Nitrogen Fertilization on Maize Sowing: Plant Growth and Seed Vigor

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Abstract

The management of fertilization in maize plants is a preponderant factor in the search for higher yields and obtaining quality seeds. In this sense, the study of nitrogen (N) management is predominant in improving the efficiency of its application. The objective of this study was to evaluate the growth, partition assimilation and vigor expression of maize seeds submitted at different doses of nitrogen applied at sowing. The treatments were constituted by different doses of nitrogen fertilization at sowing, being: 25%, 50%, 75% and 100% of the recommended dose for the crop, according to the recommendation of nitrogen fertilization. The plants were collected at regular intervals of 21 days after sowing until the end of the growth cycle. For the analysis of growth and partition of dry matter between the organs, the accumulation of dry matter and leaf area was measured. The experimental design was randomized blocks in a 4×7 factorial scheme (four N rates at sowing and seven collection times with three replications). The maize plants under the influence of nitrogen fertilization at sowing presented differential growth and distinct responses in the assimilated partition between their structures, with the intensity dependent on the dose used. Plants grown using 75% of the recommended dose of N at sowing had a higher relative growth rate, with more pronounced effects compared to the other doses. The expression of maize seed vigor increases to an increase of 82.5% N at sowing.

Keywords

Emergence Speed, Net Assimilation Rates, Relative Growth Rates, *Zea mays* L., Vigor

1. Introduction

Maize (*Zea mays*) is a plant whose origin is believed to have occurred in Mexico and was later spread by pre-Columbian peoples to the other continents [1]. The productivity of this cereal is dependent on the cultivar, soil, fertilization, climate, cultural practices, pests and diseases [2] [3]. Therefore, it is important and necessary to develop studies in an attempt to solve the various problems that involve these factors, seeking high outputs with high quality.

With regard to cultural practices, the management of fertilization is a preponderant factor in the search for higher yields and the obtaining of quality seeds. In this sense, nitrogen (N) management has been one of the most widely studied agricultural practices in the search to improve its efficiency of use. Different concentrations of nutrients affect the growth of cereals [4] [5], while an adequate supply of these elements favors the productivity of the maize [6]. N is one of the main plant nutrients, being required in large quantities [7].

The proper application of nutrients to the soil is an important factor that interferes with maize yield, microorganism activity and soil quality improvement. The proper application of nutrients to the soil is an important factor that interferes with crop yield, microorganism activity and soil quality improvement. This necessity exists because most of the N in the soil is in organic combinations, and this form is unavailable to the plants and, in general, the soil does not fulfill the demand of the crop in terms of N [8]. Therefore, to obtain higher yields of maize it is necessary to apply nitrogenous fertilizers.

Nitrogen is important in plant metabolism when participating in several metabolic routes of great importance to plants [9]. Research indicates that high concentrations of N in the root zone are beneficial in promoting rapid initial plant growth and increased grain yield [10]. In addition, the supply of N in the early stages favors a higher absorption of phosphorus [11], thereby facilitating the growth of plants.

Growth analysis is a technique that allows the primary and physiological bases of production to be determined, describing at plant differential responses of different time intervals that are related to morphophysiological conditions under different environmental and management conditions [12]. In turn, vigor is a component of the physiological quality of seeds related to the reorganization of the cell membrane system, hydrolysis, translocation and reserve allocation, and can be determined from the emergence of seedlings [13]. Therefore, the joint evaluation of the growth and expression of seed vigor makes it possible to evaluate the level of influence imposed by the factor studied.

In this sense, the objective of this research was to analyze comparatively the growth of plants and the vigor of maize seeds produced under the influence of different doses of nitrogen at sowing.

2. Materials and Methods

The experiment was conducted in the experimental area of the Department of

Plant Sciences of the Federal University of Pelotas—Capão do Leão, RS (31˚52'S and 52˚21'W). The determination of seed vigor was carried out in a germination chamber which control temperature and relative humidity. For the establishment of the crop, seeds of the yellow maize genotype were used, with a germination of 95%.

The sowing was done manually in January 2017, in soil classified as Planosols Hydromorphic. The spacing between lines and between plants was 45 and 36 centimeters, respectively, resulting in a population density of 55,000 ha^{-1} plants [14].

Soil correction was performed according to previous analysis and according to the recommendation of the Manual of Fertilization and Liming for the Rio Grande do Sul and Santa Catarina States [15]. Each plot consisted of four rows of 6 meters in length, and the two rows were considered for the plot area.

