

## Growth and Gas Exchange of *Cucurbita pepo* L. Under Nitrogen and Silicon Fertilization

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### Abstract

Zucchini (*Cucurbita pepo* L.) is a horticultural crop of great socioeconomic importance in Brazil and in the world. However, inappropriate fertilization management, such as over-fertilization of soils, may become a limiting factor for its development. Thus, the aim of this study was to evaluate the morpho-physiological behavior of zucchini submitted to nitrogen (N) doses applied via soil and foliar application of silicon (Si). The treatments were distributed in split-plot scheme in a randomized block design, with three replications. The plot was formed by silicon levels (0.0 and 6.0 g plant<sup>-1</sup>) and the subplots constituted by five nitrogen levels (30, 60, 90, 120 and 150 kg ha<sup>-1</sup>), adding up to 30 experimental units. Gas exchanges and growth parameters were assessed at 35 days after planting. The data were submitted to analysis of variance by the F test and in the cases of significance was performed a polynomial regression analysis for the nitrogen factor and Tukey test for the silicon factor. The supply of Si positively enhances the effects of N on growth characteristics and gas exchanges of zucchini. The simultaneous application of Si and N does not influence the leaf area of zucchini. The N dose of 93.9 kg ha<sup>-1</sup> provides greater assimilation of CO<sub>2</sub> in zucchini plants under the conditions in which the experiment was performed.

**Keywords:** Zucchini, fertilizers, morpho-physiological traits

### 1. Introduction

The zucchini or courgette (*Cucurbita pepo* L.) is a summer squash that belongs to the family Cucurbitaceae. The center of origin of zucchini is the tropical Americas, specifically Central Mexico and South of the United States of America. In Brazil, the average yield of this vegetable varies from 8 to 10 t ha<sup>-1</sup> (Filgueira, 2012). According to Carpes et al. (2008), the zucchini is among the ten vegetables of greatest economic value in Brazil.

Inadequate fertilization management is one of the main factors impairing the growth, development and productivity of zucchini. Nitrogen is the key limiting nutrient for most crops. Therefore, nitrogen fertilization is of fundamental importance, since nitrogen (N) is an essential macro-nutrient that directly influences plant growth and development, affecting the source-sink relationship and photoassimilate distribution between the vegetative and reproductive organs (Porto et al., 2012). In this context, scarce nitrogen content in the soil causes nitrogen deficiency in plants, negatively affecting its biochemical and physiological processes and, consequently, the growth (Pedo et al., 2014). On the other hand, beneficial effects of nitrogen fertilization are reported by several authors in different crops, such as Antunes et al. (2014) in African cucumber (*Cucumis metuliferus* L.), Porto et al. (2012) in zucchini (*Cucurbita pepo* L.) and Morais et al. (2015) in maize plants (*Zea mays* L.).

The rational use of nitrogen fertilizers is important, since excessive N content in the soil can be harmful to both the plant and the environment. In this sense, better yields can be achieved by the combination of nitrogen and silicon (Si) fertilization (Artigiani et al., 2014). Although not an essential nutrient, silicon is beneficial to plants, being associated with several indirect effects on plants, especially enhanced photosynthetic capacity, reduced transpiration and increased mechanical resistance of the cells (Ferraz et al., 2014).

Thus, the rational application of these fertilizers, in order to improve the nitrogen use efficiency in plants, is of fundamental importance for the development of sustainable agriculture (Xu, Fan, & Miller, 2012). Given the scarcity of information on the joint action of these nutrients, especially in vegetables, the aim of this work was to evaluate the morphophysiological behavior of zucchini submitted to nitrogen doses applied via soil and foliar application of silicon.

## 2. Material and Methods

The experiment was carried out under field conditions in the Agroecology Sector of the Agrarian and Exact Sciences Department (DAE) of the State University of Paraíba (UEPB), Campus IV, located in the municipality of Catolé do Rocha, Paraíba, Brazil, having the geographical coordinates of 6°20'38" S and 37°44'48" W, with an altitude above sea level of 275 m.

