



## **Gas Exchange and Nitrogen Compartmentalization of Eggplant under Nitrogen and Silicon Doses**

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### **Authors' contributions**

*This work was carried out in collaboration among all authors. Authors EGA, JLAR, KFP, RHCR and APS designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors AROG and ENS managed the analyses of the study. Author RMOSM managed the literature searches. All authors read and approved the final manuscript.*

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### **ABSTRACT**

To evaluate the effect of fertilization with N and Si on gaseous exchanges, dry mass, concentrations, accumulations and compartmentalization of nitrogen fractions in eggplant.

The experimental design was a randomized entirely design, in a 5 x 4 factorial arrangement with four replications and one plant per plot, totaling 80 experimental units.

The experiment was conducted in a protected environment at Center of Sciences and Agri-Food Technology of the Federal University of Campina Grande, Campus of Pombal, Paraíba, Brazil, between July and September 2016. The nitrogen doses applied was 25, 125, 250, 350 and 500 mg dm<sup>-3</sup> and four silicon doses was 0, 75, 150 and 200 mg dm<sup>-3</sup> both supplied by root. In pre-flowering

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stage were evaluated growth components; gas exchange, which are: photosynthesis, stomatal conductance, transpiration rate and intercellular CO<sub>2</sub> concentration; levels and accumulation of fractions of nitrogen (NO<sub>3</sub><sup>-</sup>, NH<sub>4</sub><sup>+</sup>, and total), and the silicon concentration in the leaves. There was no significant interaction ( $p > 0.05$ ) between the factors nitrogen and silicon doses for any of the evaluated variables. Nitrogen and silicon doses influenced the variables evaluated only independently each other. The nitrogen doses promoted increases in the photosynthetic rate and associated variables, dry matter yield of stem leaves and roots and in the concentration and accumulations of nitric, ammoniacal and total nitrogen in leaf, stem e roots and decrease the concentration of silicon in leaves. The silicon doses increased the leaf area index, the nitrate levels and accumulation in the roots and the silicon content in the leaves. In conclusion, the nitrogen supply increased the photosynthetic rate, dry mass and nitrogen accumulation and decreased the concentrations of silicon in leaf. Silicon did not interfered with growth of eggplant, however increased leaf area index, decreased nitrate levels and accumulations in the roots at lower doses of this element.

*Keywords: Solanum melongena L.; photosynthesis rate; beneficial element; nitrogen use efficiency.*

## 1. INTRODUCTION

Eggplant (*Solanum melongena* L.) is a fruit vegetable appreciated in many countries, including Brazil. This crop has importance in human food, due to its rich in mineral nutrients, antioxidants and vitamin [1]. For the adequate growth and development of eggplant, usually high nitrogen (N) doses are require [2,3]. However, the application of high doses of N is costly and can provide losses of this nutrient through the ammonia volatilization and nitrate leaching, causing negative environmental impacts [4]. In addition, nitrogen excess in plant can increase the susceptibility to the incidence of diseases [5]. Thus, it is necessary to establish strategies to increase the efficiency of nitrogen fertilization and to reduce the doses to be applied, especially in species with high nitrogen demand, such as eggplant.

Silicon (Si) is not an essential element, but it can provide several benefits to plants such as the induction of resistance to biotic [6,7] and abiotic stress by improving the plant architecture and light interception [8,9] and the better utilization of some nutrients, such as N [10,11].

Researches about the silicon and nitrogen interaction have been carried out mainly in grasses, especially rice [12,13,14]. In this sense, Ávila et al. [12] observed an increase in the chlorophyll and nitrogen contents under root silicon supply in rice cultivated in nutrient solution. According to Ávila et al. [12] the supposed increase of the photosynthetic rate provided by Si accumulation in the leaves of plants would increase the energy efficiency of N assimilation and therefore decrease the accumulation of soluble N forms such as

ammonium and nitrate in plant tissues. Campos et al. [13], found that the supply of Si in the culture medium, under excess of ammonium, mitigates the toxicity of ammonium in cucumber, resulting in higher accumulations of total nitrogen and dry matter of plants.

On the other hand, in other studies with rice cultivation, silicon leaf contents decreased with increasing N rates, increasing the susceptibility of the crop to the incidence of diseases [14].

Fruits vegetables not accumulate Si as well as rice, but some studies have shown that this element may contribute to increase the photosynthetic rate and plant growth for some species such as eggplant and melon [15,16]. However, there is a lack of work about the interaction between Si and N in horticultural species such as eggplant, in terms of their influence on the accumulation of mineral and total nitrogen in the tissues of the plant.