The soil of the experiment site is classified as Solubic Eucalyptus Haplic Planosol, belonging to the Pelotas mapping unit [16]. The results of the soil analysis showed the following chemical and physical characteristics: pH (H_2O): 5.0; P: 46.0 mg·dm^{-3}; K: 53 mg·dm^{-3}; S: 19.8 mg·dm^{-3}; Ca: 3.2 cmolc·dm^{-3}; Mg: 0.9 cmolc·dm^{-3}; Al: 0.8 cmolc·dm^{-3}; B: 0.0 mg·dm^{-3}; Cu: 1.4 mg·dm^{-3}; Zn: 1.8 mg·dm^{-3}; Mn: 14.2 mg·dm^{-3}; Na: 50 mg·dm^{-3}; CTC: 5.3 cmolc·dm^{-3}; base saturation: 45%; organic matter: 2.07%; clay: 19%. During the experiment, temperature, solar radiation, relative air humidity and rainfall values were recorded (Figure 1).

The treatments comprised different doses of nitrogen fertilization at sowing, those being 25%, 50%, 75% and 100% of the recommended dose of the crop, which was 30 kg N ha^{-1}, in accordance with the nitrogen fertilization recommendation for maize in Rio Grande do Sul based on the soil organic matter content, predecessor crop and yield expectancy [15]. The source of N was urea ($CO(NH_2)_2$). Complementary fertilization with a fixed dose was performed in the breeding stage.

For growth assessments, successive collections of three plants per plot were made at regular intervals of 21 days after sowing (DAS), throughout the development cycle, totally seven collection seasons. In each collection, the plants were separated into different structures (aerial part, root, stem, tassel and spikes), and the roots were washed using a fine-mesh sieve. The different structures were conditioned separately in brown paper envelopes and subjected to oven drying with forced ventilation at a temperature of 70˚C ± 2˚C until reaching a constant mass.

The leaf area (Af) was determined with the aid of an area meter, model LI3100, and the leaf area index (L) calculated using the equation L = Af/St, where St corresponds to the superficial area occupied by the plant. The total dry matter data over time (Wt) was adjusted using the simple logistic equation Wt = Wm/(1 + Ae-Bt), where Wm is the asymptotic maximum growth estimate, "A" and "B", "And" the natural basis of Napierian logarithm and "t" the time in days
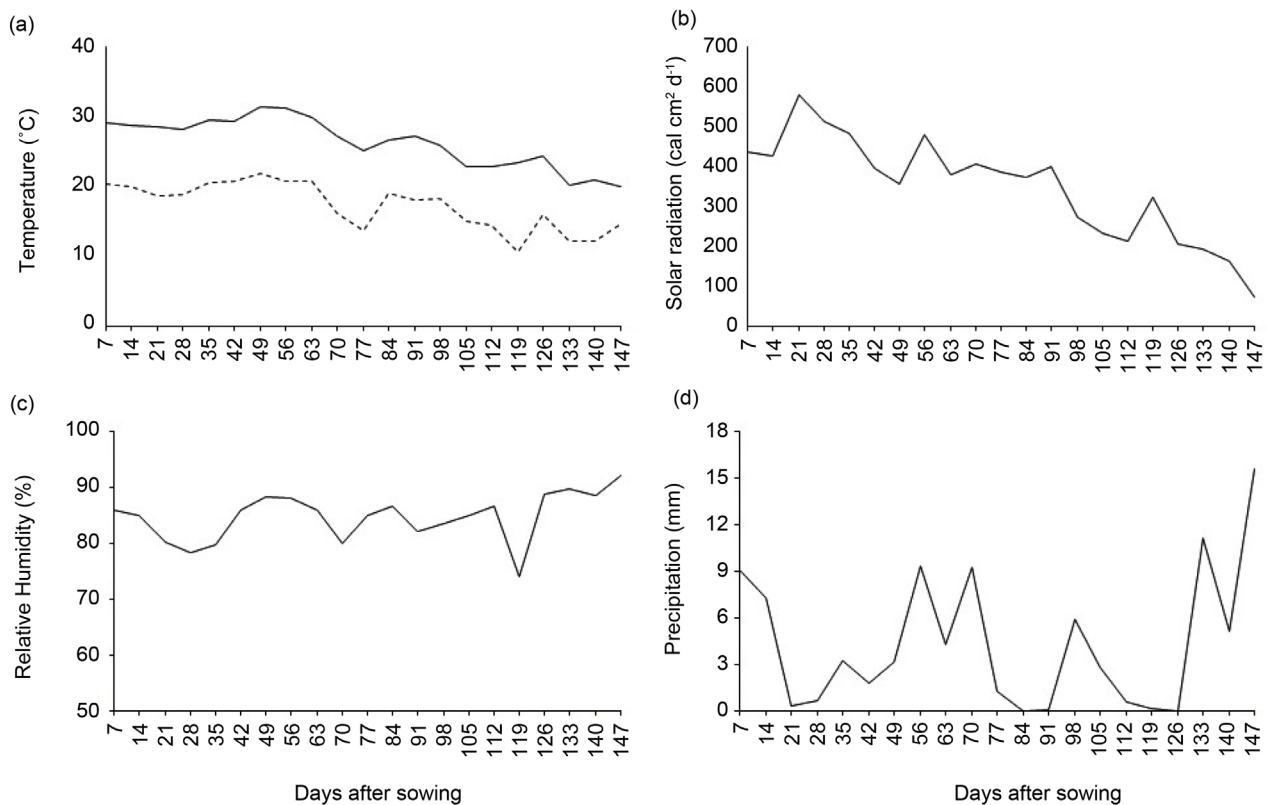
Figure 1. Maximum (——) and minimum (– – –) temperatures (a), solar radiation (b), relative humidity (c) and precipitation (d).

