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Water Depths and Nitrogen Rates on the Physiological Quality of Cotton Seeds Produced in the Arenito Caiuá Region

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Abstract

This work aimed to analyze the effect of water depths and nitrogen rates on the productivity and physiological quality of cotton seeds produced in the Arenito Caiuá region. In addition, it is expected to verify if it is possible to obtain quality seeds when they are produced in a low-altitude region. For this purpose, cotton seeds were sown, with a population of 122 thousand plants per hectare. The treatments consisted on the application of three nitrogen rates (30, 60 and 90 kg/ha) and four irrigation depths (0%, 50%, 100% and 150% of crop evapotranspiration), applied

to the soil. When ripe, the seeds were harvested and manually delinted, and productivity was measured. Analyzes were also carried out to investigate the physiological quality of newly harvested seeds and six-month stored seeds. It was observed that the water depths and nitrogen rates applied to plants affect productivity, but not the physiological quality of the seeds. Furthermore, it is possible to obtain high quality seeds in a low-altitude region.

Keywords: *Gossypium Hirsutum* L, Irrigation, Fertilization, Seed Storage

Introduction

Cotton (*Gossypium hirsutum* L.) is among the most important fiber-producing crops worldwide. World production is estimated at 27 million tons per year (Vaddula and Singh, 2023) ^[19]. In Brazil, in the last harvest, 2022/2023, about 1,663 million hectares were planted, producing an average of 4,493.6 million tons, which represents an increase of 20.8% in production compared to last year. (Conab, 2023) ^[7].

Cotton has been produced from small farms, associated with family farming, to large areas, by producers with high technological level (Aguiar *et al.*, 2014) ^[1]. In Brazil, three macro-regions deserve to be highlighted for cotton production: North-Northeast, comprising the states of Tocantins, Maranhão, Piauí, Ceará, Rio Grande do Norte, Paraíba, Pernambuco, Alagoas and Bahia, Midwest, with significant production in the states of Mato Grosso, Mato Grosso do Sul and Goiás, and South-Southeast, especially the states of São Paulo, Paraná and Minas Gerais (Aguiar *et al.*, 2014) ^[1].

Due to the increasing demand for cotton fiber worldwide, the prospects of the crop for the coming years are promising (Severino *et al.*, 2019) ^[15], which also increases the demand for inputs that guarantee the production and productivity of the crop. Among the inputs required for a good production, the seed deserves to be highlighted.

The use of quality seeds is one of the main factors responsible for the success of cotton crops (Mayrinck *et al.*, 2020) ^[13]. High-quality seeds show good performance in the field, rapid emergence and establishment of plants, even under less favorable environmental conditions (Mattioni *et al.*, 2012) ^[12]. On the other hand, low-quality seeds generally result in slow germination, uneven stands, low weather tolerance, and often planting failures, culminating in agricultural productivity loss (Mattioni *et al.*, 2012) ^[12].

The obtaining of quality seeds depends, among other factors, on proper management of irrigation and plant nutrition. The water availability, with a correct water supply, and the nutritional balance of the plant are required for the good formation and development of seeds (Bewley *et al.*, 2013) ^[4], which generally reflects on their quality.

However, water stress, whether due to water deficit or excess, and/or nutrient imbalance in the soil and the plant, can reduce germination and vigor or, ultimately, derail seed formation (Bewley *et al.*, 2013).^[4] The effect of water and nutritional stress on seed formation, development, and quality depends on the genotype, level and duration of stress, as well as the soil type, and the climate of the region.

The Umuarama city is among the regions of Paraná that presents historical prominence for the production of herbaceous cotton (Moura *et al.*, 2020)^[14]. It is located at an approximate altitude of 401 m, with a subtropical climate and a predominance of sandy to medium texture soils, known as Arenito Caiuá region (Cunha *et al.*, 1999)^[8]. Although the climate and soil characteristics of the region are not the most conducive to seed production, the correct management of water and nutrients can be decisive for the success in the production of quality seeds.

