

Nitrogen Doses And Humic Substances In Custard Apple Nutrition

ABSTRACT

Custard apple production (*Annona squamosa* L.) in the Brazilian Northeast occurs throughout the year. Its management involves pruning, irrigation, and proper nutrition. The objective of the present work was to verify the influence of different doses of nitrogen and the use of humic substances on soil chemical attributes and tree custard apple nutrition. The experiment was conducted in a commercial orchard in the municipality of Anagé, Southwest region of the state of Bahia, in a randomized block design in a 4 x 2 factorial scheme, four nitrogen doses (0, 284, 568 and 852 g per plant, urea) with and without the application of humic substances (Ks100), with 4 replicates. Soil chemical characteristics, macro and micronutrient contents were evaluated in the leaves of the custard apple tree. Nitrogen rates reduced soil pH and the availability of calcium, magnesium and boron. The application of humic substances increased the content of potassium in leaves. Under the conditions studied, high nitrogen doses influence soil fertility and the application of humic substances does not improve the absorption of most of the nutrients, except potassium, and it did not influence the chemical attributes of the soil.

Keywords: *Annona squamosa*, fertility, anonaceous, urea, fertilization.

1. INTRODUCTION

Bahia is the largest producer of Custard apple (*Annona squamosa* L.) in Brazil, with an estimated area of 3575 ha in 2012, followed by Alagoas, Pernambuco, São Paulo and Ceará [1]. The socioeconomic importance of the crop has been increasing in recent years, especially in the semi-arid region, due to the hot climate, producing fruits with good quality throughout the year [2]. However, Custard apple development is reduced in the fall/winter period. According to [3], it is essential to increase the availability of nutrients in the soil through fertilization to meet the physiological requirements for obtaining good growth and development of the custard apple plant.

Researches with different doses of nutrients under field conditions, for the anonaceae, is an important strategy to optimize the production of this crop [4]. There is little information on the nutritional requirement of the custard apple plant [5]. According to [6], the plant extracts large amounts of nutrients from the soil. Since nitrogen is the most required nutrient, nitrogen fertilization increases the vegetative and reproductive stimulus of custard apple plant [3]. Working with doses of nitrogen and potassium in different production seasons, [7] observed linear increase in leaves nitrogen contents with the increase of nitrogen fertilization in the prunings performed in the fall/winter period. However, high doses of nitrogen can cause acidification in the soil and influence nutrient availability and plant nutrition [8, 9].

Another alternative is the use of humic substances, which provides an improvement in nutrient absorption [10]. According to [11], the use of humic substances improves the absorption of the applied fertilizer and increases the nutrient content in custard apple leaves. In addition, it favors plant photosynthesis [6]. However, the effectiveness of humic substances is influenced by the studied crop, soil conditions, dose, source, and form of application [12, 13, 14]. It may act directly on the growth of the plant or on improving the conditions for the development of the crop [10].

There is little information in the literature on custard apple nutrition, on the effect of humic substances and ideal nitrogen doses in autumn/winter pruning. According [Te-to \[3\]](#), the custard apple producers modify the nitrogen/potassium ratio in the fertilization as a function of the time of the year, from 2 to 4 parts of nitrogen to 1 of potassium in autumn/winter pruning. Therefore, the objective of this work was to verify the influence of different doses of nitrogen and the application of humic substances on soil chemical attributes and custard apple plant nutrition.

2. MATERIAL AND METHODS

2.1. Site of study

The experiment was conducted from May 10th to October 24th, 2017, in a commercial custard apple orchard at Rancho Alegre farm, in the municipality of Anagé, Southwest region of the State of Bahia, located at 14°26' south latitude and 41°04' Longitude West of Greenwich, with 335 m of altitude, [\(elevation above sea level?\)](#) in which prevails the semi-arid climate, classified as Bwsh according to Köppen, with average precipitation of 656 mm year⁻¹, average annual temperature of 22.3 °C, minimum of 19.0 °C and maximum of 29.0 °C.

2.2. Experimental design

The experimental design was a randomized complete block design with 4 x 2 factorial design, with 4 N doses (0, 284, 568 and 852 g of N per plant, in the form of urea), with and without the application of humic substances, with 4 replicates. The plots were composed of 4 plants, the first and the last plant were considered as borders.