after sowing [17]. Leaf area data were adjusted by means of orthogonal polynomials [17] and the instantaneous values of the dry matter production rate (Ct) obtained by the derivatives of the adjusted equations of total dry matter (Wt) with respect to time [17].

For determining the instantaneous values of the relative growth rate (Rw), the equation Rw = 1/Wt·dw/dt was used. The instantaneous values of the net assimilation rate (Ea), leaf area ratio (Fa) and leaf mass ratio (Fw) were estimated using the following equations: Ea = 1/Af·dw/dt, cFa = Af/Wt and Fw = Wf/Wt, according to RADFORD [18].

Harvest index (Hi) was determined by the equation Hi = Ws/Wt, where Ws corresponds to the dry seed mass and Wt is the total dry matter of the plant. The conversion efficiency of solar energy (ξ) was determined using the equation ξ (%) = (100·Ct·δ)/Rs, where Rs is the mean value of incident solar radiation (cal m^{-2} day^{-1}) to the corresponding Ct and δ calorific value of 4515 cal g^{-1} according to Protásio *et al.* [19].

The vigor of the maize seeds was determined from the emergence of seedlings in soil, and the test was performed with eight replications. For this, the seeds were arranged in trays of polyethylene, containing A1 horizon from Planossolo Háplico Eutrofico Solódico. Seedling emergence had stabilized 10 days after sowing, and the percentage of seedlings that had emerged was as described by

Nakagawa [20]. Together with the emergence of seedlings in soil, the emergence velocity was evaluated from the daily number of emerged seedlings until stabilization. The counts were performed until a constant number of seedlings emerged, and the results were obtained in accordance with Nakagawa [20].

A mass of 1000 seeds was determined by a mass of 100 seeds from the experimental plot area, counted at random with the aid of a manual counter, then measured using a precision scale, using eight subsamples per plot.

The experimental design was a randomized complete block design in a 4×7 factorial scheme (four N rates at soil seeding and seven collection times with three replications). The total dry matter data were analyzed by the simple logistic equation and the discussion was based on the trend of the growth curves [12]. The vigor data were submitted to a randomized experimental design with eight replications. Assimilated partition data were converted to percentage of dry matter allocated in each evaluated plant structure, and those referring to primary growth data were submitted to analysis of variance. The data related to vigor when significant at 5% probability were represented by orthogonal polynomials, using the WinSTAT statistics software.

3. Results and Discussion

A significant interaction was observed at the probability level of 5% for the primary data of leaf area, dry matter of stalk, dry matter of roots and dry matter of ears of maize plants submitted to the influence of different doses of nitrogen (N) at sowing. The production of total dry matter (Wt) in maize plants under different N rates at sowing presented a logistic tendency with a high coefficient of determination ($R^2 \geq 0.87$). Maize plants presented slow growth until 42 days after sowing (DAS), with rapid growth after this period, presenting a maximum growth at 147 DAS (Figure 2(a), Table 1).

Table 1. Summary of variance analysis with mean squares for primary leaf area (Af), leaf dry matter (Wf), stem (Wc), root (Wr) and ear (Wesp) dry matter. Capão do Leão, UF-Pel, 2017.

