 0.00 |
| 11,333 | Benzaldehyde | 2.32 | 1.52 | 1.94 | 2.90 | 15.87 |
| 12,125 | benzene acetaldehyde | 0.44 | 0.28 | 0.29 | 0.59 | 0.80 |
| 14,087 | 2-etil-1-hexanol | 4.97 | 2.70 | 2.20 | 5.93 | 1.13 |
| 15.220 | 2-octenal | 0.49 | 0.32 | 0.37 | 0.40 | 0.35 |
| 15.742 | 1-octanol | 0.71 | 0.67 | 0.73 | 0.89 | 0.89 |
| 16.865 | beta-linalol | 0.44 | 0.36 | 0.87 | 0.43 | 0.00 |
| 17.046 | Nonanal | 2.62 | 2.27 | 2.90 | 2.35 | 6.31 |
| 19.730 | 1-nonanol | 2.64 | 1.17 | 1.13 | 0.69 | 0.53 |
| 20.181 | Decanal | 0.02 | 0.00 | 0.02 | 0.05 | 0.06 |
| 29.539 | 2-decanone | 0.20 | 0.06 | 0.15 | 0.12 | 0.10 |
| 32.955 | beta-ionone | 1.41 | 1.19 | 0.92 | 1.22 | 0.80 |
| 46.571 | 2,4-dimethyl-pentanal | 0.14 | 0.11 | 0.15 | 0.16 | 0.61 |
| 47.859 | 1-hexadecanol | 0.80 | 1.33 | 1.78 | 1.40 | 1.97 |

Carvalho *et al.* (2020) carried out the scientific and technological monitoring of aloe vera in order to map its progress in the scientific and technological field and observed several activities with pharmacological purposes shown by *Aloe vera*, which corroborates experiments that show the high potential of its properties in the treatment of diseases, especially concerning to healing, antioxidant and antimicrobial actions.

The results showed a variation in the composition of volatile compounds in both cultivation conditions (*in vitr*o and *in vivo*) as a function of the time when the plants were collected. A similar result was reported by Cota *et al.* (2019), who stated that the period of the year that the plant is collected is a very important factor in the composition of secondary compounds, since the amount, as well as the nature of the active constituents, vary over the year.

Similar to what was observed in the first experiment, concerning the total area of the VOCs, it is possible to highlight that the use of a phytoregulator did not promote a significant increase in the production of secondary metabolites. On the contrary, the increase in BAP concentrations caused a reduction in the total area of the VOCs (Figure 2). However, the total area of VOCs in *in vitro* grown plants was greater than the area observed in the *in vivo* stock plants.

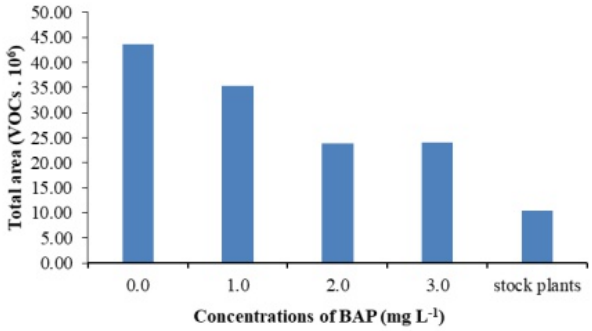


**Figure 2.** Total area of volatile compounds observed in *in vitro*-grown aloe plants in MS culture medium containing different concentrations of BAP (0.0; 1.0; 2.0 and 3.0 mg $L^{-1}$) and from *in vivo* grown plants (stock plants).
**Figura 2.** Área total dos compostos voláteis observados nas plantas de babosa cultivadas *in vitro* em meio de cultura MS contendo diferentes concentrações de BAP (0,0; 1,0; 2,0 e 3,0 mg $L^{-1}$) e das plantas cultivadas *in vivo* (Matriz).

## 4 Conclusion

The different concentrations of BAP in the MS culture medium did not promote any significant results that would recommend the use of this growth regulator in studies related to the *in vitro* cultivation of aloe, aiming at the large-scale production of seedlings.

However, based on this work, it was clear that aloe plants cultivated in vitro present higher production of volatile compounds, and consequently, higher concentrations of principles with pharmacological activities when compared to plants grown in field conditions.

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