

Ecology of Upland Rice Plants and Seeds Subjected to Growth Regulator

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Abstract

The objective of this work is to verify the influence of the plant regulator tri-nexapac-ethyl on plant growth and physiological quality of upland rice seeds. We used an upland rice genotype, and the growth regulator was the trinexapac-ethyl. The experiment was completely randomized in a 4×8 factorial design related to four concentrations of the plant regulator trinexapac-ethyl [0 (without growth regulator), 0.25, 0.50, and 0.75 L c.p. ha^{-1}] and 7 plant collection times [14, 28, 42, 56, 70, 84, 98 days after emergence (DAE)], with four replicates. To determine growth attributes, we evaluated total dry matter, dry matter production rate, relative growth rate, leaf area ratio, leaf matter, leaf area index, solar energy conversion efficiency and partition of assimilates. The physiological quality of the seeds was evaluated based on germination, first germination count, field emergence, emergence speed index and seedling dry matter. Plant growth was affected by the growth regulator. Total dry matter, dry matter production rate and solar energy conversion efficiency decreased, while leaf area index, leaf area ratio and leaf matter increased due to the effects of the growth regulator. The dry matter partition of plants changed in plants subjected to the growth regulator, with a delay in the targeting of assimilates to reproductive organs and a greater allocation to roots at the end of the cycle in plants subjected to the doses 0.50 and 0.75 L· ha^{-1} of growth regulator. Seed vigor was adversely affected by the growth regulator.

Keywords

Oryza sativa L., Trinexapac-Ethyl, Leaf Area, Dry Matter, Seed Vigor

1. Introduction

In Brazil, rice cultivation has a prominent position because of its economic and

social importance. It is a basic ingredient in the diet of most Brazilians. The production of this crop (irrigated and upland) in Brazil is around 10.6 million tons, reaching an average productivity of more than 5.2 t·ha⁻¹. The state of Rio Grande do Sul deserves a special mention. It achieves a production of approximately 7.3 million tons and an average productivity of approximately 6.8 tons per hectare [1].

The productivity of rice crops depends on several factors, including cultivar, amount of inputs and management techniques [2]. The achievement of high yields is related to the use of a set of technologies more appropriate to this crop. Correction of soil fertility is an essential prerequisite for achieving high yields.

The availability of mineral elements at appropriate levels in the soil solution favors the plant growth and the seed quality of different species [3]. However, the supply of fertilizer in excess, especially of nitrogen, predisposes plants to lodging, compromising productivity and seed quality and hindering mechanized harvesting [4]. In order to minimize this issue, resistant cultivars and growth regulators are used.

Growth regulators are chemical compounds exogenously applied to plants which exert similar effects as plant hormones [5] [6]. The ability of growth regulators to change plant physiology and morphology is an important tool in the search for high productive potentials of plants [7]. In order to evaluate the need for growth regulators, it is essential to consider soil and climatic conditions and the level of nitrogen in the soil [8].

Several studies were conducted on the growth regulator trinexapac-ethyl aiming to reduce lodging and evaluate the productivity of crops such as corn [9] wheat [7] [8] and rice [9] [10]. To evaluate plant growth, plant quality and the chemical composition of wheat seeds, [11] [12] reported reductions in height and in dry matter allocation. In contrast, there was high vigor and high levels of starch, amino acids and proteins in seeds produced by plants subjected to trinexapac-ethyl applied at doses recommended for these crops.

It is possible to verify that the use of such substances has become an important tool in crop management, mainly aiming to obtain high yields. In this sense, it is hypothesized that the application of trinexapac-ethyl changes plant growth and partitioning of assimilates, reducing the allocation of assimilates to vegetative parts and promoting allocation of assimilates to seeds.

Growth analysis is a quantitative, low-cost, accurate method to evaluate plant growth over time in relation to different media and management conditions [13]. It allows inferences on different physiological processes related to the characterization of plant performance and the interpretation and evaluation of primary production [13] [14] [15]. Physiological quality is the ability of seeds to perform vital functions such as germination, vigor and longevity [16]. Seed vigor is related to the level of seed reserves, hydrolysis, translocation and seedling allocation, as well as seed performance in field conditions, which may be measured by the seedling emergence test [17].

The objective of this study is to evaluate the effects of the plant regulator *trinexapac-ethyl* on plant growth and the physiological quality of upland rice seeds.