FV	DF	Medium Squares				
		A_f	W_f	W_c	W_r	W_{esp}
Doses (D)	3	4,773,625*	669.29*	98.15016*	1544.98*	368.11*
Seasons (S)	6	1.66E007*	3547.95*	1904.801*	2105.82*	32,448.07*
Block	2	22,328.16	2.15	2.030781	0.3399	16.29
D x E	18	701,411.90*	85.33*	63.64213*	291.26*	137.16*
Residue	54	14,770.11	0.9600	1.368481	0.7865	5.66
Total	83	-	-	-	-	-
Average		2128.68	28.59	18.11	18.79	31,93
CV (%)		5.7	3.42	6,45	4.71	7.45

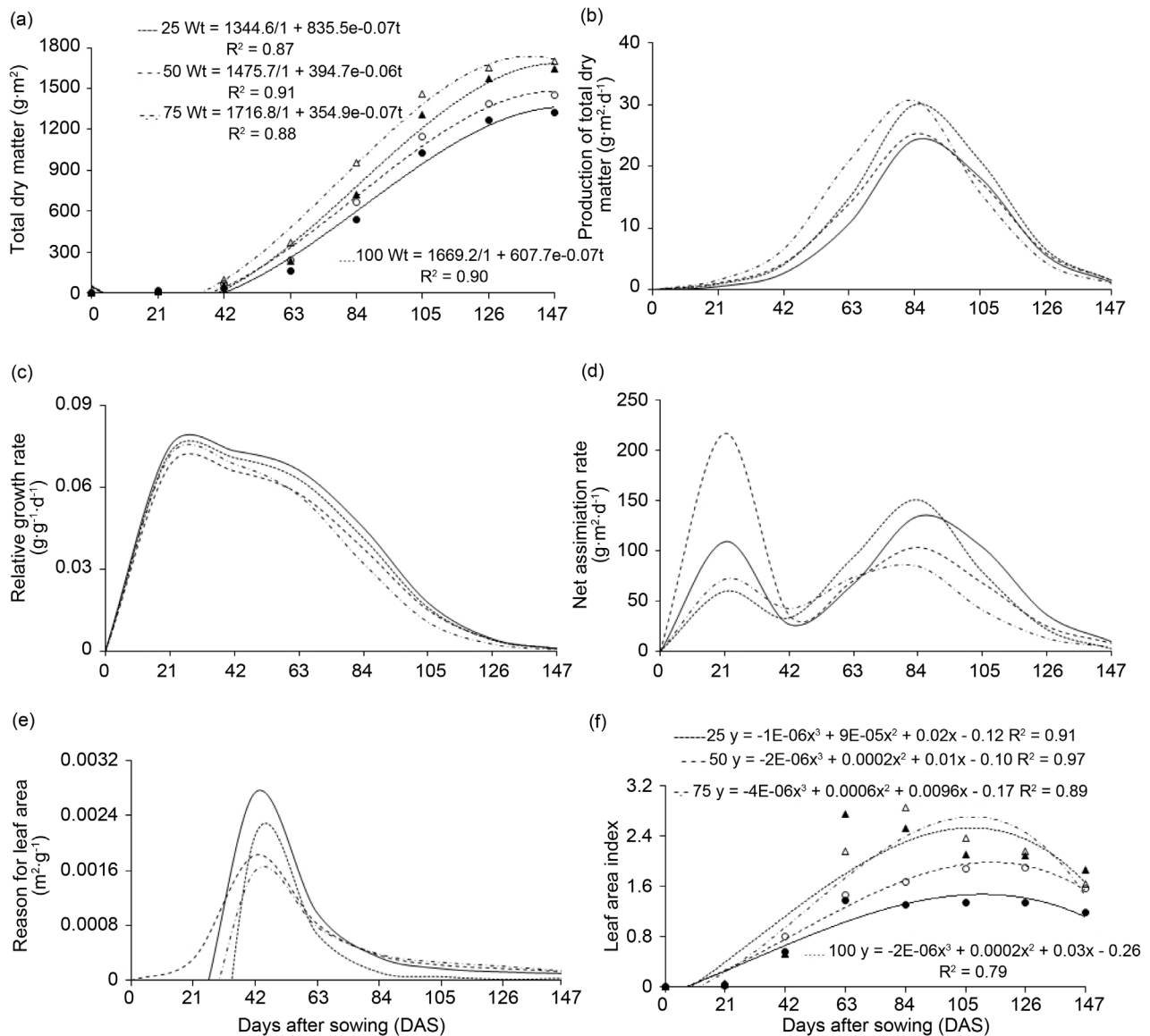Level of significance (*p = 5%) and not significant (ns).

Figure 2. Total dry matter (a), dry matter production rate (b), relative growth rate (c), net assimilation rate (d), leaf area index (e) and leaf area ratio of maize plants (*Z. mays* L.) submitted to different doses of N at sowing. 25% of the recommended dose (——), 50% of the recommended dose (– – –), 75% of the recommended dose (· – ·) and 100% of the recommended dose (······) for the culture.