The zucchini cultivar 'Caserta' was used in the experiment, being conducted in a spacing of 1 × 1 m. Planting was done by direct sowing in pits. Pits of 30 × 30 × 30 cm size were dug, then 2 L of cattle manure were added in each.

The soil of the experimental area is classified as eutrophic Fluvisol Neosol, with a sandy loam texture (Embrapa, 2011). Before the experiment started, soil samples were collected in the 0-20 cm depth for physical and fertility analysis (Table 1) and the chemical analysis of the cattle manure was performed (Table 2), according to the methodology proposed by Embrapa (2011). The soil was prepared by plowing sequenced by harrowing.

Table 1. Chemical and physical characteristics of the eutrophic fluvisol neosol used in the experiment

<i>Chemical Characteristics (Fertility)</i>												
pH	Ca	Mg	Na	K	S	H+Al	CEC	BS	OC	OM	N	PA
----- cmol <sub>c</sub> dm <sup>-3</sup> -----								----- % -----		----- mg dm <sup>-3</sup> -----		
6.7	1.49	0.54	0.10	1.72	3.85	0.00	3.85	100	0.67	1.2	0.07	16.83
<i>Physical Characteristics</i>												
Sand	Silt	Clay	TC	SD	PD	TP	FC	PWP	SAW			
----- g Kg <sup>-1</sup> -----			----- g dm <sup>-3</sup> -----			(%)		----- g Kg <sup>-1</sup> -----				
640.0	206.0	154.0	Sandy franc	1.54	2.68	42.54	146.9	76.60	70.3			

*Note.* CEC = Cation exchange capacity; BS = Base sum; OC = organic carbon; OM = organic matter; PA = Phosphor assimilable; TC = textural class; SD = soil density; PD = particle density; TP = total porosity; FC = field capacity; PWP = permanent wilting point; SAW = soil available water.

The treatments were distributed in split-plot scheme in a randomized block design (RBC), with three replications. The plot was formed by silicon levels (0.0 and 6.0 g plant<sup>-1</sup>) and the subplots constituted by five nitrogen levels (30; 60; 90; 120 and 150 kg ha<sup>-1</sup>). The nitrogen source used was urea (45% N). The silicon levels were based on an estimate of 60 kg ha<sup>-1</sup>. The silicon source used was Bugram Protect<sup>®</sup>, with 100% pure silicon dioxide, pH 6/8 and 0.90% moisture. The N doses were equally divided and applied to the soil twice, first at 15 DAP (days after planting) and the second application at 30 DAP. Si was also divided into two applications, the first at 14 DAP and the second at 28 DAP, both via foliar. Thus, in the first application 540 g of silicon dioxide was diluted in 30 L of water. In the second application 540 g was diluted in 40 L of water. The silicon solutions were uniformly applied using a 10 L capacity backpack sprayer. In total, 1.080 kg of silicon dioxide were used in the present study.

Micro-irrigation was performed through a drip irrigation system, spaced at 0.2 m and with a flow rate of 1.7 liters per hour, at a service pressure of 147 kPa, using 16 mm drip tapes. The water was supplied through an Amazon well near the experiment site, and it is classified as moderately saline (1.2 dS m<sup>-1</sup>) (Ayers & Westcot, 1999).

The physiological analyzes were performed at 35 DAP, between 9:00 and 10:00 am, using the portable infrared gas analyzer (IRGA-model LI-6400XT, LI-COR®, Nebraska, USA) with 300 mL min<sup>-1</sup> air flow and coupled light source of 1200 μmol m<sup>-2</sup> s<sup>-1</sup>. The following variables were assessed: net CO<sub>2</sub> assimilation rate (A-μmol CO<sub>2</sub> m<sup>-2</sup> s<sup>-1</sup>), stomatal conductance (gs-mol m<sup>-2</sup> H<sub>2</sub>O s<sup>-1</sup>), CO<sub>2</sub> concentration in intercellular spaces (Ci-μmol CO<sub>2</sub> m<sup>-2</sup> s<sup>-1</sup>), transpiration rate (E-mmol H<sub>2</sub>O m<sup>-2</sup> s<sup>-1</sup>) and leaf temperature (TF-°C) and from these, the water use efficiency (WUE-A/E) and instantaneous carboxylation efficiency (iCE-A/Ci) were calculated. For the growth parameters, the following variables were measured: plant height, stem diameter, leaf number and leaf area.