2. Materials and Methods

The study was conducted in a chapel greenhouse. The analyses were performed at the Seed Analysis Didactic Laboratory of the Graduate Program in Seed Sciences and Technology, Eliseu Maciel Agronomy College, Federal University of Pelotas, at an altitude of 13 m, 31°52'S and 52°21'W.

The experiment was completely randomized in a 4×7 factorial design related to four concentrations of the plant growth regulator trinexapac-ethyl [0 (without growth regulator), 0.25, 0.50, and 0.75 L of the commercial product (p.c) ha^{-1}] and 7 plant collection times [14, 28, 42, 56, 70, 84, 98 days after emergence (DAE)], with four replicates.

We used an upland rice genotype. Its sowing was carried out in polyethylene buckets with a capacity of 12 liters. The substrate was the A1-horizon of a solodic Haplic Eutrophic Planosol present within the Pelotas mapping unit [18]. The chemical and physical characteristics of the soil are pH (H_2O): 5.1, P: 121.2 $\text{mg}\cdot\text{dm}^{-3}$, K: 123 $\text{mg}\cdot\text{dm}^{-3}$, Ca: 2.7 $\text{cmol}_c\text{ dm}^{-3}$, Mg: 0.9 $\text{cmol}_c\text{ dm}^{-3}$, Al: 0.2 $\text{cmol}_c\text{ dm}^{-3}$, B: 0.3 $\text{mg}\cdot\text{dm}^{-3}$, Cu: 5.6 $\text{mg}\cdot\text{dm}^{-3}$, Zn: 2.2 $\text{mg}\cdot\text{dm}^{-3}$, Mn: 9 $\text{mg}\cdot\text{dm}^{-3}$, CEC: 8.1 $\text{cmol}_c\text{ dm}^{-3}$, base saturation: 52%, organic matter: 1.8%, and clay: 17%.

The fertilization was performed according to the recommendation of the Manual of Fertilization and Liming of the states of Rio Grande do Sul and Santa Catarina [19]. Ten seeds were sown per pot, and a thinning was performed. Four plants remained in each pot.

Phosphate and potassium fertilization were performed during the pre-sowing period. 0.015 $\text{kg}\cdot\text{m}^3$ of P_2O_5 was incorporated into the soil as triple super phosphate and 0.015 $\text{kg}\cdot\text{m}^3$ of K_2O was incorporated as potassium chloride. The nitrogen fertilization consisted of 0.010 $\text{kg}\cdot\text{m}^3$ of nitrogen as urea incorporated during pre-sowing, and 0.045 $\text{kg}\cdot\text{m}^3$ of nitrogen as urea was used as cover fertilization, divided into two applications (the first at the beginning of tillering and the second at the beginning of stretching). For the application of cover fertilization, the nitrogen source was urea (45% of N), which was evenly distributed throughout the surface area of the pots.

The plant regulator used for the experiment was *trinexapac-ethyl*, trade name Moddus®, “{4-(cyclopropyl-a-hydroxy-methylene)-3,5-dioxocyclohexanecarboxylic acid ethyl ester}”, the concentration of active ingredient in this commercial product is 250 g·L. The application of the plant growth regulator was carried out during the V9 stage [20], when there is a differentiation of the panicle primordium 33 days after sowing. The application was made using a CO_2 pressurized bar sprayer and fan-like tips (110-020), with a spray volume of 150 $\text{L}\cdot\text{ha}^{-1}$ and doses corresponding to each treatment. Pest and disease control was carried out whenever necessary and as recommended for the crop by spraying insecticides and fungicides.

In order to evaluate plant growth, successive collections were carried out at regular intervals of fourteen days after emergence until the physiological maturity of seeds. The plant organs were separated into shoot (leaf blade, stem, stalk and reproductive structures, whenever present) and roots. The plant parts were packed in brown paper bags and dried in a forced-air circulation oven at $70^{\circ}\text{C} \pm 2^{\circ}\text{C}$ until constant mass.

Primary data for leaf area (A_t), dry leaf matter (W_t), stalk (W_s), roots (W_r) and panicle (W_{pan}) were adjusted by orthogonal polynomials [29]. The data for accumulated primary dry matter (W_t) were adjusted by the simple logistic equation: $W_t = W_m / (1 + Ae^{-Bt})$, where W_m is the asymptotic maximum growth estimate, "A" and "B" are fitting constants, "e" is the natural basis of neperian logarithm, and "t" is time in days after emergence [21].