2.3. Plant and Area Characteristics

The custard apple plants of the experiment were 20 years old, originated from seed propagation, planted in 7 x 4 m spacing. They had a mean height of 3.65 m, crown diameter of 4.42 m and trunk diameter of 0.14 m, at 20 cm height from the soil. The irrigation used is of the micro sprinkler type, with flow rate of 30 liters hour⁻¹, a daily irrigation shift of 3 hours, one sprinkler being used for each two plants. The soil analysis presented in Table 1 followed the methodology of [15], the soil texture is sandy clay loam.

Table 1. Soil chemical analysis of the area planted with custard apple, Anagé-BA, 2017

Depth	pH (H ₂ O)	P ⁺ mg dm ⁻³	K ⁺	Ca ⁺²	Mg ⁺²	Al ⁺³	H ⁺	Na ⁺	V %	O.M.
0-20cm	7.1	35	0.54	4.4	3.5	0	1.1	0.13	89	1.3
20-40 cm	7.0	14	0.53	4.2	3.2	0	1.2	0.11	87	0.8

* Mehlich 1.

The climatic characteristics of the experimental period are presented in Figure 1, according to data from INPE meteorological station 32494.

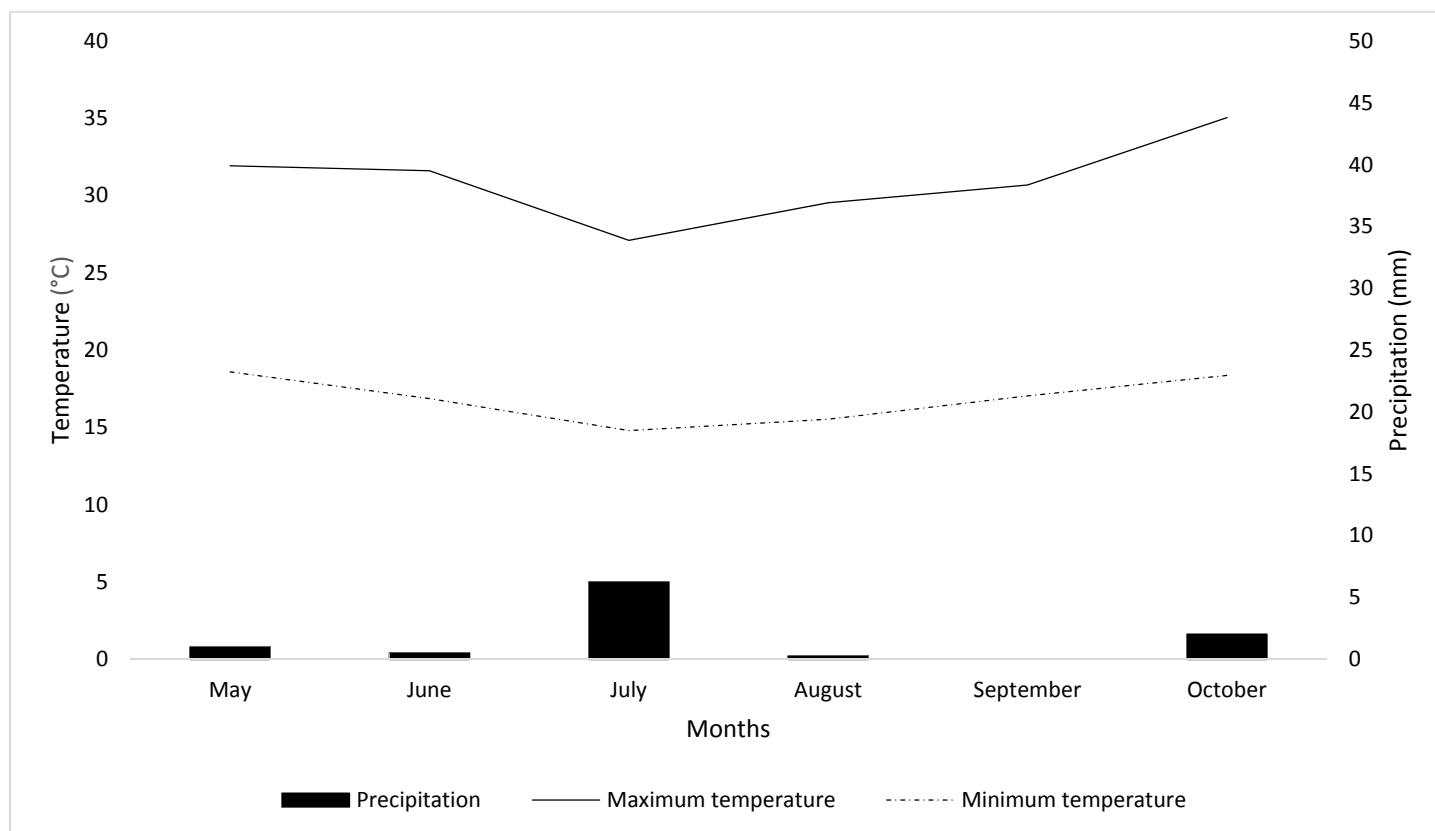


Figure 1. Monthly precipitation, mean maximum and minimum temperatures from May to October, Anagé-BA, 2017

In order to characterize the nutritional status of the custard apple plants, the fifth and sixth leaves of the middle third were collected in 20 plants randomly prior to the implantation of the experiment [16]. Data of the initial leaf nutritional characteristics of the custard apple plants are presented in Table 2, and analysis was performed according to the methodology described by [15].

Table 2. Leaf nutrient content of custard apple before the implantation of the experiment, Anagé-BA, 2017

N	P	K	Ca	Mg	S	B	Cu	Fe	Mn	Zn
g kg ⁻¹					mg kg ⁻¹					
32.7	2.27	14.6	11.49	4.16	2.13	63.97	17.87	160.04	65.65	10.85

Pruning was done with scissors, in order to standardize the branches, leaving them with approximately 0.2 m, the manual defoliation of these branches was also performed. After pruning, the plants were fertilized with 50 g of FTE BR 12 and 200 g of TOPMIX (52% of P₂O₅ and 11% of N), applied at 0.2 m depth in crown projection. 350 g of potassium chloride (58% of K₂O) was applied, in 7 doses every 15 days, starting 45 days after pruning.