The use of N doses at sowing increased Wt at doses of 75%, 100%, 50% and 25% N, reaching 1702.87, 1647.99, 1453.19 and 1327.37 g·m^{-2}, respectively. The differences between dry matter accumulations of 75% and 100% doses in relation to the others may be related to the higher initial availability of N for growth, while the doses of 5% and 25% were not sufficient for large dry matter additions.

The higher accumulation of dry matter obtained in the 75% dose can be related to the high net assimilated production [21] by the maize plants, since the N in the plant acts by participating in the molecules of organic compounds. In addition, the negative effect of a high N dose on sowing on initial plant performance may be a consequence of the phytotoxic effect of high doses of fertilizer

on seedling emergence [22] [23].

The maximum dry matter (Ct) production rate was reached at approximately 84 DAS for the N rates at sowing (Figure 1(b)). Plants at 75% N had higher Ct than plants under 100%, 50% and 25% N, showing greater efficiency in the increase of dry matter per unit area in relation to time. Similar trends were found by Pedó *et al.* [24] in rye plants and by Koch *et al.* [25] in wheat.

The increase in the dry matter production rate can be attributed to the increase in the leaf area and the amount of assimilates synthesized, destined to the growth and the vegetal development, due to the greater availability of the element in the initial stages in pepper [26]. On the other hand, the subsequent decrease in Ct is related to the increase in plant age and nonphotosynthetic tissues in beans [27].

The relative growth rate (Rw) of maize plants under different N rates at sowing showed maximum values at 21 DAS, with a subsequent decrease up to 147 DAS (Figure 2(c)). The maximum values of Rw were obtained by plants at 25% N, followed by doses of 100%, 75% and 50% N, demonstrating that increasing N rates at sowing influenced Rw. According to Aumonde *et al.* [21], the high initial Rw can be attributed to the high photosynthetic capacity of the leaves at the beginning of plant growth, and according to Lopes *et al.* [28] its decrease is due to auto-shading.

The growth of the plant in terms of volume increase, dry mass, linear dimensions and structural units is a function of the storage of structural carbon [21]. In this sense, the increase in initial growth may be associated with greater availability of the element in the early stages of cultivation. The net assimilation rate (Ea) in maize plants submitted to different doses of N at sowing presented the best results with 50% of the N dose recommended at 21 DAS in relation to the other doses (Figure 2(d)). At 21 DAS, the highest values were found at doses of 50%, 25%, 75% and 100%, at the beginning of the vegetative growth, with a subsequent decrease at these doses. At 84 DAS the second peak was found for doses of 100%, 25%, 50% and 75 %, respectively. According to Gondim *et al.* [29], the Ea tends to be higher at the beginning of the cycle when auto-shading is low and, according to Lopes *et al.* [28], the second peak is attributed to the beginning of the reproductive phase of the plants.

It is possible that plants at the highest N doses reached the greatest difference between crude photosynthesis and respiration, resulting in higher total dry matter production. In this sense, the highest Ea are related to the increase of the efficiency of the leaf area for photoassimilate production, with the second Ea peak being due to the emergence of the plant reproductive structures [30]. Ea curves showing two peaks during plant ontogeny have been described by Silva *et al.* [31] in barley, Falqueto *et al.* [32] in rice and Pedó *et al.* [24] in rye.

The leaf area index (L) was adjusted to the cubic model, with a high coefficient of determination ($R^2 \geq 0.92$). At the beginning of the growth the L was reduced and reached the maximum near 105 DAS (Figure 2(e)) with a subsequent

decrease. The highest values of L were obtained by maize plants at the 75% dose followed by 100%, 50% and 25% doses. Aumonde *et al.* [21] reported that the decrease in L at the end of the cycle is a result of foliar senescence. The L is a growth variable that indicates the leaf surface in relation to the soil surface, and according to Peixoto and Peixoto [33], L is responsible for the interception of solar energy.

It can be seen that the application of N at sowing exerted a positive influence on L, providing an increase in the area of the soil occupied by leaves. An adequate supply of N is considered one of the main factors in the maize crop, since this nutrient exerts an important function in the biochemical processes of the plant. In addition, it affects leaf initiation and expansion rates, leaf size and leaf senescence intensity [34].

The maximum leaf area ratio (Fa) was reached at 42 DAS for all N rates, with subsequent decreases up to 147 DAS (**Figure 2(f)**). The highest values were obtained at the dose of 25% followed by the doses of 100%, 50% and 75%. According to Peixoto and Peixoto [33], the decrease in Fa is due to self-shading, caused by the upper leaves above the lower ones [35]. A similar trend was found by Koch *et al.* [25] when evaluating the effect of growth regulator doses on wheat plants under high nitrogen fertilization rates.