The leaf area index (L) was calculated by the formula: $L = A_t/S_t$, where S_t is the surface area of the pot occupied by the plant. The values for total dry matter production rate (C_t) were obtained by time derivatives of adjusted equations of total dry matter (W_t) [13]. For the determination of instantaneous values of relative growth rate (R_w), the equation used was: $R_w = 1/W_t \cdot d_w/d_t$, and the instantaneous values of leaf area ratio (F_a) and leaf matter (F_w) were estimated by the equations: $E_a = 1/A_t \cdot d_w/d_t$, $F_a = A_t/W_t$, and $F_w = W_t/W_t$, according to [13].

The solar energy conversion efficiency (ξ) was determined by the equation $\xi(\%) = (100 \times C_t \times \delta)/R_s$, where R_s is the mean value of the incident solar radiation ($\text{cal m}^{-2} \cdot \text{day}^{-1}$) fourteen days before the corresponding C_t , and δ calorific value of $3702.11 \text{ cal g}^{-1}$, according to [22]. The dry matter partition among plant structures (roots, stalk, leaves and panicle) was determined by measuring the mass allocated to each plant structure with a subsequent transformation of primary dry matter allocation data of each organ into percentage.

Soon after the occurrence of physiological maturity, the seeds were harvested with a moisture content of 25%. After harvest, the seeds were dried in a forced-air circulation oven at 33°C , and stored in a cold room for later evaluation of physiological quality. To evaluate the effects of the plant regulator on physiological quality, the seeds were subjected to the evaluations below.

1) Germination: five replicates were used, with four subsamples of 50 seeds for each treatment sown in a roll of *Germitest* germinating paper, moistened with a volume of water 2.5 times the mass of the dry substrate, and kept in a BOD germination chamber at 25°C and a photoperiod of 12 hours. The counts were performed at fourteen days after sowing according to the Rules for Seed Analysis [23]. The results were expressed as percentage of normal plants.

2) First germination count: performed together with the germination test. The count was performed five days after sowing according to the Rules for Seed Analysis [23]. The results were expressed as percentage of normal plants.

3) Germination speed index: obtained from daily counts of germinated seeds (minimal root protrusion of 3 - 4 mm until obtaining a constant number of germinated seeds. The calculation of the germination speed index was per-

formed using the Maguire equation [24].

4) Emergence of seedlings in the field: performed by sowing five replicates of 100 seeds per treatment in Planosol beds. The evaluation was performed 21 days after sowing. The percentage of seedling emergence was then determined [25].

5) Emergence speed index: determined in conjunction with the seedling emergence test in the field by daily counting the number of seedlings emerged since the onset of emergence until the process stabilized. The calculation of the emergence speed index was performed using the Maguire equation [24].

6) Seedling dry matter: performed in laboratory at fourteen days together with the germination test, and at 21 days in the field together with the seedling emergence test. To determine dry matter, five replicates were collected containing 10 seedlings per treatment. The seedlings were separated into shoots and roots, placed in brown paper envelopes, and taken to a forced-air circulation oven at 70°C until constant mass, which was determined using a precision scale. The results were expressed as mg seedling⁻¹.

Primary growth data were submitted to analysis of variance at 5% probability and analyzed using simple logistic equation. Values of partition of assimilates were converted into percentage [15]. An analysis of variance was conducted on data referring to the physiological quality of seeds. When significant at a 5% probability, they were adjusted by orthogonal polynomials.

3. Results and Discussion

The dry matter values of seedling shoots and roots obtained at 14 days in conjunction with the germination test and at 21 days in conjunction with the field emergence test did not show significant differences among treatments.

In all treatments, the total dry matter (W_t) of rice plants adjusted to a logistic tendency, with a high coefficient of determination ($R = 0.99$), as can be observed in **Figure 1**. During the initial period of the crop cycle of plants in all treatments, a phase of slow growth occurred until 42 DAE, following a phase of intense growth. From 56 DAE, it is possible to observe a reduction in the growth of plants subjected to the growth regulator. At 98 DAE, there was an intense reduction in dry matter allocation in plants subjected to growth regulator, with a reduction of approximately 25%, 40% and 41% in plants subjected to the doses 0.25, 0.50 and 0.75 L·ha⁻¹, respectively, compared to plants without growth regulator.