The data were submitted to analysis of variance by the F test at 5% probability. Polynomial regression analysis for the nitrogen factor and Tukey test for the silicon factor were performed in case of significance. The statistical program R was used (R Core Team, 2018).

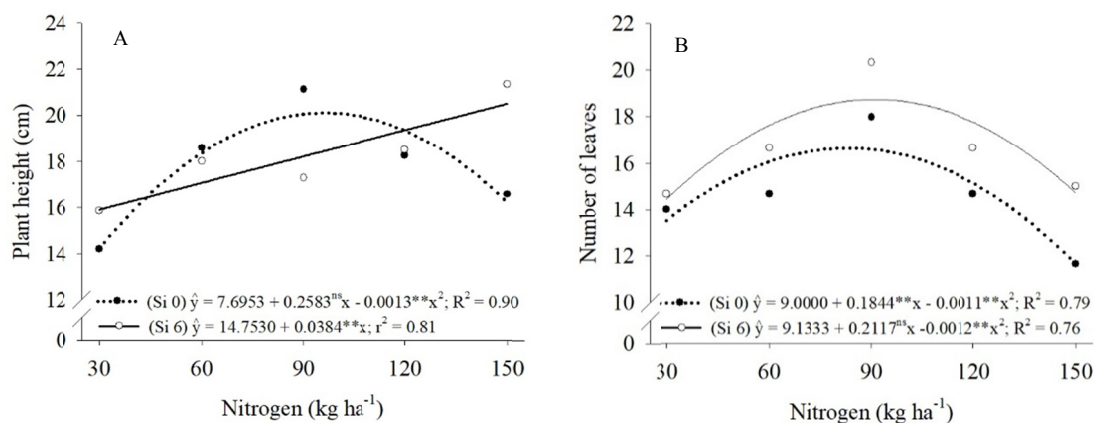
### 3. Results and Discussion

There was interaction between the studied factors, except for the leaf area. Plant height, stem diameter, leaf number and leaf area were influenced by the interaction between nitrogen (N) and silicon (Si), being the highest values for these variables obtained with the application of Si (Table 2).

Table 2. Average values for plant height, stem diameter, leaf number and leaf area of zucchini (*Cucurbita pepo* L.) under nitrogen doses and silicon leaf fertilization

Parameters	Silicon (g planta <sup>-1</sup> )	Nitrogen (kg ha <sup>-1</sup> )				
		30	60	90	120	150
Plant length	0	14.23 b	17.99 a	21.15 a	16.57 b	19.24 b
	6	15.87 a	19.00 a	17.29 b	18.51 a	21.37 a
Stem diameter	0	18.32 b	19.02 a	19.64 a	19.95 a	21.15 a
	6	19.11 a	16.14 b	17.17 b	17.26 b	20.24 b
Number of leaves	0	14.00 a	14.67 b	18.00 b	14.67 b	11.67 b
	6	14.67 a	16.67 a	20.33 a	16.67 a	15.00 a
Leaf area	0	6480.55 a	7387.90 a	7454.20 a	6419.40 a	7035.92 a
	6	5768.75 b	6157.45 b	5173.02 b	5461.33 b	5598.22 b

For the plant height, in the treatment without application of Si, the values adjusted to a quadratic effect, with the greatest increase obtained at 99.4 kg ha<sup>-1</sup> of N. On the other hand, under the application of Si, a linear increase in plant height is observed as the N doses increases. There was a 28.9% increase in plant height at the nitrogen dose of 150 kg ha<sup>-1</sup> when compared to the control treatment (30 kg ha<sup>-1</sup>) (Figure 1A). For the leaf number, the application of silicon provided better results, with greater efficiency being found at the N dose of 88.2 kg ha<sup>-1</sup> (Figure 1B). The beneficial effect of silicon can be associated with several indirect factors that stimulate growth and plant production. Silicon in plants promotes strengthening of the cell walls, increasing the amount of water in the plant and, consequently, of photoassimilates (Adrees et al., 2015).