The leaf mass ratio (Fw) of maize plants was high at 42 DAS for both N rates at sowing. The 100% N dose presented the highest leaf dry matter allocation at 42 DAS, followed by 50%, 75% and 25% N at sowing (**Figure 3(a)**), with a subsequent decrease at all doses. The use of 100% N at sowing reflected an accumulation of approximately 68% more leaf dry matter, compared to the 25% N dose at sowing.

The conversion efficiency of solar energy (ξ) showed increasing peaks up to 105 DAS (**Figure 3(b)**), with maximum values of 3.55%, 3.40%, 3.72% and 4.06% for plants at 25%, 50%, 75% and 100% N at sowing, respectively, with a subsequent decline to 147 DAS. ξ presented a high peak at 84 DAS, together with the maximum obtained at Ct (**Figure 2(b)**). Up to 84 DAS, the dose of 75% of N presented greater ξ than the other doses, whereas at 105 DAS the highest ξ was obtained at doses of 100% N at sowing. ξ is a variable that relates the production of dry matter in energy units [36], reflecting the highest total dry matter allocation at 75% N at sowing.

Increasing the dose of N applied, most of the time, provides an increase in the structures and increases the yield of the crop [37]. Nitrogen fertilization in maize showed a positive effect on leaf area index and the number of seeds/ears at plant height, as well as on biomass yield [38]. Similar trends of ξ were found in works with beans by Reyes-Cuesta *et al.* [27], with soybean by Melges *et al.* [39], Marenco and Lopes [40] and with wheat by Koch *et al.* [25].

There was similarity in the dry matter partition between the 50% and 75% N rates in the sowing of maize plants (**Figure 3(d)**, **Figure 3(e)**). However, 75% N plants show less leaf dry matter allocation and higher roots in the range of 63 to
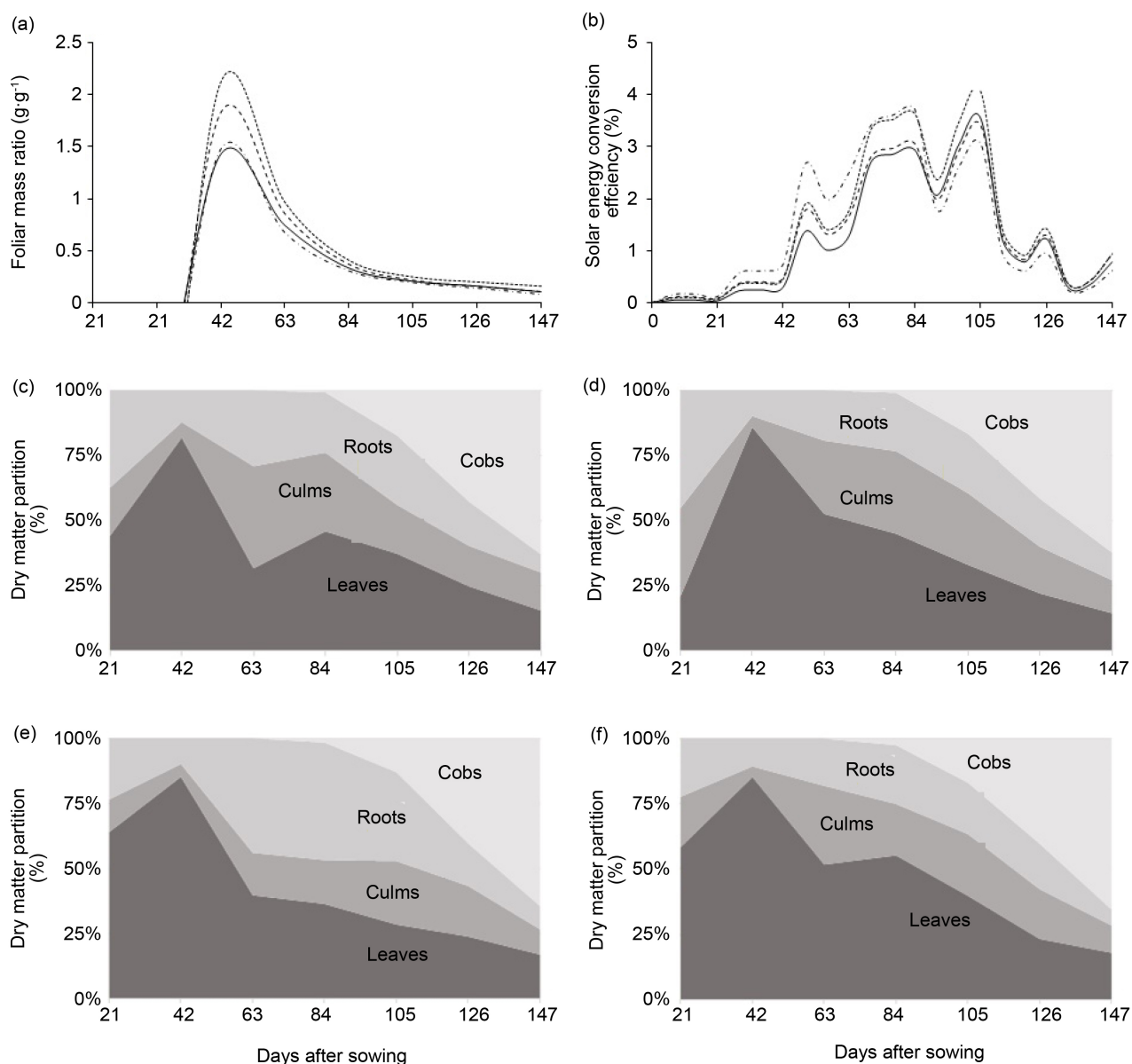
Figure 3. Foliar mass ratio (a), conversion efficiency of solar energy (b) of maize plants (*Z. mays* L.) submitted to different doses of N at sowing. 25% of the recommended dose (———), 50% of the recommended dose (– – –), 75% of the recommended dose (⋅ ━ ⋅) and 100% of the recommended dose (∙∙∙∙∙) for the culture. And dry matter partition of different structures of maize plants (*Z. mays* L.) submitted to different doses of N at sowing. 25% of the recommended dose (c), 50% of the recommended dose (d), 75% of the recommended dose (e) and 100% of the recommended dose (f) for the culture.