3.4. Treatments

88 The application of the humic substance was performed after pruning, applying 10 g per plant of commercial product Ks
 89 100 (Omnia Brazil), distributed in the projection of the crown. The physico-chemical characteristics of this product,
 90 analyzed according to the methodology described by [17], are: N = 0.45%, P2O5 = (total) 0.08%, P2O5 (2% citric acid) =
 91 0.02 %, K2O (H2O) = 2.00%, Ca = 0.26%, Mg = 0.30%, S = 0.12%, Fe = 0.26%, CO = 4.75%, pH = 9 , 65, moisture =
 92 11.10%, density = 0.82 M / V, Si = 3.85%, Al = 1.85%, fulvic acid = 2.34% and humic acid = 3.65%[\(present these data in](#)
 93 [table](#). The different nitrogen doses were applied every 15 days from the pruning operation, using urea as the source of N
 94 (46% of N), applied under the projection of the crown.

95

96 **3.5. Variables studied and statistical analysis**

97

98 The nutritional status of the plants was evaluated collecting leaves following the methodology proposed by [16], at 156
 99 days after pruning. The leaves were packed in paper bags and washed in water, rinsed with distilled water and dried in an
 100 oven with forced air circulation at 65 °C until constant mass was obtained.

101 Soil samples were also collected in the crown projection in the 0-0.20 m layer, and the samples were dried in shade. After
 102 that, the samples were sent to the laboratory where the chemical analyzes of macro and micronutrients were carried out
 103 according to the methodology of [15].

104 After testing for normality and homogeneity the analysis of variance was performed using SISVAR Software [18], and
 105 quantitative data were submitted to regression analysis.

106 **3. RESULTS**

107

108 **3.1. Soil chemistry**

109

110 The levels of remaining P, K, H + Al (Table 3), base saturation and soil organic matter at 156 days after pruning, when it
 111 was observed that they were not influenced by the applied treatments. According to the classes proposed by [19], the
 112 values of potassium, base saturation and soil organic matter content are very good, medium and low, respectively.