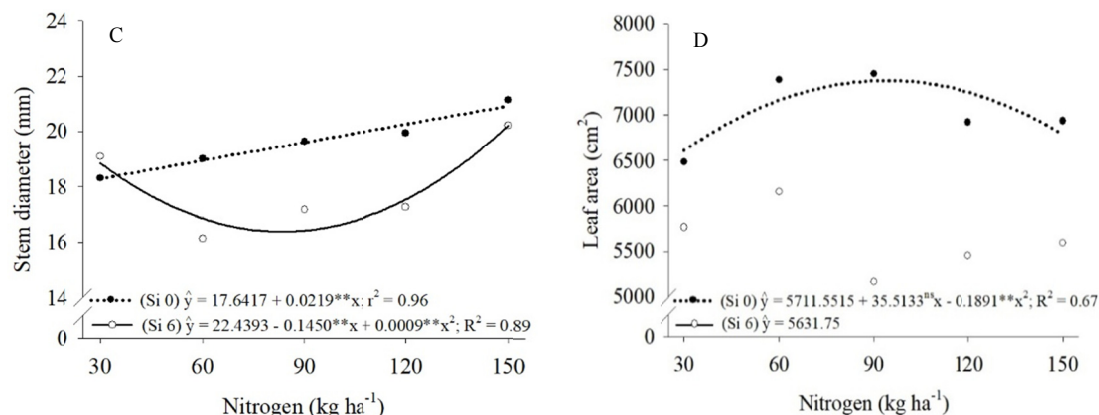


Figure 1. Plant height (A), leaf number (B), stem diameter (C), and leaf area (D) in zucchini (*Cucurbita pepo* L.) under nitrogen doses and silicon leaf fertilization

For the stem diameter and leaf area, the best results were obtained in the treatments without silicon application, with higher increases at the N doses of 150 and 93.9 kg ha<sup>-1</sup>, respectively (Figures 1C and 1D). This behavior can be attributed to the fact that nitrogen is part of the photosynthetic apparatus, e.g., being constituent of chlorophyll molecules, providing increased photosynthetically active leaf area, leading to greater CO<sub>2</sub> assimilation, thus resulting in improved growth and development (Basra, Iqbal, & Afbal, 2014).

Positive results from the use of nitrogen fertilizers were found by Liu et al. (2015) in lettuce plants (*Lactuca sativa* L.). Beneficial effects of silicon are reported by Keller et al. (2015) in hydroponically grown wheat seedlings (*Triticum turgidum* L.) under different doses of copper.

Stomatal conductance, net CO<sub>2</sub> assimilation, transpiration rate, internal CO<sub>2</sub> concentration, water use efficiency, instantaneous carboxylation efficiency and leaf temperature were altered by the interaction between nitrogen (N) and silicon (Si), with the highest values for these variables obtained with the application of Si (Table 3).

Table 3. Mean values for stomatal conductance (gs), net CO<sub>2</sub> assimilation rate (A), transpiration rate (E), CO<sub>2</sub> concentration in intercellular spaces (Ci), water use efficiency (WUE), instantaneous carboxylation efficiency (iCE) and leaf temperature (T<sub>Leaf</sub>) of zucchini (*Cucurbita pepo* L.) under nitrogen doses and silicon leaf fertilization