The objective of this work was to evaluate the effect of fertilization with N and Si on gaseous exchanges, dry mass production, concentrations, accumulations and compartmentalization of nitrogen fractions in eggplant.

## 2. MATERIALS AND METHODS

### 2.1 Location, Experiment Description and Conduction

The experiment was carried out in a greenhouse and in the Laboratory of Soil Fertility and Plant Nutrition, in a greenhouse of the Center of Science and Technology Agrifood, at the Federal University of Campina Grande (UFCG/CCTA), Campus of Pombal-PB, Brazil.

In the experiment were used samples of a Fluvent soil collected in the 0-40 cm layer in an area belonging to the CCTA Campus, which was pounded to break up clods and sieved through a 2.0 mm mesh sieve, for chemical and physics characterization according to the procedures described in Embrapa [17]. The soil sample showed the following attributes: pH (CaCl<sub>2</sub>) = 6,7; the exchangeable concentration of K<sup>+</sup>, Na<sup>+</sup>, Mg<sup>2+</sup>, Ca<sup>2+</sup> and Al<sup>3+</sup> of 0.22; 0.11; 2.3, 4.7 and 0.0 cmol<sub>c</sub> dm<sup>-3</sup>, respectively, P = 53 mg kg<sup>-1</sup>, and sandy loam texture.

The treatments were arranged in a 5 x 4 factorial arrangement, comprising five doses of N (25, 125, 250, 350 and 500 mg dm<sup>-3</sup>) and four doses of Si (0, 75, 150 and 200 mg dm<sup>-3</sup>). The experiment was carried out in a completely randomized design and four replications, totaling 80 experimental units. Each experimental unit consisted of a pot containing 6 dm<sup>3</sup> of soil, with two plants per pot.

Eggplant seedlings from variety 'Embú', were produced on expanded polystyrene trays with 128 cells filled with Tropstrato® (commercial substrate), which received one seed per cell. Thirty days after sowing, two leaves were transplanted per pot. Irrigations were performed manually according to the needs of the crop.

The nitrogen doses were applied in the form of urea (45% N) and Si in the form of potassium silicate, using commercial product Quimifol Silicio® (100 g Si/L and 83 g K/L). In order to avoid possible N losses by volatilization and K losses by leaching, doses were divided into three applications (at 15, 30 and 40 days after transplanting). Fertilization with macro (except N) and micronutrients were performed according to Malavolta [18], applying the following doses in mg dm<sup>-3</sup>: P = 100; K = 160; Ca = 230; Mg = 20; S = 155; B = 0.5; Cu = 1.5; Fe = 10; Mn = 4; Mo = 0.15 and Zn = 5.0. The sources of the nutrients used were as follows: simple superphosphate, KCl, MgSO<sub>4</sub>.7H<sub>2</sub>O, H<sub>3</sub>BO<sub>3</sub>, CuSO<sub>4</sub>.5H<sub>2</sub>O, Fe-EDTA, MnSO<sub>4</sub>.4H<sub>2</sub>O, ammonium molybdate and ZnSO<sub>4</sub>.7H<sub>2</sub>O. For potassium fertilization, the amounts of K supplied by potassium silicate were discounted to balance the nutrient doses between the treatments.

## 2.2 Evaluations

At the beginning of the flowering (45 days after transplanting-DAT), leaf transpiration rate (E), intercellular CO<sub>2</sub> concentration (Ci), stomatal conductance (gs), and net CO<sub>2</sub> assimilation rate

(A) were estimated. The evaluations were performed using infrared gas analyzer (IRGA) with a constant light source of 1,200 μmol of photons m<sup>-2</sup> s<sup>-1</sup>, starting the analysis at 7:00 p.m. employing two freshly ripened leaves per plant. On the same day and time, the leaf area index was also evaluated by photosynthetically active radiation method, using an equipment denominated ceptometer (AcupPAR model LP-80). These analyzes were performing in three replicates per plant.

Still at 45 DAT, plants were separated into roots, stems and leaves and placed in Kraft paper bags, identified with the respective treatments and dried in a forced-air oven (60°C) until constant weight to obtain root dry matter (RDM), stem (SDM), leaves (LDM) and by the sum all dry mass (TDM) was estimate. In these tissues, total nitrogen (total-N) concentration were analyzed according to Malavolta [19], nitric nitrogen (NO<sub>3</sub><sup>-</sup>-N) and ammonium nitrogen (NH<sub>4</sub><sup>+</sup>-N) were analyzed as described in Tedesco [20]. By multiplying the data of the concentration of the nitrogen fractions by the respective dry mass produced for each plant part, the total accumulated these nitrogen forms in these tissues were obtained. In the leaves, the silicon concentration were also determined according to Furlani & Gallo [21] using the colorimetric method of molybdenum blue.