113

114 Table 3. Mean values of remaining phosphorus, potassium, hydrogen + aluminum, base saturation and organic matter
 115 after to the application of humic substances (HS), general mean and coefficient of variation (C.V.) in soil cultivated with
 116 custard apple, Anagé-BA, 2017

	P(rem)	K ⁺	H+Al	V	O.M.
	mg dm ⁻³	cmol _c dm ⁻³	mmol _c dm ⁻³	%	g dm ⁻³
With HS	54.45 ^{ns}	0.64 ^{ns}	41.18 ^{ns}	46.63 ^{ns}	16.47 ^{ns}
Without HS	57.38	0.53	29.43	54.73	13.42
Mean	55.92	0.58	35.31	52.20	14.95
C.V. (%)	8.97	37.19	63.70	33.74	33.29

117 ^{ns} non-significant by F test (*P*=0.05)

118

119 Micronutrients (Table 4) were not influenced by the application of humic substances and there was no significant
 120 regression for the nitrogen doses. According to the classes proposed by [19], the levels found in the present experiment
 121 for copper, manganese and zinc are high and iron is low.

122

123 **Table 4. Average micronutrient content as a function of the application of humic substances (HS), general mean**
 124 **and coefficient of variation (C.V.) of soil cultivated with custard apple, Anagé-BA, 2017**

	Cu	Fe	Mn	Zn
	mg dm ⁻³			
With HS	2.85 ^{ns}	14.83 ^{ns}	14.98 ^{ns}	10.99 ^{ns}
Without HS	2.68	12.83	12.24	11.44
Mean	2.77	13.84	13.60	11.21
C.V. (%)	40.70	38.76	43.54	57.78

125 ^{ns} non-significant by F test ($P=0.05$)

126

127 The increase of the nitrogen doses caused reduction of 1.21 in soil pH, in the calcium content of 15.18 mmol_c dm⁻³ and
 128 | 9.71 mmol_c dm⁻³ in the magnesium content (Figure 2 (A, C and D)). ~~corroborating~~Corroborating with the results found by
 129 [8] working with doses of urea in dwarf coconut. This variation in pH caused the availability of aluminum to increase 4.48
 130 mmol_c dm⁻³ as a function of N rates (Figure 1B). This same behavior was observed by [9] working with doses and sources
 131 of nitrogen in soil cultivated with mombaça grass.

132 The phosphorus content increased by 3.21 mmol_c dm⁻³ as a function of the growth of the nitrogen doses (Figure 2E).
 133 Different behavior from that observed by [9], where the levels of phosphorus reduced with increasing doses of nitrogen.
 134 Boron levels reduced by 0.06 with the nitrogen rates.

