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Effect of fertilisation with urea on development in the ornamental bromeliad *Aechmea fasciata*[1]

Efeito da fertilização com ureia no desenvolvimento da bromélia ornamental *Aechmea fasciata*

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ABSTRACT - The aim of this study was to evaluate the effect of urea fertiliser on the bromeliad, *Aechmea fasciata*. Plants were submitted to weekly treatments with a solution of 0.0, 0.5, 1.0, 1.5 or 2.0 g urea L^{-1}. After 210 days of the experiment, biometric variables of growth and development of the root and shoot systems were analysed, together with foliar levels of chlorophyll *a* and *b*, and of carotenoids. The experimental design was of randomised blocks with four replications; eight plants were used per plot. The data were submitted to regression analysis. The highest values for chlorophyll *a* and carotenoid content were obtained at a concentration of 1.23 g L^{-1} urea, and for chlorophyll *b* at a concentration 1.75 g L^{-1} urea. Plants of *A. fasciata* submitted to a solution with no nitrogen showed marked chlorosis of the leaves and reduced growth and development. The mean value for the maximum levels of photosynthetic pigments was 1.40 g L^{-1} urea, and this can be used as an indicator of the nutritional status of nitrogen in the species. From the results, an application of 1.46 g L^{-1} urea to the tank in *A. fasciata* is recommended, as this gave the greatest accumulation of total dry weight and an increase in the number of leaves.

Key words: Bromeliaceae. Nitrogen. Mineral nutrition. Chlorophyll.

RESUMO - Objetivou-se avaliar o efeito da fertilização com ureia na bromélia *Aechmea fasciata*. As plantas foram submetidas aos tratamentos com as soluções 0,0; 0,5; 1,0; 1,5 e 2,0 g ureia L^{-1}, aplicadas semanalmente. Após 210 dias de experimentação, foram analisadas as variáveis biométricas de crescimento e desenvolvimento dos sistemas radicular e aéreo, e os teores foliares das clorofilas *a* e *b*, e carotenoides. O delineamento experimental foi em blocos casualizados com quatro repetições, oito plantas por parcela, sendo os dados submetidos à análise de regressão. Os maiores valores de teores de clorofila *a* e carotenoides foram obtidos na concentração 1,23 g L^{-1} de ureia e de clorofila *b* na concentração 1,75 g L^{-1} de ureia. As plantas submetidas à solução com omissão de nitrogênio apresentaram acentuada clorose das folhas e diminuição do crescimento e desenvolvimento de *A. fasciata*. A média dos teores de máximos dos pigmentos fotossintetizantes foi de 1,40 g L^{-1} ureia, podendo ser utilizada como indicadora do estado nutricional de nitrogênio na espécie. Os resultados obtidos permitem recomendar a aplicação no tanque de 1,46 g L^{-1} de ureia em *A. fasciata*, uma vez que possibilitou o maior acúmulo de massa de matéria seca total e aumento no número de folhas.

Palavras-chave: Bromeliaceae. Nitrogênio. Nutrição mineral. Clorofila.

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INTRODUCTION

There is a lack of information on the production of bromeliads for commercial purposes which would promote an increase in productivity and the maintenance of plant health. It is known that balanced fertilisation produces better quality plants, with resistance to disease and pests (KANASHIRO, 2007). Bromeliads are characterised by having leaves arranged in a rosette to form a tank, which accumulates rainwater, particulate matter and organic compounds that are absorbed by trichomes present on the leaf surface (BENZING, 1990).

Studies into nitrogen nutrition in ornamental bromeliads have been carried out, especially by *in vitro* experiment, demonstrating that tank-forming epiphytic bromeliads are able to absorb and assimilate urea efficiently (TAKAHASHI; CECCANTINI; MERCIER, 2007). Epiphytic bromeliads show a preference for organic N, while terrestrial bromeliads prefer inorganic N. Furthermore, bromeliads are exposed to constant nutritional deficiency in their habitat, and have adapted to increase the efficiency of N absorption and assimilation (ENDRES; MERCIER, 2001). The leaf base in the bromeliad, which is the principle region in contact with nutrients, takes on the role of the roots in nitrogen absorption, nitrate reduction and the hydrolysis of urea, since the main function of the roots is to anchor the bromeliad to the host plant (TAKAHASHI; MERCIER, 2011).