## 2.3 Statistical Analysis

The data were subjected using the analysis of variance by F test (p <0.05). Mean values for the nitrogen and silicon doses were analyzed by polynomial regression at 5% probability. Statistical software Sisvar version 5.6 was used for data analysis [22].

## 3. RESULTS AND DISCUSSION

There was no significant interaction (p >0.05) between the factors nitrogen and silicon doses for any of the evaluated variables (Tables 1, 2, 3 and 4). However, the nitrogen doses significantly (p < 0.05) affected all the plant characteristics evaluated, while the silicon doses affected only some of these characteristics.

The doses of N promoted increases in transpiration rate photosynthetic rate and stomatal conductance and a decrease in internal CO<sub>2</sub> concentration (Table 1). This effect was also observed by Souza et al. [2]. The increase of the photosynthetic rate is due to a greater stomatal

opening, which increases the gas exchange and the stomatal conductance of the plant [23,24]. On the other hand, the decrease in the intercellular CO<sub>2</sub> concentration with increasing doses of N is perhaps, a response of the increase of the photosynthetic rate and stomatal conductance, decreasing the internal CO<sub>2</sub> [24]. Silicon doses not affected gas exchange in eggplant.

The increase of the photosynthetic rate is probably due to the N action in the composition of the chlorophyll molecules, in which the active sites responsible for photosynthesis are found [25,26], as observed by Zhang et al. [27]. At lower doses of N, probably occurred a decrease in the turgescence potential of foliar tissues, causing stomata closure, increased CO<sub>2</sub> diffusion resistance, and a consequent decrease in the photosynthetic rate [24,26].

The leaf area index (LAI), leaves dry mass (LDM), stems dry mass (SDM), roots dry mass (RDM) and total dry mass (TDM) (Table 2) adjusted to the quadratic regression model with N doses, where maximum values were obtained at the doses of 262, 427, 445 and 373 and mg dm<sup>-3</sup> of N, respectively. In other works, a positive effect of the N supply to the eggplant was observed on the growth [2,3]. The decrease in dry mass production at higher doses is probably due to the toxicity caused by excess N, which occurs mainly when it is supplied as urea [28]. The silicon doses also increases the LAI that adjusted to the quadratic regression model. This effect possibly is due improved of leaf

architecture and thus increased interception of sunlight, as reported by Ávila et al. [12].

The LAI measures the relation of the area covered by the leaves in relation to the area of the soil occupied by the plant and is related to the leaf expansion and consequently to the vegetative growth of the leaves. Thus the photosynthetic rate is generally positively related to this variable [29]. The lowest LAI values as well as LDM, SDM and RDM in the lowest doses of N are due to the N requirement for the plant, a fact related to the role of this nutrient on the cellular division and expansion [6].

All nitrogen fractions concentrations were positively affected by N doses (Table 3). N-ammonium concentrations in the plant tissues were higher in the leaves while nitrate was concentrated mainly in the stem and root. Alves et al. [30] observed higher levels of N-nitrate in the sunflower in stem in relation to the leaves. In addition, the ammonium is absorbed by the roots and is almost all assimilated in these tissues [30]. In turn, N-total concentration in all plant tissues were linearly elevated as a function of the N rates applied in accordance with the N-mineral fractions. The highest levels of these N fractions were observed in leaf tissues, a fact that is justified by the composition of these tissues, which are rich in chlorophylls and several nitrogen compounds, such as amino acids and proteins, since they are the main tissues of assimilation of N [31]. In relation to silicon doses,

**Table 1. Intercellular CO<sub>2</sub> concentration (Ci), transpiration rate (E), stomatal conductance (gs) and photosynthesis rate (A), in eggplant leaves as function of nitrogen and silicon doses**

Nitrogen doses (mg dm <sup>-3</sup> )	Ci (mg L <sup>-1</sup> )	E (mmol m <sup>-2</sup> s <sup>-1</sup> )	gs (mmol m <sup>-2</sup> s <sup>-1</sup> )	A(μmol m <sup>-2</sup> s <sup>-1</sup> )
25	293.84	4.33	0.32	8.91
125	276.63	4.11	0.33	11.68
250	261.03	4.76	0.40	14.29
350	237.81	4.62	0.37	16.02
500	247.00	5.00	0.44	16.81
β <sub>1</sub>	**	*	*	*
β <sub>2</sub>	**	-	-	**
Silicon doses (mg dm <sup>-3</sup> )				
0	266.03	4.63	0.37	13.35
75	266.03	4.41	0.38	13.72
150	254.48	4.62	0.36	13.93
200	266.53	4.60	0.37	13.18
β <sub>1</sub>	ns	ns	ns	ns
β <sub>2</sub>	ns	ns	ns	ns