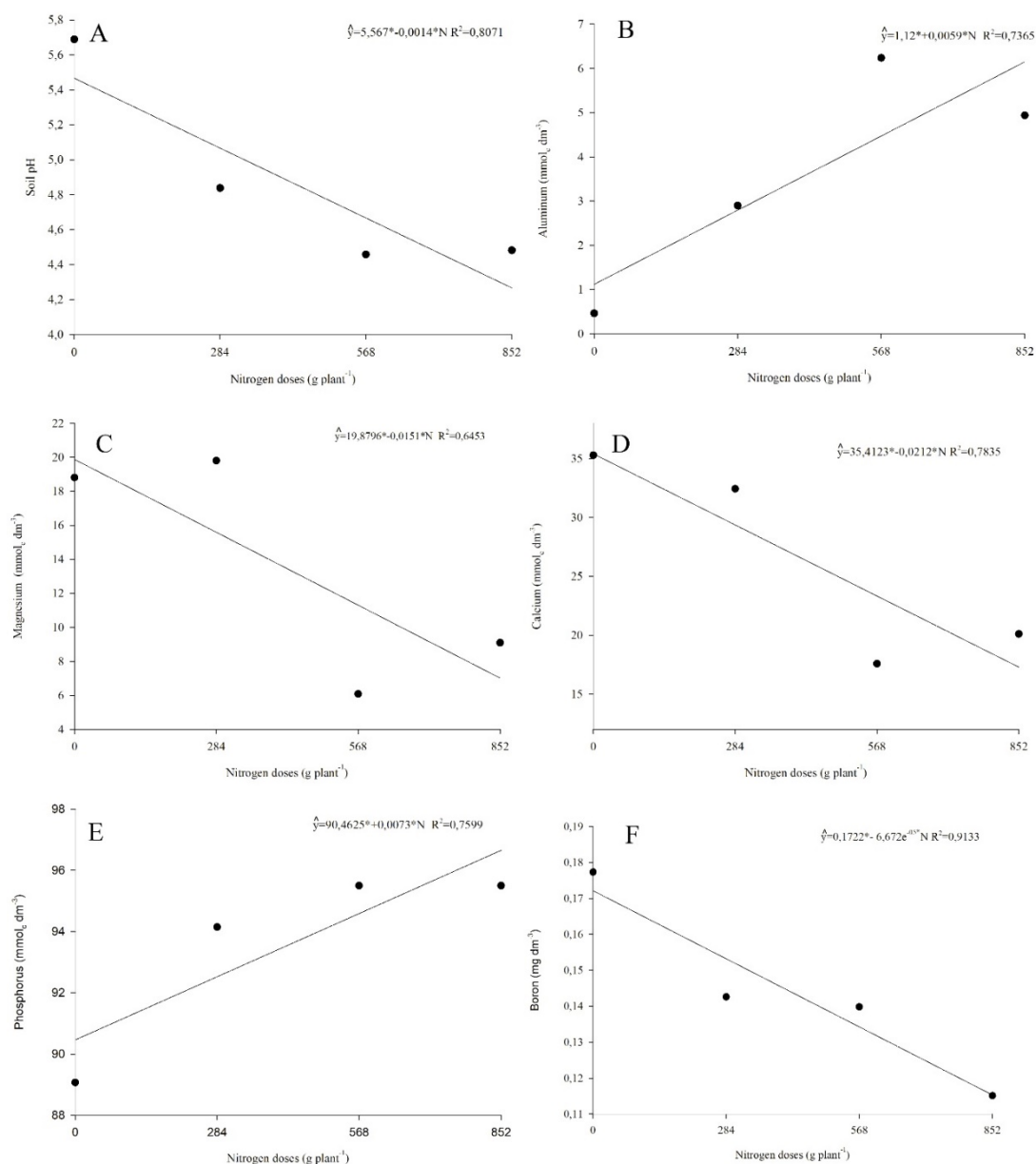


Figure 2. PH values (CaCl₂, A) and aluminum (B), magnesium (C), calcium (D), phosphorus (E) and boron (F) values of soil cultivated with custard apple as a function of nitrogen doses, Anagé-BA, 2017. * Significant ($P=0.05$).

3.2. Nutrition of the plants

The application of humic substances increased the potassium contents and did not influence the contents of the other nutrients in the custard apple leaves (Table 5). This behavior differs from that observed by [11], working with custard apple, which observed an increase in nitrogen contents with addition of humic substances. Working with pineapple seedlings, [20] observed increases in potassium content in their leaves with the use of humic substances. The levels of N are in the ideal range according to [3], since P, K and Mg are above and S and B are below. Studying nitrogen rates and application of humic substance [11] obtained lower levels of N, P, K, Mg and S. Working with organic fertilizer [21] found similar levels of N, Mg and S, lower than P, K and higher Ca. The boron and manganese contents are below the average levels shown by [22]. Iron and copper contents are close to those found by the same authors. The contents of B, Cu and Zn are close to those found by [5] and below those found by [21]. The same authors found smaller Fe and Mn contents.