Given the current trend of expanding seed production into previously uncharted regions and the lack of research concerning seed production in low-lying areas, it becomes essential to explore the quality characteristics of seeds cultivated in these regions. This research is justified both under well-managed conditions with adequate water and nutrient supply and under conditions of water and nutrient stress.

Therefore, the aim of this work was to analyze the effect of water depths and nitrogen rates on the productivity and physiological quality of cotton seeds produced in the Arenito Caiuá region. In addition, it is expected to verify if it is possible to obtain quality seeds when they are produced in a low-altitude region.

Material and Methods

The experiment was carried out in the Seed Laboratory, and in the experimental area of the State University of Maringá, Umuarama Regional Campus, located at 23°47' South latitude and 53°14' West longitude. According to the Köppen classification, the region's climate is of the humid mesothermal subtropical type (Cfa), with an average annual temperature of 28°C, and precipitation of 1,658 mm (Iapar, 2020)^[9].

In this research, two experiments were carried out: field experiment, and laboratory experiment, as described below.

Field experiment

Cotton seeds of the cultivar FiberMax® were sown under a population of approximately 122 thousand plants per hectare. The treatments consisted of the application of three nitrogen rates and four irrigation depths applied to the soil during the development of cotton seed cultivation.

The rates of 30, 60 and 90 kg ha⁻¹ were applied at 40 days after the emergence of seedling (DAE), using urea as a nitrogen source. In addition, irrigation depths of 0%, 50%, 100% and 150% of crop evapotranspiration (ETc) were applied from the 10 DAE, remaining up to 115 DAE.

The experimental design was carried out in split plots, with three replications. The plots were 4.5 meters long and 4.3 meters wide, containing 5 cotton lines, with spacing of 0.90 m between rows. The three central lines of each plot were considered as effective experimental areas for the evaluations.

The localized irrigation method was adopted, with a drip

system, through the use of drip tapes with emitters with nominal flow of 2 L h⁻¹ and spaced at 0.15 m.

Irrigation management was performed via climate, with reference evapotranspiration calculated by the method proposed in FAO Bulletin 56 (Allen *et al.*, 1998)^[3], with methodology adapted from the Penman-Monteith method, as shown in the equation below:

$$ET_o = \frac{0,408 \cdot S \cdot (R_n - G) + \gamma \cdot \frac{900}{T_a + 273} \cdot u_2 \cdot (e_s - e_a)}{S + \gamma \cdot (1 + 0,34 \cdot u_2)}$$

Where S is the tangent of the air vapor saturation pressure curve, R_n is the radiation balance, in MJ m⁻² d⁻¹, G is the heat flux in the ground, in MJ m⁻² d⁻¹, T_a is the average air temperature, in °C, u₂ is the wind speed at 2 m, in m s⁻¹, e_s - e_a is the vapor pressure deficit, in kPa, and γ is the psychrometric constant.

Meteorological data were obtained using an automatic meteorological station installed next to the experimental area, and crop evapotranspiration (ETc) was calculated (Allen *et al.*, 1998)^[3]. The irrigation depth was calculated using an application efficiency of 0.95.

All cultural treatments of weed, pest, and disease management were carried out throughout the crop cycle. After physiological maturity, the seeds were harvested and cleared manually. The seeds of each plot were weighed, and the values for kilograms per hectare were extrapolated, considering a population of 122,000 plants per hectare.

Analysis of variance was performed and the effect of nitrogen rates and water depths and their interaction on seed yield was verified. Treatment means were compared using the Tukey test at 5% probability, and the R Software.

Laboratory Experiment

Freshly harvested seeds were submitted to germination and vigor tests. After that, they were stored for 6 months, under laboratory conditions (without humidity and temperature control), and once again the same tests were performed, using the methodologies described below:

Moisture Content - Performed immediately before the execution of the tests, as determined by the greenhouse method, at 105 ± 3 °C, for 24 hours, using four replicates of 25 seeds each (Brasil, 2009)^[5].