105 DAS than 50% N roots. Although nutritional requirements are lower in the early stages [10]. In addition, the high N concentrations in the root zone are beneficial in promoting rapid initial plant growth and root growth [10] [41].

The use of the 100% N dose at sowing increased the leaf dry matter partition up to 84 DAS (**Figure 3(f)**), while reducing the root partition. It is worth noting that, due to the increase in the dry matter partition in leaves during the development cycle, the accumulation in the other structures was altered, which can cause structural problems and the lodging of maize plants along the ontogeny

[42]. The tests detected significant differences in seed physiological quality, and the emergence and emergence speed of maize seedlings from seeds produced under different N rates (**Figure 4**).

Seedling emergence tended to increase with increasing doses of N (**Figure 4(a)**), adjusting in a quadratic model, with a determination coefficient of $R^2 \geq 0.70$. Thus, the expression of maize seed vigor was positively influenced by the N increase. At doses higher than the point of maximum technical efficiency (82.5%) there was a tendency for the emergence to reduce. At the emergence speed, the maximum technical efficiency is 58% of the dose of N, which presents the fastest emergence of seedlings (**Figure 4(b)**). The highest quality at these doses probably occurs because nitrogen fertilization interferes with protein content and may affect seed quality [43], since the reserve proteins are hydrolyzed during the germination of the seeds to supply (N, sulfur and carbon skeletons the embryonic axis and the seedlings during the early stages of development) [44].