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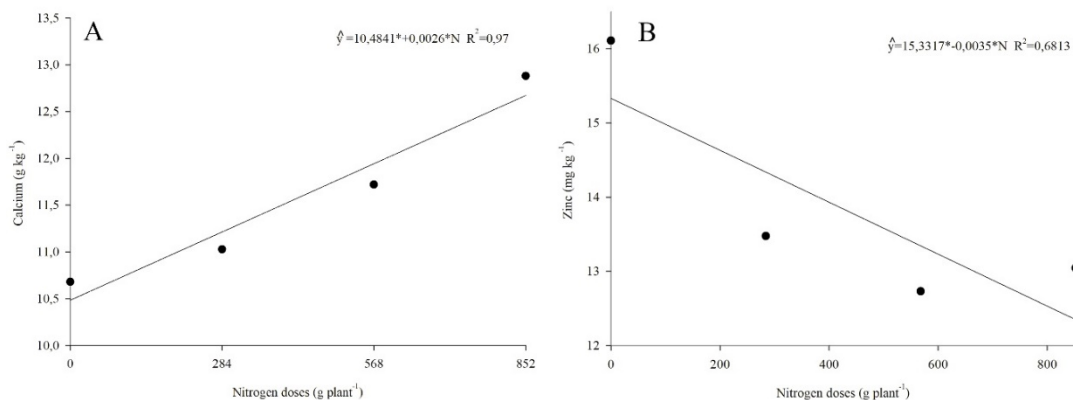
152 **Table 5. Average leaf nutrient content due to the use of humic substances (HS), general averages and coefficient**
153 **of variation (C.V.) the custard apple, Anagé-BA, 2017**

	N	P	K	Mg	S	B	Cu	Fe	Mn
	g kg ⁻¹					mg kg ⁻¹			
With HS	30.6 ^{ns}	1.98 ^{ns}	14.65 [*]	3.53 ^{ns}	2.21 ^{ns}	38.48 ^{ns}	6.26 ^{ns}	145.68 ^{ns}	62.65 ^{ns}
Without HS	31.29	2.04	16.71	3.62	2.24	43.35	6.48	146.13	65.03
Mean	30.97	2.01	15.18	3.58	2.23	36.18	6.37	145.91	63.84
C.V. (%)	4.48	10.58	9.25	12.74	9.28	42.55	7.28	9.87	18.72

154 ^{*}, ^{ns} significant and non-significant by F test ($P=0.05$)

155

156 Leaf calcium levels (Figure 3A) showed tendency to increase with increasing nitrogen doses, increasing 1.98 g kg⁻¹ and
157 zinc contents (Figure 3B) had reduction of 1.08 mg kg⁻¹ depending on the nitrogen doses. The levels of zinc and calcium
158 are below ideal according to [3, 22]. Calcium levels are close to that observed by [5, 11].



159

160 **Figure 3. Calcium (A) and zinc (B) content in custard apple leaf as a function of nitrogen doses, Anagé-BA, 2017.**
161 *** Significant ($P=0.05$).**

162

163 4. DISCUSSION

164

165 4.1. Soil Chemistry

166

167 Soil acidification was probably caused by nitrification reaction from ammonium to nitrate. The urea initially has an alkaline
168 reaction in the soil, later the nitrification performed by microorganisms of the genus Nitrosomonas and Nitrobacter cause
169 acidification in the soil [23, 24]. Therefore, it can be affirmed that the application of nitrogen fertilizers acidifies the soil [25].

170 The reduction in Ca and Mg contents occurred due to changes in availability caused by pH. According to [26], this can
171 occur due to the leaching loss of cations (Ca^{2+} and Mg^{2+}), being replaced in the soil cation exchange complex by
172 aluminum.

The phosphorus content increased with the nitrogen doses probably due to the favoring of the decomposition of the pruning remains. The degradation of straw is favored by the application of nitrogen [27], because the microorganisms involved require nitrogen for decomposition.

The reduction in soil boron availability with increased nitrogen rates is probably associated with pH variation. By studying the variation in boron adsorption as a function of pH, [28] found that low and very high values favor adsorption, reducing availability. In this work, the pH at the highest nitrogen dose reached 4.5, being within the favorable range of adsorption. In addition, [29] reports that in acidic pH, organic matter is the main source of boron, with the increase associated with the same behavior described for phosphorus.

4.2. Nutrition of the plants

The increase in potassium leaf content with the use of humic substances is associated with increased absorption. The application of humic substances increases the electrochemical gradient between the roots and the soil [30] and acts on the expression of the genes encoding H⁺ ATPases proteins and their activation [13]. The same authors report that the action of the humic substances are directly related to pH, this shows that the lack of results in the other nutrients may be associated with this.