The reduced production of dry matter at the beginning of plant cycle is common and linked to the small leaf area during this period. However, throughout the development cycle of plants, a normal tendency of increase in total dry matter occurs mainly due to an increase in the leaf area available to the photosynthetic process [12], which, to some extent, favors the interception of solar radiation by plants.

The reduction in W_t values at 98 DAE in plants subjected to growth regulator

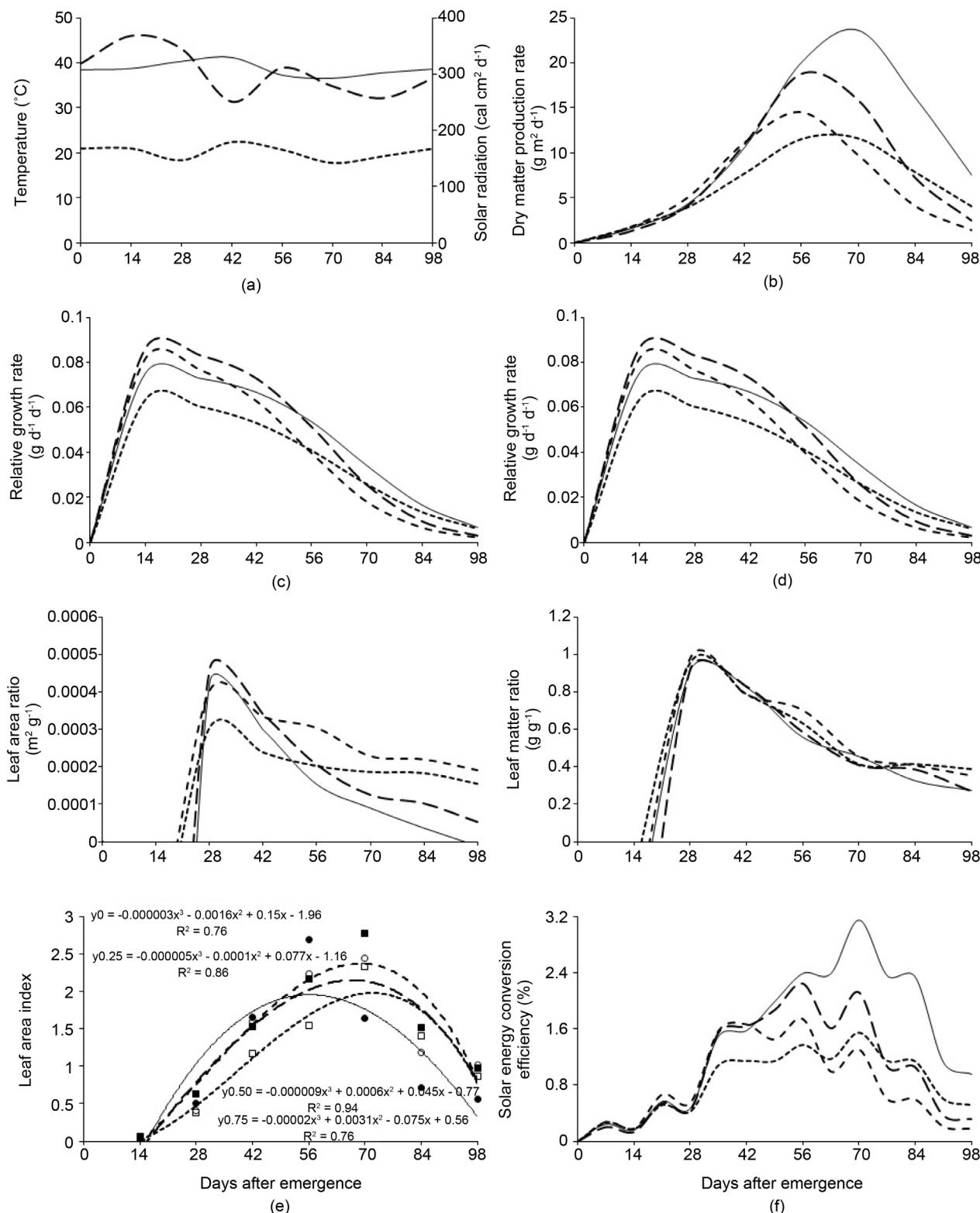


Figure 1. Maximum (---) and minimum (---) temperature and solar radiation (----) (a), total dry matter (b), dry matter production rate (c), relative growth rate (d), leaf area ratio (e), leaf matter ratio (f), leaf area index (g), and solar energy conversion efficiency (h) of upland rice plants subjected to different doses of the growth regulator trinexapac-ethyl. 0 (----), 0.25 (---), 0.50 (---) and 0.75 (.....) L·ha⁻¹ of commercial product (c.p.).

are related to its effects on the reduction of active gibberellin synthesis, which is responsible for cell division and stretching [26]. The inhibition of this synthesis results in a momentary stop in plant growth, resulting in a smaller stature and in dry matter allocation, a fact verified until the end of the plant cycle.