Parameters	Silicon (g planta <sup>-1</sup> )	Nitrogen (kg ha <sup>-1</sup> )				
		30	60	90	120	150
gs	0	0.239 b	0.263 b	0.324 b	0.213 b	0.204 b
	6	0.278 a	0.308 a	0.370 a	0.245 a	0.237 a
A	0	9.384 b	11.623 b	17.598 b	14.200 b	8.577 b
	6	10.691 a	12.736 a	19.374 a	16.776 a	10.778 a
E	0	4.846 b	5.494 b	8.586 b	6.624 a	4.692 b
	6	6.588 a	7.295 a	9.712 a	6.053 b	5.652 a
Ci	0	243.167 b	259.417 b	285.000 a	238.167 b	224.333 b
	6	268.750 a	276.750 a	288.733 a	243.917 a	229.250 a
WUE	0	1.910 a	2.065 a	2.074 a	2.129 b	1.744 b
	6	1.624 b	1.736 b	1.995 b	2.687 a	1.874 a
iCE	0	0.039 a	0.045 a	0.062 b	0.060 b	0.038 b
	6	0.040 a	0.046 a	0.067 a	0.069 a	0.047 a
T <sub>leaf</sub>	0	40.533 a	35.933 b	41.808 b	38.257 a	37.433 a
	6	39.783 a	41.313 a	43.268 a	39.200 a	37.531 a

For A, it can be observed that the highest rate (17.80 μmol of CO<sub>2</sub> m<sup>-2</sup> s<sup>-1</sup>) was registered at the N dose of 93.9 kg ha<sup>-1</sup>, showing an 11.5% increase when compared to the treatment without application of Si, which presented a

rate of  $15.76 \mu\text{mol of CO}_2 \text{ m}^{-2} \text{ s}^{-1}$  (Figure 2A). This result may be related to the fact that silicon promotes changes in the plant structure, improving the opening angle of the leaves, which favors a better use of light and consequently boosts photosynthetic efficiency. Also, the results found can be related to a higher nitrate uptake, since the process requires two moles of ATP for each mol of absorbed nitrate (Ávila et al., 2010; Ribeiro et al., 2011).