The low calcium contents occurred due to the high levels of potassium and magnesium in the soil, which compete for the same absorption sites [29], which is justified by the high levels of foliar potassium shown above. The increase in foliar calcium content with nitrogen doses is associated with reduced competition with magnesium contents, which reduced the effect of pH.

Applications of nitrogen doses led to an imbalance in the nutrition of the plants, where boron, zinc, manganese, and sulfur contents are limiting their development. Sulfur and manganese are directly linked to the nitrogen assimilation reduction process [29].

Therefore, other studies should be carried out, varying the sources of nitrogen, in order to avoid the effect of acidification of the soil, and to verify if the use of high nitrogen doses in the crop / crop cycle conducted in the autumn / winter is justified. Improving also fertilization with micronutrients and with sulfur.

5. CONCLUSION

Under the conditions in which the present work was conducted, we concluded that high nitrogen levels cause acidification in the soil, reducing the availability of calcium, magnesium and boron and increasing the levels of aluminum and phosphorus. The application of humic substance in soil cultivated with custard apple does not affect the absorption of most of nutrients with the exception of potassium, causing an increase, and it does not influence the chemical attributes of the soil.

6. REFERENCES

1. LEMOS EEPde. The production of anonaceous plants in Brazil. Revista Brasileira de Fruticultura. 2014; 36: 077-085.
2. São José AR, Pires MdeM, Freitas ALGEde, Ribeiro DP, Perez LAA. News and perspectives of the anonáceas in the world. Revista Brasileira de Fruticultura. 2014; 36: 086-0993.

- 213 3. São José AR, Prado NB, Bomfim MP, Rebouças THN, Mendes HTAE. March of nutrient absorption in anonaceous.
214 Revista Brasileira de Fruticultura. 2014; 36: 176-183.
- 215 4. Rozane DE, Natale W. Liming, fertilization and mineral nutrition of anonaceae. Revista Brasileira de Fruticultura. 2014;
216 36: 166-175.
- 217 5. Cavalcante LF, Pereira WE, Curvêlo CRS, Birth JAM, Cavalcante IHL. Nutritional status of pines under organic soil
218 fertilization. Agronomic Science Journal. 2012; 43: 579-588.
- 219 7. Souza IVB. Characteristics and quality of pine nut fruits (*Annona squamosa* L.), in the State of Bahia, due to NK
220 fertilization. PhD Thesis, State University of Southwest of Bahia, Vitória da Conquista, 2016. 156p.
- 221 6. Cavalcante IHL, Cunha MdosS, Rocha LFda, Santos EM, Junior GBdaS. Physiological indexes of the pine tree as a
222 function of nitrogen fertilization and humic substances. Agrarian Sciences Journal. 2014; 57: 85-89.
- 223 8. Sobral LF, Nogueira LC. Influence of nitrogen and potassium, via fertirrigation, on soil attributes, foliar critical levels and
224 dwarf coconut production. Brazilian Journal of Soil Science. 2008; 32: 1675-1682.
- 225 9. Rosado TL, Gontijo I, Almeida MSde, Andrade FV. Sources and doses of nitrogen and changes in chemical attributes
226 of a latosol cultivated with mombaça grass. Brazilian Journal of Soil Science. 2014; 38: 840-849.
- 227
228 10. Nardi S, Pizzeghello D, Muscolo A, Vianello A. Physiological effects of humic substances on higher plants. Soil
229 Biology and Biochemistry. 2002; 34: 1527-1536.
- 230 11. Cunha MS, Junior GBS, Cavalcante ÍHL, Santos EM, Albano FG, Rocha LF. Nutritional status of custard apple
231 (*Annona squamosa*) as a function of nitrogen fertilizing and humic substances. Journal of the Faculty of Agronomy.
232 2014; 31: 493-509.
- 233 12. Canellas LP, Olivares FL, Aguiar NO, Jones DL, Nebbioso A, Mazzei P, Piccolo A. Humic and fulvic acids as
234 biostimulants in horticulture. Scientia Horticulturae. 2015; 196: 16-17.
- 235 13. Ramos AC, Dobbss LB, Santos LA, Fernandes MS, Olivares FL, Aguiar NO, Canellas LP. Humic matter elicits proton
236 and calcium fluxes and signaling dependent on Ca²⁺ -dependent protein kinase (CDPK) at early stages of lateral
237 plant root development. Chemical and Biological Technologies in Agriculture. 2015; 2: 1-12.
- 238 14. Silva AC, Canellas LP, Olivares FL, Dobbss LB, Aguiar NO, Frade DAR, Rezende CE, Peres LEP. Promotion of root
239 growth of tomato seedlings by humic substances isolated from peat bogs. Brazilian Journal of Soil Science. 2011;
240 35: 1609-1617.
- 241 15. Brazilian Agricultural Research Corporation - EMBRAPA. Manual of chemical analyzes of soils, plants and fertilizers.
242 National Soil Research Center. 2.ed. rev. Rio de Janeiro, 2009.
- 243 16. Pin ACdeQ. Gravile. In: Chrysostom LA, Naumov A. (eds.). Fertilizing for high productivity and quality: tropical fruit
244 trees of Brazil. Translation Lindburgo Araújo Crisóstomo. Fortaleza: Embrapa Agroindústria Tropical, 2009. pp:
245 206-222.
- 246 17. Ministry of Agriculture, Livestock and Supply (MAPA). Manual of official analytical methods for fertilizers and
247 correctives. Brasília, MAPA, 2017.
- 248 18. Ferreira DF. Sisvar: a computer statistical analysis system. Science and Agrotechnology. 2011; 35: 1039-1042.
- 249 19. Ribeiro AC, Guimarães PTG, Alvarez V VH. Recommendations for the use of correctives and fertilizers - 5th
250 Approach. Soil Fertility Commission of the State of Minas Gerais, Viçosa, MG.1999.
- 251 20. Santos PCdos, Silva MPSda, Freitas SdeJ, Berilli SdaS, Altoé JA, Silva AdeA, Carvalho AJCde. Humic and
252 brassinosteroid acids in the growth and nutritional state of shoots of pineapple crowns. Brazilian Journal of
253 Agricultural Sciences. 2014; 9: 532-537.
- 254 21. Leonel S, Araújo JF, Tecchio MA. Biofertilization and organomineral fertilization: denutrient concentration in the leaf
255 and yield of pine nut fruits. Irriga Magazine. 2015; 1: 40-51.