β<sub>1</sub>, β<sub>2</sub> -Linear and quadratic parameters, respectively, of the adjusted functions: linear ( $y = \beta_0 + \beta_1.x$ ) and quadratic ( $y = \beta_0 + \beta_1.x + \beta_2.x^2$ ). \*\* P<0.01; \* P<0.05; ns P>0.05

**Table 2. Leaf area index (LAI), leaf dry mass (LDM), stem dry mass (SDM), root dry mass (RDM), in eggplant plants under nitrogen and silicon doses**

Nitrogen doses(mg dm <sup>-3</sup> )	LAI	LDM	SDM	RDM	TDM
	-----g per plant-----				
25	0.50	5.05	3.18	3.34	11.58
125	1.43	9.63	6.57	5.73	21.94
250	1.88	11.44	7.52	6.56	25.51
350	2.12	12.08	7.49	5.96	25.52
500	1.75	12.83	8.01	5.73	26.56
$\beta_1$	**	**	*	**	
$\beta_2$	**	**	**	**	
Silicon doses(mg dm <sup>-3</sup> )					
0	1.43	10.09	6.82	5.46	22.38
75	1.63	10.29	6.44	5.47	22.20
150	1.56	10.47	6.38	5.60	22.45
200	1.52	9.96	6.57	5.33	21.86
$\beta_1$	*	ns	ns	ns	ns
$\beta_2$	*	ns	ns	ns	ns

$\beta_1, \beta_2$  -Linear and quadratic parameters, respectively, of the adjusted functions: linear ( $y = \beta_0 + \beta_1.x$ ) and quadratic ( $y = \beta_0 + \beta_1.x + \beta_2.x^2$ ). \*\*  $P < 0.01$ ; \*  $P < 0.05$ ; <sup>ns</sup>  $P > 0.05$

**Table 3. Ammonium (NH<sub>4</sub><sup>+</sup>-N), nitrate (NO<sub>3</sub><sup>-</sup>-N) total nitrogen concentration in leaves of eggplant plants under nitrogen and silicon doses**

Nitrogen doses(mg dm <sup>-3</sup> )	NH <sub>4</sub> <sup>+</sup> -N			NO <sub>3</sub> <sup>-</sup> -N			Total-N		
	Leaf	Stem	Root	Leaf	Stem	Root	Leaf	Stem	Root
	-----mg kg <sup>-1</sup> -----								
25	0.93	0.21	0.17	0.13	0.08	0.25	14.56	7.00	9.69
125	1.00	0.24	0.23	0.12	0.11	0.18	23.31	9.06	12.81
250	1.49	0.31	0.41	0.08	0.19	0.13	32.31	13.50	19.31
350	1.88	0.63	0.73	0.13	0.62	0.32	39.44	18.69	21.06
500	2.19	1.19	0.86	0.26	0.98	0.83	46.94	24.25	22.50
$\beta_1$	**	**	**	**	**	**	**	**	**
$\beta_2$	**	**	**	**	**	**	**	**	**
Silicon doses(mg dm <sup>-3</sup> )									
0	1.60	0.50	0.50	0.14	0.44	0.36	32.40	14.80	17.10
75	1.55	0.51	0.45	0.12	0.36	0.31	30.15	14.00	17.20
150	1.38	0.54	0.50	0.17	0.41	0.24	30.95	14.95	16.85
200	1.46	0.50	0.47	0.14	0.37	0.45	31.75	14.25	17.15
$\beta_1$	ns	ns	ns	ns	ns	*	ns	ns	ns
$\beta_2$	ns	ns	ns	ns	ns	*	ns	ns	ns

$\beta_1, \beta_2$  -Linear and quadratic parameters, respectively, of the adjusted functions: linear ( $y = \beta_0 + \beta_1.x$ ) and quadratic ( $y = \beta_0 + \beta_1.x + \beta_2.x^2$ ). \*\*  $P < 0.01$ ; \*  $P < 0.05$ ; <sup>ns</sup>  $P > 0.05$

there was effect solely for the nitrate-N concentration in the roots, which decreased in the doses of 75 and 150 mgdm<sup>-3</sup> and increased in the highest dose. Similar results were obtained in rice by Ávila et al. [12] and indicates that possibly the silicon decrease the translocation of N-nitrate to the aerial part or increased its assimilation in the roots.