The dry matter production rate (C_t) reached low values up to 42 DAE, a fact related to a low dry matter production during this period, as observed in **Figure 1(b)**. The growth regulator caused a temporal-quantitative change in the value of C_t , indicating a change in dry matter production per unit area and per time (**Figure 1(c)**).

The maximum values of C_t were obtained at 56 DAE in plants subjected to growth regulator at the doses 0.25 and 0.50 L·ha⁻¹, and at 70 DAE in plants without growth regulator and also plants subjected to the dose 0.75 L·ha⁻¹. The highest production rate values were observed for plants without growth regulator. A reduction by 20%, 38% and 50% in relation to the control was observed for plants subjected to the application of 0.25, 0.50 and 0.75 L·ha⁻¹ of growth regulator, respectively.

The increase in C_t values is related to leaf number increase, the amount of photoassimilates synthesized and their distribution among plant organs [15]. Its subsequent reduction tendency occurred due to an increase in non-photosynthetic tissues and also due to auto-shading [27]. The reduction in C_t values in treatments subjected to growth regulator, in relation to treatments without growth regulator, is probably due to a temporary inhibition of plant growth resulting from the inhibition of the synthesis of the gibberellin that the product provides.

The values for relative growth rate (R_w) were maximal at 14 DAE, with a subsequent and systematic decrease up to 98 DAE (**Figure 1(d)**). After 42 DAE, the R_w values of plants that received the growth regulator were lower than the values of control plants (without growth regulator), indicating a low increase in dry matter in relation to the preexisting dry matter in plants subjected to the growth regulator. At 56 DAE, there was a reduction by 4%, 24% and 26% in the R_w values in plants subjected to the doses 0.25, 0.75 and 0.50 L·ha⁻¹ of growth regulator, respectively, in relation to plants without growth regulator.

During the early stages of development, the highest values of R_w are explained by the fact that plants have young leaves, with a high photosynthetic capacity and a high growth rate [14] [15]. In contrast, such reduction in values is related to an increase in the proportion of non-photosynthetic tissues and also to shading [27] [28].

The leaf area ratio (F_a) reached the highest values at the beginning of the plant cycle, at 28 DAE (**Figure 1(e)**). Plants subjected to a dose of 0.75 L·ha⁻¹ experienced a marked decrease in values at 28 DAE. From 42 DAEs, plants subjected to the doses 0.50 and 0.75 L·ha⁻¹ evidenced a less marked decrease in F_a values up to 98 DAE, whereas plants subjected to a dose of 0.25 L·ha⁻¹ without growth regulator showed a marked decrease in values. Such more and less pronounced decrease tendencies provided high F_a values for plants subjected to the doses 0.50

and 0.75 L·ha⁻¹ of growth regulator from 56 DAE until the end of the plant cycle.

The superiority of F_a values from 42 DAE in plants subjected to growth regulator in relation to plants without it probably occurs due to a marked decrease in total dry matter allocation (**Figure 1(b)**) and an increase in leaf area as observed for the plants of these treatments, indicating a larger leaf area in relation to the total plant dry matter.

According to [29], the application of trinexapac-ethyl indirectly affects the leaf area of wheat plants. It is linked to effects on the number of leaves, leaf length and leaf width. It is important to note that the intensity of such effects varies among different cultivars.

It is important to note that in situations of environmental normality, the occurrence of higher F_a values at the beginning of the plant cycle occurs because the greater proportion of assimilates formed in during photosynthesis is directed to the formation of leaves, which are the preferential metabolic drain during this period [14]. However, a decrease in F_a values is related to an increase in non-photosynthetic tissues and to changes in the preferential metabolic drain occurring during the cycle [15].