- 256 22. São José AR, Souza IVB, Morais OM, Rebouças TNH. Anonáceas: production and market. Vitória da Conquista:
257 UESB / DFZ. 1997.
- 258 23. Urquiaga S, Malavolta E. Urea: An organic fertilizer with potential for organic agriculture. Caderno de Ciência &
259 Tecnologia. 2002; 19: 333-339
- 260 .
- 261 24. Moreira FMS, Siqueira OJ. Microbiology and soil biochemistry. 2.ed. Federal University of Lavras, Lavras, 2006.
- 262 25. GE S, ZHU Z, JIANG Y. Long-term impact of fertilization on soil pH and fertility in an apple production system. Journal
263 of Soil Science and Plant Nutrition. 2018; 18: 282-293.
- 264 26. Lange A, Carvalho JLNde, Damin V, Cruz JC, Marques JJ. Changes in soil attributes due to application of nitrogen
265 and straw in a no - tillage system in maize crop. Rural Science. 2006; 36: 460-467.
- 266 27. Vitti AC, Trivelin PCO, Cantarella H, Franco HCJ, Faroni CE, Otto R, Trivelin MO, Tovajar GJ. Mineralization of the
267 straw and growth of sugarcane roots related to nitrogen fertilization of plantation. Brazilian Journal of Soil Science.
268 2008; 32: 2757-2762.
- 269 28. Soares MR, Casagrande JC, Alleoni LRF. Adsorption of boron in acidic soils as a function of pH variation. Brazilian
270 Journal of Soil Science. 2008; 32: 111-120.
- 271 29. Malavolta E. Mineral nutrition of plants. São Paulo: Agronômica Ceres, 638p
- 272 .
- 273 30. Zandonadi DB, Canellas LP, Façanha AR. Indoleacetic and humic acids induce lateral root development through a
274 concerted plasmalemma and tonoplast H⁺pumps activation. Planta. 2007; 225: 1583–1595.
- 275