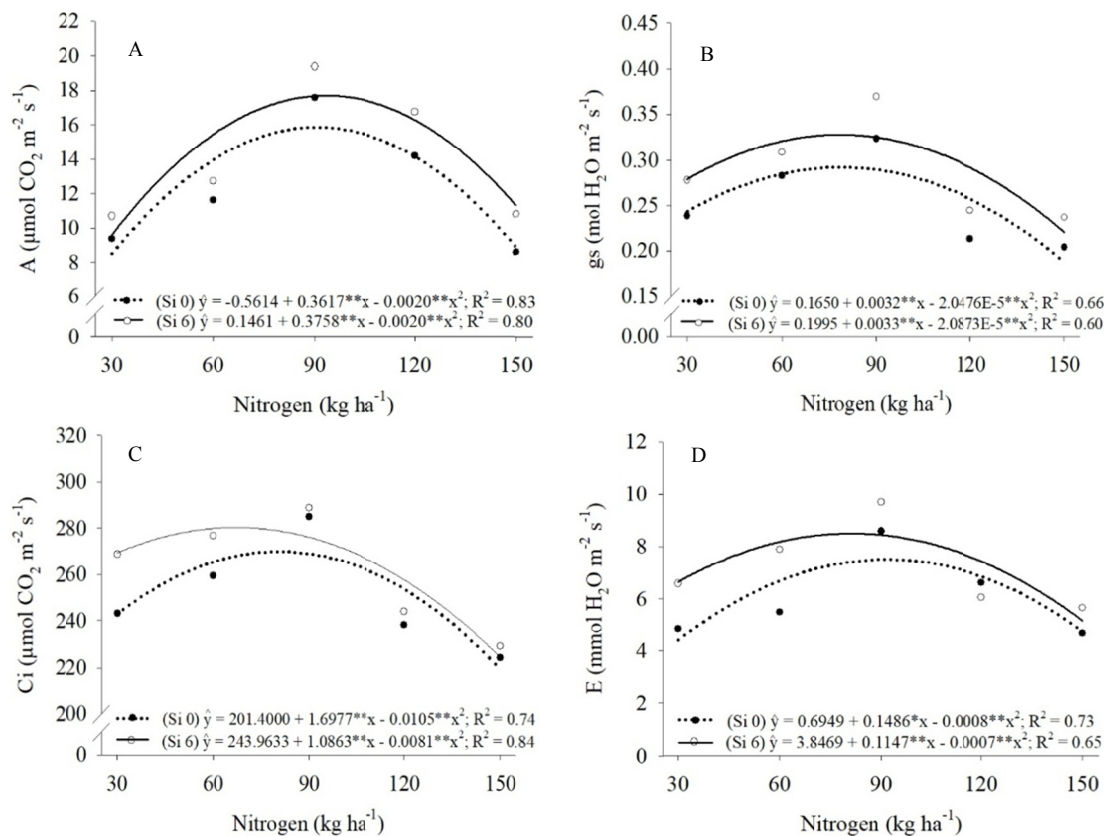


Figure 2. Net CO<sub>2</sub> assimilation rate (A-A), stomatal conductance (gs-B), CO<sub>2</sub> concentration in intercellular spaces (Ci-C) and transpiration rate (E-D) of zucchini (*Cucurbita pepo* L.) under nitrogen doses and silicon leaf fertilization

The variables gs and Ci presented a similar behavior to A, adjusting to a quadratic effect. The best increments for gs ( $0.33 \text{ mol of H}_2\text{O m}^{-2} \text{ s}^{-1}$ ) and Ci ( $280.38 \mu\text{mol of CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ ) were observed at the N doses of 79.1 and 67.1 kg ha<sup>-1</sup> (Figure 2B and 2C), respectively, under silicon foliar fertilization. This performance may be due to the increases recorded in A, since increased carbon assimilation may induce a greater stomatal opening, thus favoring the entry of CO<sub>2</sub> into the cells. The increase of the photosynthesis rate in the mesophyll cells causes the reduction of internal CO<sub>2</sub> concentration, which leads to the opening of the stomata (Taiz, Zeiger, Moller, & Murphy, 2017).

Regarding E, the highest rate ( $8.55 \text{ mmol of H}_2\text{O m}^{-2} \text{ s}^{-1}$ ) was registered at the N dose of 81.9 kg ha<sup>-1</sup>, showing a 14.2% increase when compared to the treatment without application of Si ( $7.49 \text{ mmol of H}_2\text{O m}^{-2} \text{ s}^{-1}$ ) (Figure 2D). The increase observed in the transpiration rate may be related to stomatal conductance, since the opening and closing of the stomata regulates the gas exchange between the internal and external media of the leaves.

Increases in stomatal conductance, net CO<sub>2</sub> assimilation and transpiration rate were also found by Curvêlo et al. (2013) and Ramos et al. (2013) in cotton (*Gossypium hirsutum* L.) and zucchini (*Cucurbita pepo* L.) under silicon foliar application.

Regarding iCE, the best result (0.07) was obtained at the N dose of 100.4 kg ha<sup>-1</sup>, an increase of 40% compared to the treatment without silicon (0.05) (Figure 3A). This result can be explained by the increase in CO<sub>2</sub> assimilation, where a larger amount of carbon is being fixed by Rubisco (Ferraz et al., 2012).