Urea is the most used nitrogen fertiliser in Brazil, and when applied to the soil, may undergo hydrolysis through the action of the urease enzyme, which converts r-NH_2 into NH_4^+ so that it may be absorbed by the plant (VILLALBA; OTTO; TRIVELIN, 2014). In nature, urea when supplied in high concentrations by animals inhabiting the bromeliad tanks is an important source of N for the bromeliads (RODRIGUES *et al*., 2016), and is the preferred source of nitrogen for *Vriesea gigantea*, possibly due to the simultaneous uptake of carbon and nitrogen, which are limiting in its natural habitat, since the assimilation of urea depends on the previous hydrolysis of NH_4^+ and CO_2, a reaction which is catalysed by urease (CAMBUÍ; GASPAR; MERCIER, 2009).

The species, *Aechmea fasciata*, popularly known as caraguatá or gravatá, is native to Brazil, being endemic to warm environments and displaying an epiphytic growth habit. This species was one of the first bromeliads to be used as an ornamental plant due to its rusticity and the ornamental character of its inflorescences (SMITH; DOWNS, 1979). *A. fasciata* is one of the most cultivated and commercialised bromeliads in Brazil; management and reproduction are simple, and it is easily grown *in vitro* (TAVARES *et al.*, 2008). This gives a commercial advantage, since this technique allows the rapid multiplication of genetically identical plant material from selected matrices (MAYER *et al.*, 2008), enabling pest-free propagation, and accelerating conventional propagation methods to provide high-quality seedlings in sufficient number to meet commercial demand throughout the year (SCHIAVINATO *et al.*, 2008). The average production cycle of *A. fasciata* grown from seeds is around 2 years and 2 months (SANCHES, 2009); therefore, studies which aim to optimise growth in bromeliads are extremely important, reducing the period of cultivation and the production costs involved.

The aim of the present study was to evaluate the effects of different concentrations of urea on growth and development in the ornamental bromeliad, *Aechmea fasciata*.

MATERIAL AND METHODS

The experiment was carried out at the Centre for Ornamental Plant Research of the Botanical Institute in São Paulo, Brazil. The plants were grown in a greenhouse with a clear plastic film polyethylene covering, at a mean daily irradiance of 170 μmol m^{-2} s^{-1} (SKP 200, Skye Instruments Ltda.) and a mean daily temperature of 27.5 °C. Irrigation was by micro-sprinkler (NaanDanJain®, Modular, with a flow rate of 141 L h^{-1}) in two daily 15-minute applications. Analysis of the irrigation water showed: pH - 7.7, SAR (sodium absorption ratio) - 0.23, EC - 0.080 dS m^{-1} and ions (in $mmol_c$ L^{-1}) K^+ - 0.07, Ca^{++} - 0.320, Mg^{++} - 0.060, Cl^- - 0.960, Na^+ - 0.100, CO_3^{-2} - 0.000, and HCO_3^- - 0.420.

Plants of *Aechmea fasciata* were used, with a leaf length of 20.8 (±3.7) cm, stem diameter of 7.8 (±1.8) mm, root length of 9.0 (±2.6) cm, 8 (±1) leaves, total fresh weight of 5.6 (±1.0) g and total dry weight of 0.53 (± 0.11) g (n = 20). The plants were transplanted into 0.45 L pots of black polyethylene containing a mixture of composted pine bark and carbonised rice husks (1:1) as substrate.

The treatments consisted of the application of 50 mL of a solution with 0.0, 0.5, 1.0, 1.5 or 2.0 g L^{-1} urea (0.0, 16.6, 33.2, 50.0 or 66.6 mM N), applied weekly into the plant tank, with the excess solution poured onto the substrate. For all treatments, a supplement of 50 mL of 0:30:20 (NPK) solution at a concentration of 0.25 mL L^{-1} was given after the treatments with urea were applied to the substrate.