Leaf matter ratio (F_w) reached maximum values at the beginning of the plant cycle, obtaining the highest values at 28 DAE for plants of all treatments (**Figure 1(f)**). At 56 DAE, plants subjected to the doses 0.50 and 0.75 L·ha⁻¹ of growth regulator obtained a second peak of F_w , maintaining high values at the end of the cycle. At 98 DAE, for plants subjected to the doses 0.50 and 0.75 L·ha⁻¹, we observed an increase in F_w values of approximately 29% and 41%, respectively, in comparison to plants without growth regulator.

High F_w values at the beginning of development occur because leaves are the preferred metabolic drain during this period. However, during plant ontogeny, there are changes in the preferential metabolic drain, *i.e.*, stalks; later, reproductive organs become the preferential metabolic drains [15]. Similar tendencies were observed by [30] for rice by studying the growth of different rice cultivars and by [18] for wheat by studying the effects of trinexapac-ethyl on plant growth.

Leaf area index (L) values had a temporal-quantitative change in maximum values (**Figure 1(g)**). For plants without growth regulator, the highest values were obtained at 56 DAE. However, for the plants of other treatments, subjected to the growth regulator, the maximum values were achieved at 70 DAE. We observed the maximum values of 28%, 32% and 24% for plants without growth regulator, and 0.25 and 0.50 L·ha⁻¹ for plants subjected to growth regulator, respectively, in relation to plants without growth regulator.

The values of L increased in plants subjected to the growth regulator, indicating an increase in the soil area occupied by leaves. This was also observed by [31] for corn by evaluating the effects of doses and times of application of trinexapac-ethyl on corn hybrids, and by [11] for wheat by evaluating the effects of trinexapac-ethyl doses on plant growth.

The solar energy conversion efficiency (ξ) was changed by the growth regulator, presenting a temporal-quantitative change among plants of different treatments (Figure 1(h)). The maximum values for plants without growth regulator and plants subjected to a dose of 0.75 L-ha^{-1} of growth regulator were found at 70 DAE, whereas plants subjected to the doses 0.25 and 0.50 L-ha^{-1} of growth regulator reached the maximum values at 56 DAE. Plants without growth regulator reached the highest values for ξ . When subjected to the doses 0.25 , 0.50 and 0.75 , there were decreases of approximately 29% , 45% and 51% , respectively, in relation to control plants.

The increase tendency found for ξ corroborates what was observed for C_t (Figure 1(c)), and relates to solar radiation (Figure 1(a)). Solar energy conversion efficiency is influenced by light interception by the canopy of plants, by leaf area and by photosynthetic active radiation [32]. The growth regulator trinex-apac-ethyl acts on the intermeral meristems, causing a decrease in the levels of active gibberellin, which results in a reduced plant height [25] [33].

The dry matter partition in control plants showed that, up to 42 days after emergence (DAE), the highest assimilate allocation occurred on leaves, followed by roots (Figures 2(a)-(d)). From 42 DAE, we verified a decrease in allocation to these structures and an increase in the allocation of dry matter to the stalk. At 56 DAE, a new change in the preferential metabolic drainage occurred, with prioritization of allocation to reproductive organs (panicles and seeds).