Germination test - Four replicates of 50 seeds per treatment were used. The seeds were sown equidistantly on Germitest paper moistened with a volume of water equivalent to 2.5 times the weight of the dry substrate and kept in a germinator at 25 °C. Evaluations were carried out with a record of the percentage of normal seedlings on the 4th and 12th days after sowing (Brasil, 2009)^[5].

Accelerated Aging - Four replicates of 50 seeds each were used. The seeds were distributed in a single layer on a metal mesh tray attached to the Gerbox that contained, at the bottom, 40 mL of distilled water. The boxes were capped to obtain 100% of relative humidity (RH) inside and kept in a BOD incubator at 41°C for 48 hours (Krzyzanowski, *et al.*, 2020)^[10].

The effect of the interaction between nitrogen rates and water depth was not analyzed for the performance of the physiological quality tests of the seeds. The field treatments were numbered from 1 to 12, as described in Table 1.

Table 1: Description of the treatments, considering the water depths applied according to the crop evapotranspiration (ETc) and nitrogen (N) rates applied in kilograms per hectare (kg/ha) in the cotton crop (*Gossypium hirsutum* L.)

| Treatments | Codes | Description |
|------------|----------------------|--------------------------------------------------------|
| T1 | 0% ETc - 30Kg/ha N | No water depth (0% ETc) and 30 kg/ha of N were applied |
| T2 | 0% ETc - 60Kg/ha N | No water depth (0% ETc) and 60 kg/ha of N were applied |
| T3 | 0% ETc - 90Kg/ha N | No water depth (0% ETc) and 90 kg/ha of N were applied |
| T4 | 50% ETc - 30Kg/ha N | Water depth of 50% ETc and 30 kg/ha of N |
| T5 | 50% ETc - 60Kg/ha N | Water depth of 50% ETc and 60 kg/ha of N |
| T6 | 50% ETc - 90Kg/ha N | Water depth of 50% ETc and 90 kg/ha of N |
| T7 | 100% ETc - 30Kg/ha N | Water depth of 100% ETc and 30 kg/ha of N |
| T8 | 100% ETc - 60Kg/ha N | Water depth of 100% ETc and 60 kg/ha of N |
| T9 | 100% ETc - 90Kg/ha N | Water depth of 100% ETc and 90 kg/ha of N |
| T10 | 150% ETc - 30Kg/ha N | Water depth of 150% ETc and 30 kg/ha of N |
| T11 | 150% ETc - 60Kg/ha N | Water depth of 150% ETc and 60 kg/ha of N |
| T12 | 150% ETc - 90Kg/ha N | Water depth of 150% ETc and 90 kg/ha of N |

Statistical analyses: The physiological quality data of the seeds were analyzed in a 12 x 2 factorial scheme (12 treatments and 2 storage periods – harvested and 6-month stored seeds), in a completely randomized design. Analysis of variances was performed, and the means were compared using the Tukey test at a 5% probability, and the R Software.

Results and Discussion

The results obtained from the analysis of variance show that there was a significant interaction between the treatments of water depth and nitrogen fertilization for the yield of cotton seeds. And, the means of the treatments are shown in Table 2.

Table 2: Yield of cotton seeds (*Gossypium hirsutum* L.), when different water depths were applied, considering the percentage of crop evapotranspiration (%ETc), and nitrogen rates in kilograms per hectare (kg/ha)

| Water Depths (% ETc) | Nitrogen rates (Kg/ha) | | |
|----------------------|------------------------|---------|----------|
| | 30 | 60 | 90 |
| 0 | 2890 Ac | 3071 Ab | 3277 Ab |
| 50 | 3451 Ab | 3928 Aa | 3857 Aa |
| 100 | 3590 Ab | 3490 Ab | 3398 Aab |
| 150 | 4309 Aa | 3994 Aa | 3379 Bab |
| CV (%) | 16,7 | 11,6 | 7,6 |

Lowercase letters compare water depths within the same nitrogen level. Capital letters compare nitrogen levels for the same water depth. Means followed by the same letter do not differ from each other according to the Tukey test at a 5% probability. CV = coefficient of variation