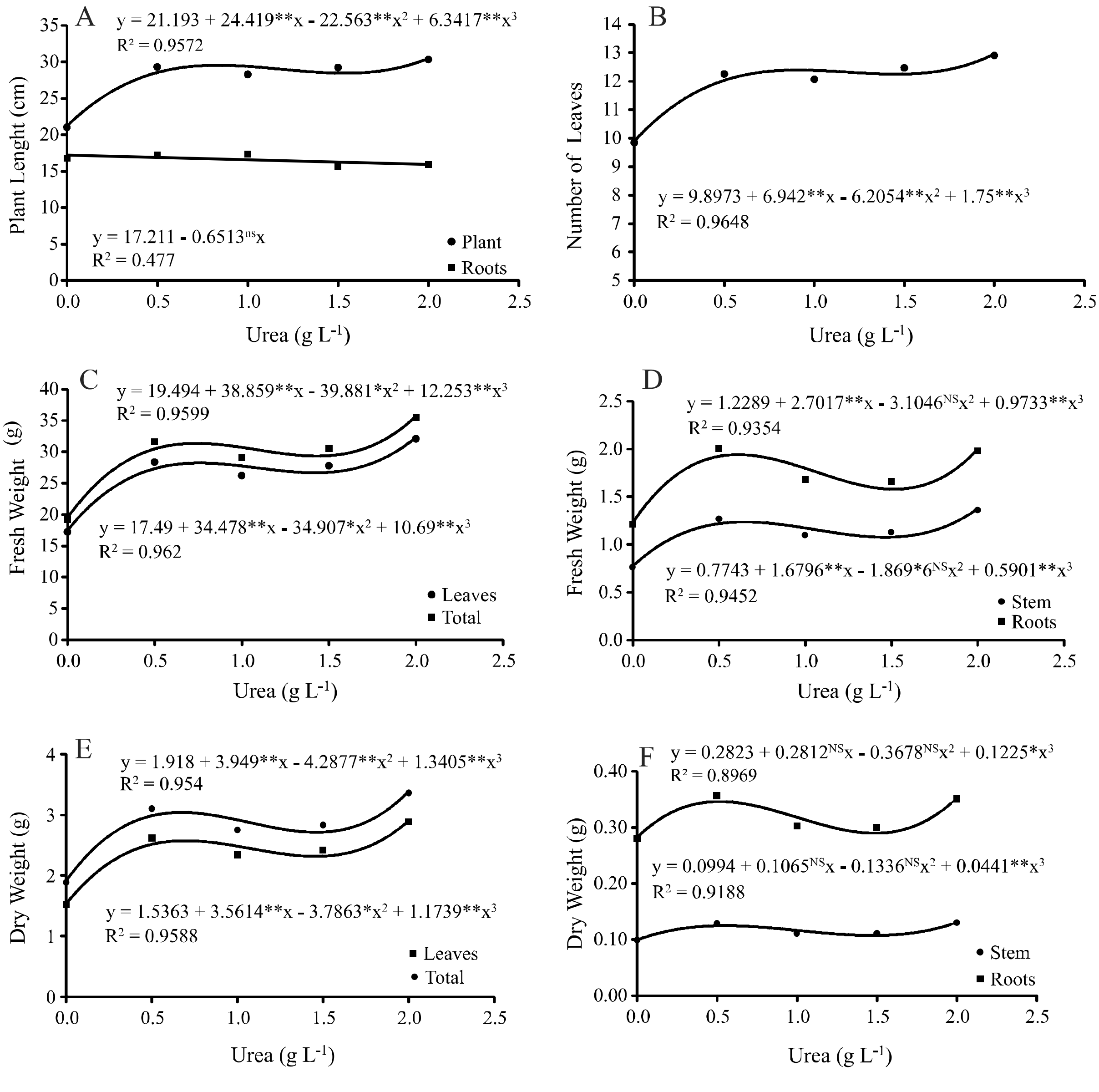
After 210 days of the experiment, the plants were divided into leaves, roots and stem, and evaluated for shoot length (from the substrate surface to the upper extremity of the leaves), root-system length, number of leaves, fresh and dry weight of the leaves, roots and stem, and total fresh and dry weight. The biochemical variables under analysis were the foliar levels of chlorophyll *a* and *b*, and carotenoids in the apical portion of the fourth expanded leaf, as per Carvalho *et al*. (2013).

The experimental design was of randomised blocks with four replications, containing eight vessels per lot. All the plants were evaluated for the biometric variables, and four plants per lot for analysis of photosynthetic pigments. Data were submitted to analysis of variance and regression using the SISVAR statistical software.

RESULTS AND DISCUSSION

The biometric variables (Figures 1A to 1F) displayed a positive response to the increases in urea concentration applied to the plant tank, and better fit the cubic regressions. The variables, plant length and number of leaves showed that for the range being considered (0 to 2 g L^{-1} urea), the mean values for the variables increased with increases in the urea concentration, reaching the greatest value at concentrations of 1.54 and 1.46 g L^{-1} urea respectively (Figures 1A and 1B). An increase of approximately 30% in the values for leaf number and leaf

Figure 1 - Adjusted mean values and functions for the variables: (A) - plant and root length, (B) - number of leaves, (C) - leaf and total fresh weight, (D) - stem and root fresh weight, (E) - leaf and total dry weight, and (F) - stem and root dry weight in *A. fasciata*, at doses of 0.0, 0.5, 1.0, 1.5 or 2.0 g L^{-1} urea (p<0.01 = **, p<0.05 = *; NS = not significant)



length was found by Santos *et al*. (2015) in *Nidularium fulgens*, for increases in urea concentration (500 mg N dm^{-3}). Ferreira *et al*. (2007) saw an increase of 10% in plant height in *Neoregelia cruenta* submitted to a treatment of urea at 2%, when compared to the control treatment.