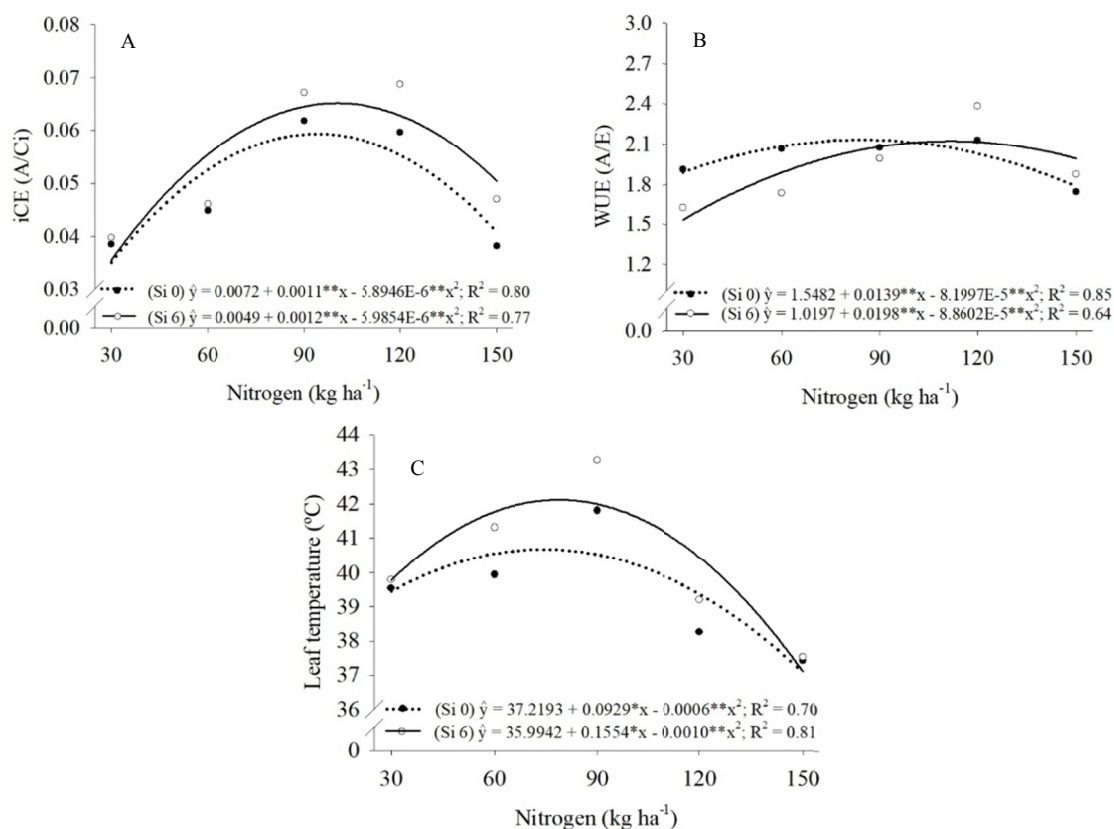


Figure 3. Instantaneous carboxylation efficiency (iCE-A), water use efficiency (WUE-B) and leaf temperature (Tleaf-C) of zucchini (*Cucurbita pepo* L.) under nitrogen doses and silicon leaf fertilization

The WUE adjusted to a quadratic effect, with the highest efficiency (2.13) obtained at the N dose of 111.7 kg ha<sup>-1</sup>, in the treatment under silicon application (Figure 3B). This result is important, since the WUE is the ratio of the amount of CO<sub>2</sub> that the plant assimilates (A) to molecule of water lost through the transpiration process (E) (Lima, Silva, Santos, & Souza, 2018). Similar results were obtained by Zamboni et al. (2016) in grapevine (*Vitis vinifera* L.) submitted to nitrogen fertilization.

The interaction between silicon and nitrogen provided a higher leaf temperature (42.03 °C), with the highest increase registered at the N dose of 77.7 kg ha<sup>-1</sup>, representing a 3% increase in relation to the treatment without silicon (40.8 °C) (Figure 3C). The leaf temperature is related to the transpiration process. The process of transpiration provides the plant with evaporative cooling, when releasing the water vapor through the stomata and cuticles. Through the process of transpiration, the thermal energy is balanced by the loss of heat. Since most of the water within the plant is released through the stomata, leaf temperature is influenced by stomatal conductance, once reduced stomata opening may lead to raise in leaf temperature. Under high temperatures on the leaf surface, plants may suffer some damage in biochemical and physiological processes.

#### 4. Conclusions

The supply of Si positively enhances the effects of N on growth characteristics and gas exchanges of zucchini.

The simultaneous application of Si and N does not influence the leaf area of zucchini.

The interaction between Si and N positively influences the gas exchanges of zucchini

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