There was no difference between the nitrogen rates for the 0, 50 and 100% ETc of water depths. However, when the highest nitrogen rate, 90 Kg/ha, and the highest water depth (150% ETc) were applied, there was a reduction of approximately 615 kg/ha in seed yield. Excess nitrogen can lead to excess uptake of NO₃ by plants, leading to decreased concentration of lignin and phenols, which are important for plant defense (Torres-Olivar *et al.* 2014) [17]. When in contact with water in the soil, there may be the formation of ammonium (NH₄), and the excess of ammonium can lead to rapid elongation of the stem and even block calcium absorption, which can be detrimental to plant development (Alarcón, 2002) [2], and which justifies the reduction of productivity in the environment with higher nitrogen and water supply (Table 2).

The highest water depth applied to the soil (150% ETc), when associated with the rates of 30 and 60 kg/ha, and the

highest nitrogen rate (90 kg/ha), in association with the water depth of 50% of the ETc, were the treatments that provided the highest seed productivity. On the other hand, the absence of irrigation (0% ETc) led to lower productivity, regardless of the nitrogen rate applied. These results emphasize the critical role of water supply in both seed production and cotton crop development, highlighting the essential nature of water in this agricultural process. It is estimated that it takes 10 liters of water to produce one kilogram of plume (Vaddula and Singh, 2023) [19].

The experiment was carried out in Umuarama – Paraná – Brazil. In this region, known as Caiuá Sandstone, the soils have a texture that varies from sandy to medium, with high sand content, low clay percentage, with sand content reaching 85% to 90% and have critical levels of phosphorus, potassium, calcium, magnesium and, not rarely, low levels of organic matter (Coêlho *et al.*, 2021) [6]. Since it is a hot region, there is rapid evapotranspiration of water, which justifies the application of a higher water depth (150% of the ETc) to present high productivity, while the absence of irrigation has presented lower productivity.

Through the tests performed to infer the physiological quality of the seeds, it was evidenced that the seeds presented a moisture content of approximately 9% for both the newly harvested seeds and those stored for 6 months.

For the freshly harvested seeds, there was no difference between the treatments of nitrogen fertilization and application of water depths. However, there was a decrease in germination for most treatments as the months stored increased, with the exception of treatments T1 (0% ETc 30 Kg), T3 (0% ETc 90 Kg), T9 (100% ETc 90 Kg N), and T10 (150 ETc 30 Kg) (Fig 1).

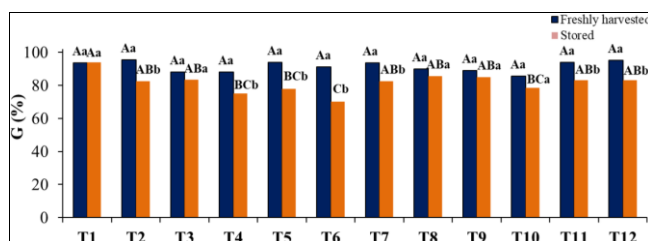


Fig 1: Germination of freshly harvested cotton seeds (in blue) and after 6-month storage (in orange), when applied to the different treatments of irrigation depth and nitrogen fertilization. T1 – 0 ETc 30 Kg N, T2 - 0 ETc 60 Kg N, T3 0 ETc 90 Kg N, T4 - 50 ETc 30 Kg N, T5 - 50 ETc 60 Kg N, T6 - 50 ETc 90 Kg N, T7 - 100 ETc 30 Kg N, T8 - 100 ETc 60 Kg N, T9 - 100 ETc 90 Kg N, T10 - 150 ETc 30 Kg N, T11 - 150 ETc 60 Kg N, T12 - 150 ETc 90 Kg N. Equal lowercase letters do not differ from each other, when

comparing between storage periods, and equal capital letters do not differ from each other, when comparing treatments within each storage period, according to the Tukey test at a 5% probability

It can be stated that the initial germination was high (above 90%) for all treatments, and even after the six-month storage, they still presented germination above 80%. Besides that, the seeds presented a high percentage of normal seedlings after aging, an average of 88% when freshly harvested, with no difference between treatments. However, there was a reduction in the percentage of normal seedlings after the accelerated aging test, when they were stored for 6 months (Fig 2).