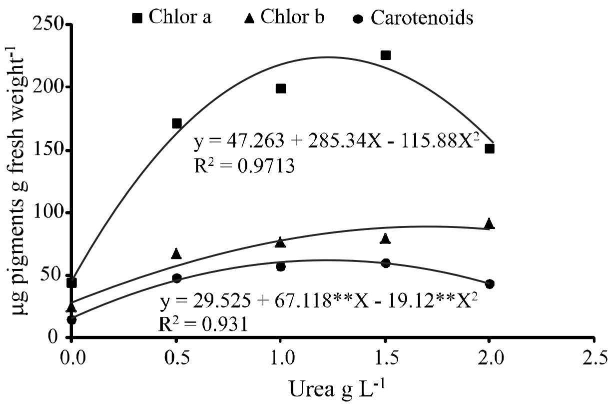
The increase in urea concentration had no significant effect on root-system length in *A. fasciata* (Figure 1A). Compounds derived from urea, such as N, N'-diphenylurea and TDZ stimulate the biosynthesis of cytokinins, a hormone that inhibits root growth. Nitrogen sources therefore play an important role in biomass partitioning (root to shoot ratio), mediated by the endogenous hormone levels in the foliar tissue of *Vriesea philippocoburgii* and *Tillandsia pohliana* (MERCIER *et al*., 1997). The lesser root development can also be explained by the epiphytic habit of the species, where the roots have the main function of anchoring the plant to the substrate, with nutrients being absorbed by trichomes present on the leaf surface (REITZ, 1983). Plants of *Laelia purpurata*, an epiphytic orchid, displayed changes in the morphological characteristics of the roots, such as a reduction in the number of root cell layers, and thickness of the velomen, exodermis and vascular cylinder, which were less functional when cultivated in the presence of urea (SILVA JÚNIOR *et al*., 2013). Plants of *Neoregelia cruenta* took 300 days to show the effects of urea application, which according to Ferreira *et al*. (2007), is due to the time required by the seedlings to use the nitrogen to synthesise amino acids and proteins; as the bromeliad seedlings reach full vegetative development, they begin to respond better, and in increasing linear fashion, to the urea fertilisation, nitrogen being the principal promoter of vegetative growth.

The fresh weight of the leaves (Figure 1C), stem (Figure 1D) and roots (Figure 1D), and total fresh weight (Figure 1C) increased with the increases in urea concentration, reaching their greatest values at concentrations of 1.42, 1.46, 1.52 and 1.43 g L^{-1} urea respectively. The values for leaf (Figure 1E), stem (Figure 1F), root (Figure 1F) and total dry weight (Figure 1E) showed the highest values respectively at concentrations of 1.46, 1.47, 1.49 and 1.46 g L^{-1} urea. Production of the urease enzyme near the site of urea absorption or the site of its release in the bromeliad tank, enables fast and efficient use of the nitrogen, showing how epiphytic bromeliads develop well in environments of limited nitrogen availability (CAMBUÍ; GASPAR; MERCIER, 2009). Urea increased the foliar levels of CO_2 in *Vriesea gigantea* due to the breakdown of the molecule in regions near the chloroplasts, vacuoles and walls, indicating that the carbon dioxide is being used, and that urea can be another source of carbon in bromeliads (MIOTO *et al*., 2015). The effect of increasing urea concentrations on Bromeliaceae was also seen in shoots of *Tillandsia pohliana* and *Vrisea philippocoburgii*, as the urea altered the balance between cytokinins and auxins, stimulating leaf development (MERCIER *et al*., 1997).

The lowest values for biomass for all variables were seen in the treatment with no urea in the nutrient solution. Cultivating plants under a nitrogen deficiency results in a reduction in mesophyll conductance and impairs the entry of CO_2 to the carboxylation sites (CRUZ *et al*., 2007), consequently decreasing the photosynthetic capacity of the plant and the assimilation and production of vegetal matter. Nitrogen is the limiting element in the pineapple (Bromeliaceae); besides reducing vegetative growth in the plant, it produces smaller fruit with a whitish pulp and chlorosis of the leaves of the crown (RAMOS *et al*., 2009).