The harvest index (Hi) was adjusted to the quadratic model, with a determination coefficient of $R^2 \geq 0.95$. The Hi indicates higher carbon allocation in the seeds in relation to the total dry matter of the plants. From the 50% N dose at sowing the Hi was elevated (**Figure 4(c)**). In this sense, the carbon allocation is related to the availability of N for the physiological and biochemical processes in the early stages of the crop. Similar results were obtained by Gava *et al.* [45] in maize, with an increase of N in the highest N rates, evidencing that there was a
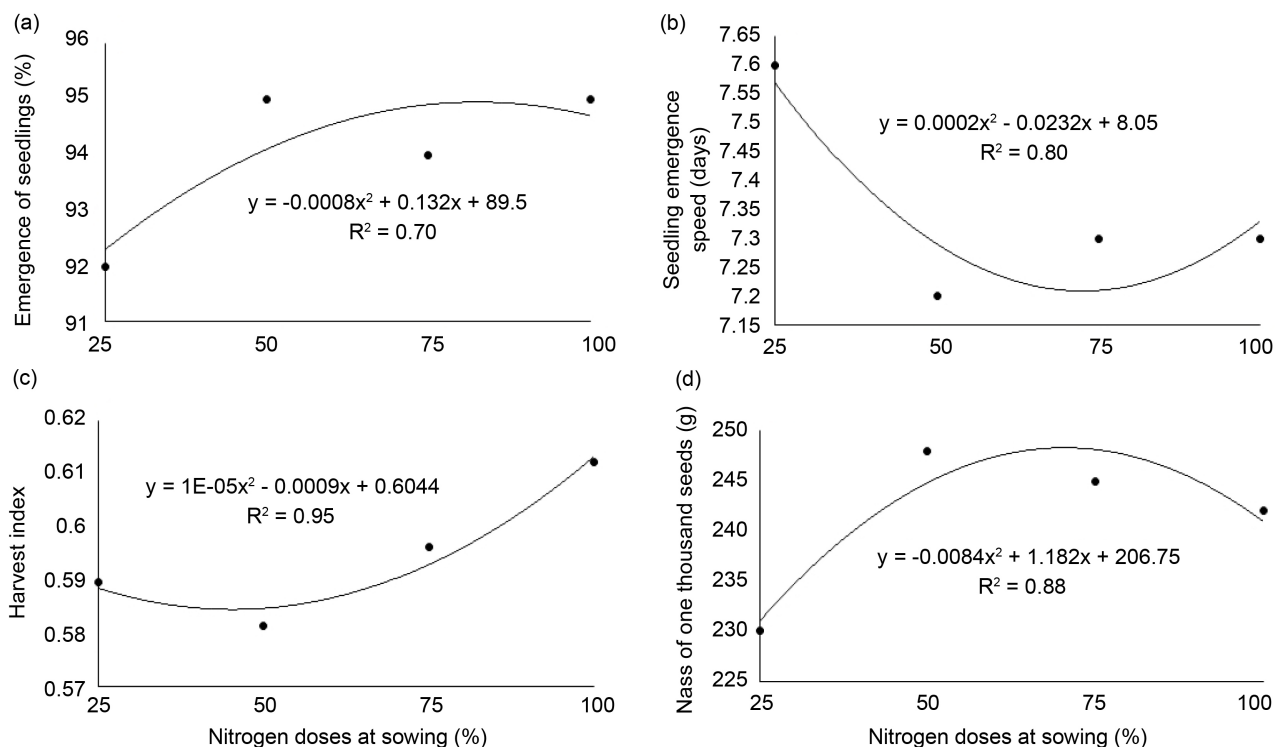

Figure 4. Emergence (a), seedling emergence velocity (b), crop index (c) and mass of one thousand maize (*Z. mays* L.) seeds originated from plants submitted to different doses of N in seeding.

good translocation of photoassimilates from the leaves to the grains, that is, the efficiency of the conversion of phytomass to grains was satisfactory.

The use of this index allows us to identify the ability of a genotype to combine high total production capacity and to allocate the accumulated dry matter to components of economic interest, in this case grain/seed production [46]. According to the same authors, the grain harvest index can vary from 0.60 for cultivars of high productivity to 0.10 for cultivars of low productivity. Consequently, the results of Hi obtained demonstrate an increase in total production as the dose of N fertilization in the base increases.

The mass of 1000 seeds responded to the application of N doses adjusting to the quadratic function (**Figure 4(d)**), increasing to the point of maximum technical efficiency (70.36% of the N dose) with a subsequent reduction. According to Escosteguy *et al.* [47], N determines the development of maize plants with a significant increase in leaf area and dry matter production, resulting in higher grain yield. This is confirmed by observing the relationship between the application of N and the mass of 1000 seeds observed in **Figure 4(d)**. Similar results were observed by Queiroz *et al.* [48] and Maestrelo *et al.* [7].

The reduction of the mass of 1,000 seeds from the point of maximum technical efficiency is possibly related to the fact that an increase in the accumulation of dry phytomass occurs, thereby increasing the maintenance breath and, consequently, reducing the translocation of photoassimilates to the grains. In general, the dry matter yield of a crop is closely related to L and Ea [39]. The maximum Ct is related to the maximum ξ (**Figure 2(a)**). However, the Ea in maize plants submitted to different doses of N at sowing presented the best results with 50% of the recommended dose, being related to L and together with the sending of photoassimilates to reproductive organs [28].

It was possible to demonstrate that in maize plants under the rate of 75% of N at sowing, they obtained higher Wt and Ct, while at the dose of 100% of N a higher Fw was shown. According to Aumonde *et al.* [21], Fw provides an estimate of the fraction of assimilates retained in the leaves and not exported to the other organs of the plant, reflecting the mass and vigor of the seeds with the highest N rates at sowing. In this sense, the reduction of the percentage of dry matter in roots under the effect of doses of N possibly occurred as a result of the increase of greater allocation of dry matter in the reproductive structures, aimed at greater allocation of carbon in the spikes.

4. Conclusions

Maize plants under the influence of nitrogen fertilization at sowing present differential responses in the growth and partition of assimilation between their structures, with the intensity depending on the dose used.

Plants under the effect of applying 75% of the recommended dose of N at sowing have a higher relative growth rate, with more pronounced effects than the other doses. The vigor expression of maize seeds increased up to 82.5% N at sowing.

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