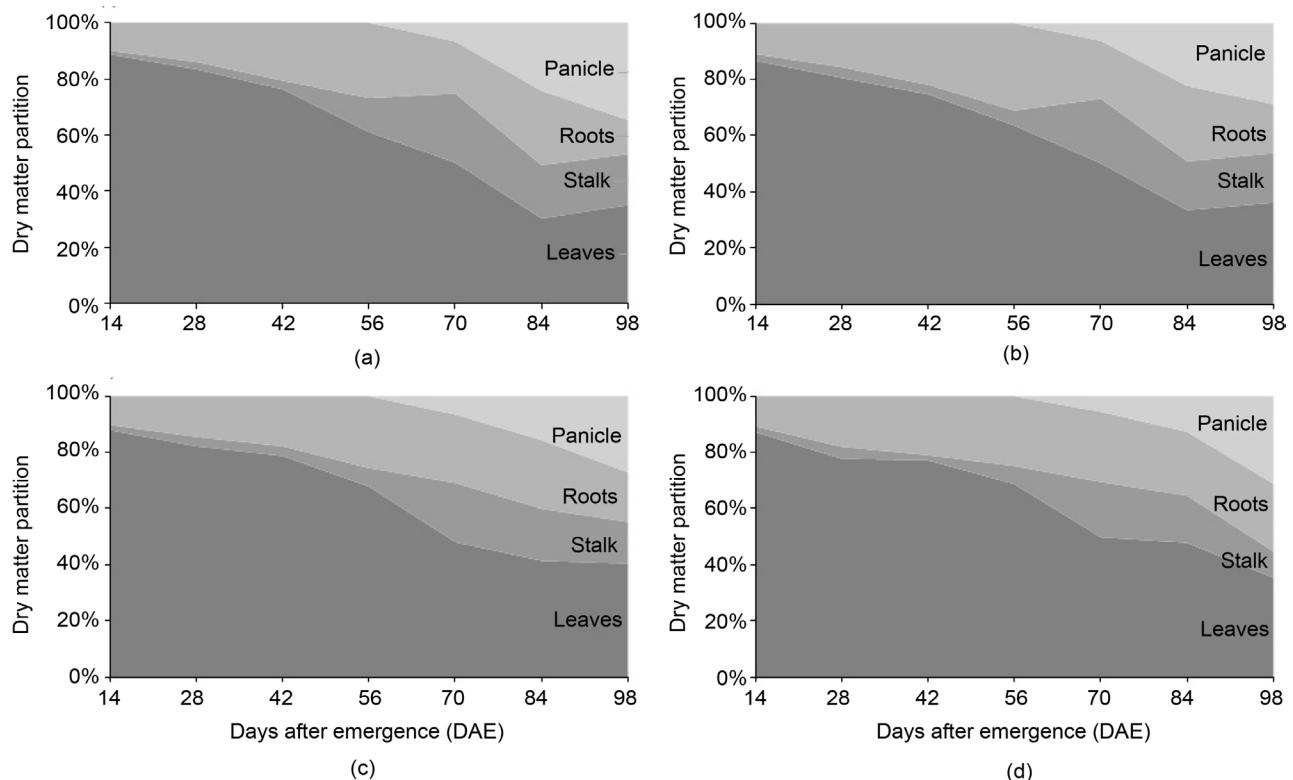


Figure 2. Dry matter partition in different structures of upland rice plants subjected to the application of trinexapac-ethyl. $0 =$ no application (a), 0.25 (b), 0.50 (c) and 0.75 (d) L-ha^{-1} of commercial product (p.c.).

Plants subjected to the doses 0.50 and 0.75 L·ha⁻¹ of growth regulator (**Figure 2(c)** and **Figure 2(d)**) obtained a low intensity of initial allocation to reproductive organs in relation to plants of other treatments, which is, in part, a result of the delayed panicle emission observed for these plants. The delay in emission of reproductive organs was also observed for wheat plants subjected to the application of trinexapac-ethyl. This may be the result of a delay in growth during the stalk elongation stage [11]. In plants subjected to the highest dose of growth regulator (0.75 L·ha⁻¹), there was a high dry matter allocation to roots at the end of the cycle in relation to the stalk, which may be a result different from plants of other treatments.

Germination values adjusted to a linear tendency, and reduced up to the highest regulator dose (0.75 L·ha⁻¹), corresponding to a decrease of 2.4% in relation to plants without growth regulator (**Figure 3(b)**). The first germination count, field emergence and emergence speed index reached a linear decrease as the dose of growth regulator applied to plants increased (**Figure 3(c)**, **Figure 3(d)**). A reduction of 4.4%, 20% and 12.5% in the highest dose (0.75 L·ha⁻¹) could be observed in relation to the control without growth regulator.

The decrease in vigor expression evidenced by the tests, especially at the highest dose (0.75 L·ha⁻¹), may be related to a severe reduction in plant structure, as evidenced by the marked decrease in dry matter allocation (**Figure 1(b)**) and in