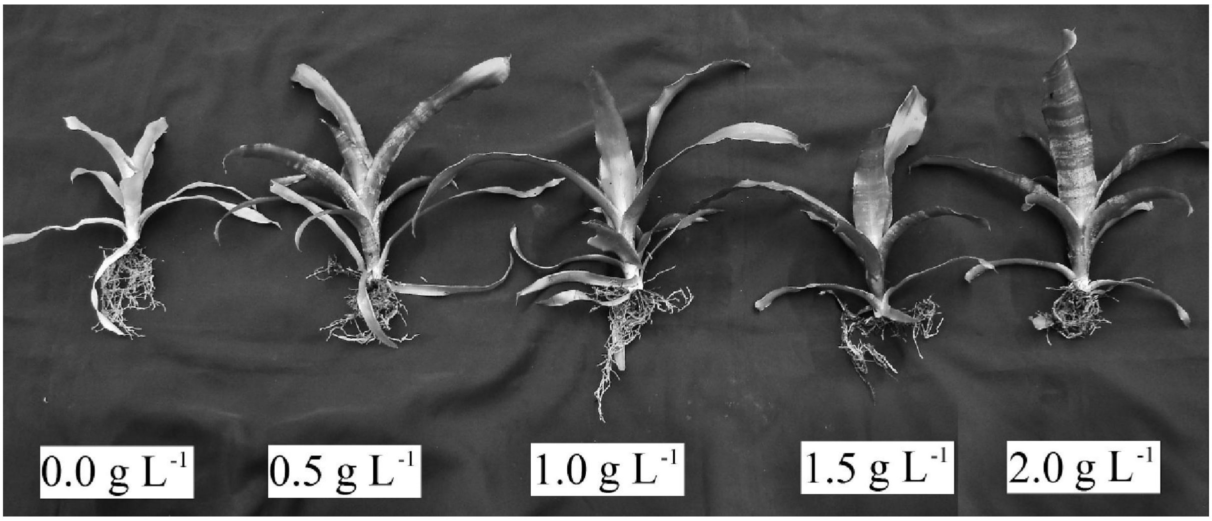
Regression analysis of the levels of photosynthetic pigments in leaves of *A. fasciata* (chlorophyll *a* and *b*, and carotenoids) was significant for the quadratic equation in all variables. *A. fasciata* displayed the greatest levels of chlorophyll *a* at a concentration of 1.23 g L^{-1} urea, chlorophyll *b* at a concentration of 1.76 g L^{-1} urea, and carotenoid pigments at a concentration of 1.21 g L^{-1} urea (Figure 2). The average maximum productivity of photosynthetic pigments was at 1.40 g L^{-1} urea, showing high correlation with dry weight production, which reached maximum productivity at a mean value of 1.43 g L^{-1} urea.

Figure 2 - Adjusted mean values and functions for the variables chlorophyll *a* and *b*, and carotenoids, in the leaves of *A. fasciata*, at doses of 0.0, 0.5, 1.0, 1.5 or 2.0 g L-^{1} urea (p<0.01 = **)



The plants grown in the absence of urea presented leaves with a yellowish colouration and less shoot growth, typical responses to nitrogen deficiency (Figure 3). The phenotypic aspects of plants cultivated in solution in the

Figure 3 - Plants of *A. fasciata* treated with different concentrations of urea (0.0, 0.5, 1.0, 1.5 or 2.0 g L^{-1})



absence of urea are the same as those found by Oliveira, Araújo and Dutra (1996), who stated that plants with nitrogen deficiency appeared atrophied, with leaves showing a pale green to yellow colouration beginning with the older leaves, and which is related to the presence of nitrogen in the molecular structure of chlorophyll. However, epiphytes show a low capacity for growth in response to the supply of nutrients in solution, and this is related to nutritional restrictions found under natural conditions (BENZING, 1983). High doses of nitrogen can cause phytotoxicity by the release of ammonium during the process of urea hydrolysis, which raises the ammonium levels of the soil solution (SANTOS *et al*., 2011); however no signs of phytotoxicity from urea were seen in the plants of *A fasciata*.

In the present study, the proportional increase seen in pigment levels for the variables chlorophyll *a*, chlorophyll *b* and carotenoids in *A. fasciata* treated with urea in increasing concentrations, was also seen in the pineapple by Leonardo *et al*. (2013). The chlorophyll content of the leaves can be used to evaluate the nutritional level of nitrogen (N) in plants, as the amount of pigment has a positive correlation with N levels in the plant (VIANA *et al*., 2008).

CONCLUSION

A concentration of 1.46 g L^{-1} urea affords greater dry weight production and higher foliar levels of chlorophyll and carotenoids in *Aechmea fasciata*. The absence of nitrogen is detrimental to the development of the species, and results in reduced growth and marked chlorosis of the leaves.

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