All the N fractions accumulated in to dry mass of eggplant increased linearly with the N doses and were higher aerial part (leaves + stem) (Table 4).

These results reflect the combination of the dry mass production and the higher concentration of the N fractions in the leaves and the stem. The shoot accumulated more ammonium-N than nitrate-N, whereas in the roots, the values for both nitrogen forms were similar. In general, plants tend to accumulate more nitrate than ammonium in tissues, depending on the form of nitrogen supplied [28]. The higher accumulation of N-ammonium in the aerial part in relation to the N-nitrate may have been a consequence of the use of urea as source of N.

**Table 4. Ammonium (NH<sub>4</sub><sup>+</sup>-N), nitrate (NO<sub>3</sub><sup>-</sup>-N), and total nitrogen accumulation in shoot and root, nitrogen use efficiency (NUE) and silicon content in leaves in eggplant under nitrogen and silicon doses**

Nitrogen doses(mg dm <sup>-3</sup> )	NH <sub>4</sub> <sup>+</sup> -N		NO <sub>3</sub> <sup>-</sup> -N		Total-N		NUE	Leaf Si
	Shoot	Root	Shoot	Root	Shoot	Root		
	-----mg per plant-----						g <sup>2</sup> /mg N	gkg <sup>-1</sup>
25	5.34	0.57	0.78	0.83	96.59	32.45	1.06	5.78
125	11.15	1.33	1.75	0.99	285.18	73.00	1.36	5.93
250	19.19	2.63	2.25	0.83	474.13	126.59	1.10	3.73
350	27.25	4.34	5.88	1.90	621.65	126.82	0.88	4.32
500	37.53	4.95	10.50	4.80	808.12	129.26	0.76	4.85
$\beta_1$	**	**	**	**	**	*	**	*
$\beta_2$	-	-	-	-	-	-	*	*
Silicon doses(mg dm <sup>-3</sup> )								
0	19.56	2.75	4.43	1.97	427.95	93.41	1.01	4.50
75	19.29	2.46	3.59	1.68	400.58	94.01	1.06	4.16
150	17.98	2.80	4.39	1.37	419.54	94.35	1.06	5.42
200	17.79	2.49	3.79	2.41	409.86	91.37	1.00	5.61
$\beta_1$	ns	ns	ns	*	ns	ns	ns	*
$\beta_2$	ns	ns	ns	*	ns	ns	ns	-

$\beta_1, \beta_2$  -Linear and quadratic parameters, respectively, of the adjusted functions: linear ( $y = \beta_0 + \beta_1.x$ ) and quadratic ( $y = \beta_0 + \beta_1.x + \beta_2.x^2$ ). \*\*  $P < 0.01$ ; \*  $P < 0.05$ ; <sup>ns</sup>  $P > 0.05$

The fact that N doses gave a linear response for total N, and quadratic for dry mass, indicates that the plants did not respond in growth in the same proportion as the amount of N accumulated by the plant, that is, above the maximum point there was accumulation of unmetabolized or mineral nitrogen [7] with increasing doses of N. The NUE values decreased with N rates as a result of decreasing dry mass production at the higher N doses. Leaf content of Si decreased with N doses, possibly due to "dilution" caused by increasing leaf dry matter production due to increasing doses of N [14] or due to the lower absorption of Si. Wu et al. [32] studying the interaction between N and Si in rice, observed that in the highest doses of N tested, there was a decrease in leaf concentration of Si and that this decrease was associated with the decrease of the expression of the OsLsi1 and OsLsi2 genes, which encode for the synthesis of Si transporters.

The Si doses influenced only the nitrate-N accumulation in the roots and the silicon concentration in the leaves. The leaf content of Si obtained in the highest dose of silicon was equivalent to 11.9 g/kg of SiO<sub>2</sub> in the mass and is within the range of contents generally observed for dicotyledonous species that is 10 to 30 g/kg of SiO<sub>2</sub> [19].

#### 4. CONCLUSION

The nitrogen supply increased the photosynthetic rate and all growth components of the eggplant,

especially the dry mass of leaves and stem and decreased the silicon leaf concentrations.

The eggplant accumulated more ammonium in the leaves and the stem, while the nitrate was accumulated more in the roots.

Silicon did not interfere with eggplant dry mass production, but increased leaf area index, decreased nitrate levels and accumulations in the roots at lower doses of this element.

#### COMPETING INTERESTS

Authors have declared that no competing interests exist.

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