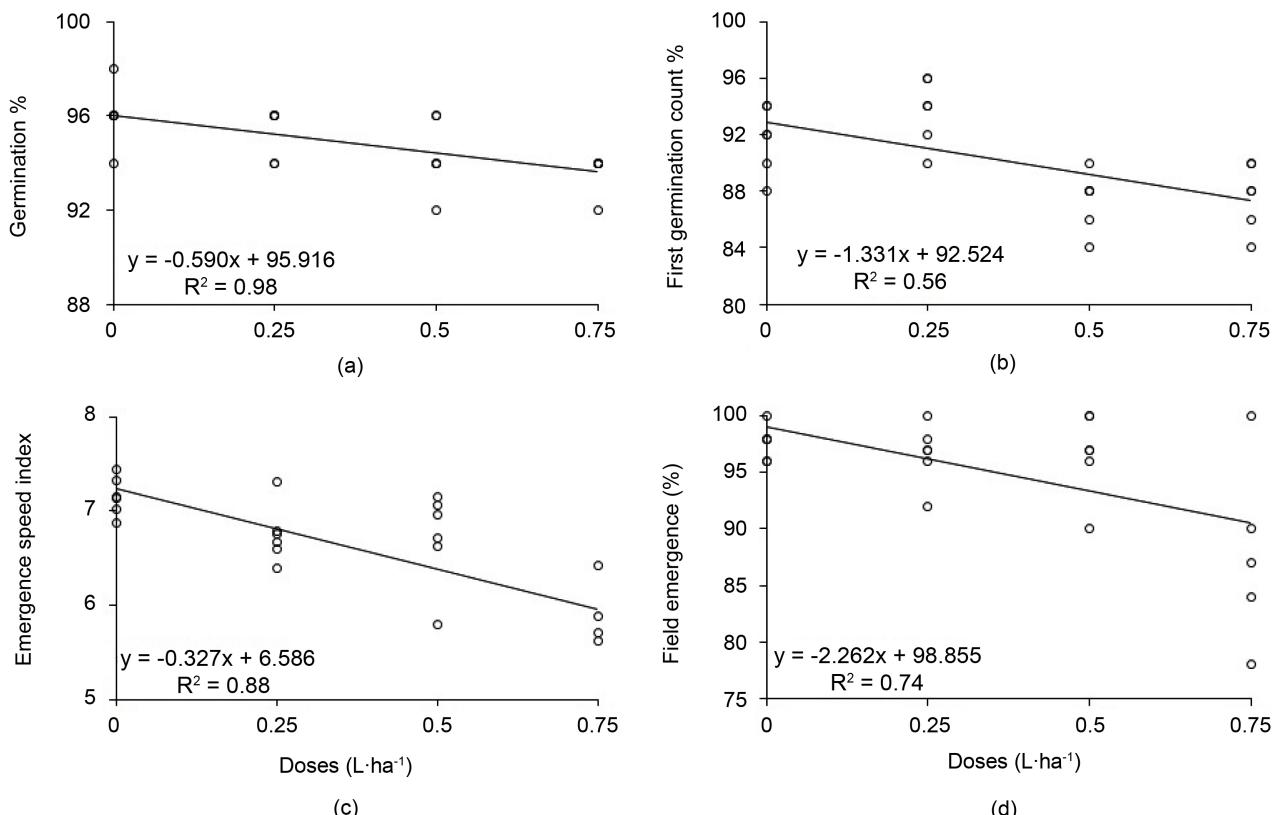


Figure 3. Germination (a), first germination count (b), emergence speed index (c) and field emergence (d) of upland rice seedlings subjected to different doses of the growth regulator trinexapac-ethyl.

leaf area (**Figure 1(e)**, **Figure 1(g)**), which could be a possible consequence of a lower intake of assimilates.

During the production of seeds, several factors may negatively affect their quality. In this case, direct factors act on seeds, while indirect factors affect the physiological and biochemical processes which occur in the matrix plant, changing the partition of assimilates between different organs, biosynthesis of compounds and, consequently, seed vigor [17] [34].

Based on this, it is possible to state that the growth regulator causes changes in the growth of upland rice plants, with a reduction in dry matter allocation, and, in contrast, an increase in the values of leaf area index, leaf area ratio and leaf matter. Solar energy conversion efficiency is decreased by the growth regulator. Another interesting fact is the delay in the partitioning of assimilates for reproductive organs and the high proportion of dry matter allocated to roots at the end of the crop cycle. Seed vigor decreased by the application of the growth regulator at high doses, which is an indication of a low efficiency in the reorganization of the cell membrane system, hydrolysis and allocation of reserves to seedlings.

4. Conclusions

Plant growth was affected by the growth regulator.

Total dry matter, dry matter production rate and solar energy conversion efficiency decreased, while leaf area index, leaf area ratio and leaf matter increased due to the effects of the growth regulator.

The dry matter partition of plants changed in plants subjected to the growth regulator, with a delay in the targeting of assimilates to reproductive organs and a greater allocation to roots at the end of the cycle in plants subjected to the doses 0.50 and 0.75 L·ha⁻¹ of growth regulator.

Seed vigor was adversely affected by high doses of growth regulator.

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