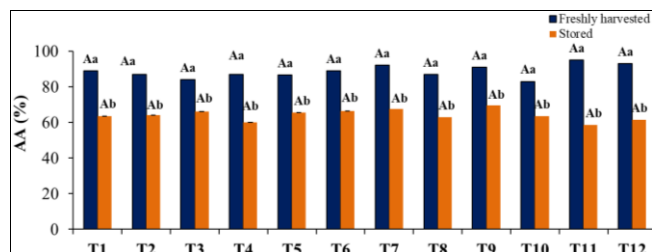


Fig 2: Accelerated aging (AA) in freshly harvested cotton seeds (in blue) and after six-month storage (in orange), when the different treatments of irrigation depth and nitrogen fertilization were applied. T1 – 0 ETc 30 Kg N, T2 - 0 ETc 60 Kg N, T3 0 ETc 90 Kg N, T4 - 50 ETc 30 Kg N, T5 - 50 ETc 60 Kg N, T6 - 50 ETc 90 Kg N, T7 - 100 ETc 30 Kg N, T8 - 100 ETc 60 Kg N, T9 - 100 ETc 90 Kg N, T10 - 150 ETc 30 Kg N, T11 - 150 ETc 60 Kg N, T12 - 150 ETc 90 Kg N. Equal lowercase letters do not differ from each other, when comparing storage periods, and equal capital letters do not differ from each other, when comparing treatments within each storage period, according to the Tukey test at a 5% probability

Accelerated aging test is one of the most widely used vigor tests for cotton seeds. In this test, seeds are subjected to high temperature and relative humidity, and subsequently, the percentage of germination after this stress is evaluated (Krzyzanowski *et al.*, 2020) [10]. The seeds with higher vigor tend to have a higher percentage of normal seedlings than the ones with least vigor, as evidenced in this experiment by the newly harvested seeds in relation to the stored ones (Fig 2).

Regarding the length of the seedlings, it can be stated that all treatments provided seedling length above 15 centimeters when freshly harvested. And even with a decrease in length in some treatments after the six-month storage, the seedlings still had a minimum length of 10 centimeters, which can be considered as vigorous (Fig 3).

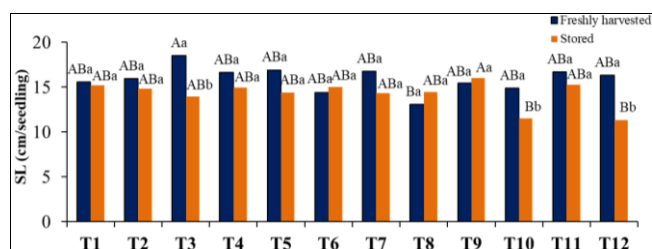


Fig 3: Seedling length (SL) of freshly harvested cotton (in blue) and after six-month storage (in orange), when applied to different treatments of water depths and nitrogen fertilization. T1 – 0 ETc 30 Kg N, T2 - 0 ETc 60 Kg N, T3 0 ETc 90 Kg N, T4 - 50 ETc 30 Kg N, T5 - 50 ETc 60 Kg N, T6 - 50 ETc 90 Kg N, T7 - 100 ETc 30 Kg N, T8 - 100 ETc 60 Kg N, T9 - 100 ETc 90 Kg N, T10 - 150

ETc 30 Kg N, T11 - 150 ETc 60 Kg N, T12 - 150 ETc 90 Kg N.

Equal lowercase letters do not differ from each other, when comparing between storage periods, and equal capital letters also do not differ from each other, when comparing treatments within each storage period, according to the Tukey test at a 5% probability

Seedling length is a widely used test for the evaluation of the vigor of cotton seeds. Currently, these tests are being widely used in seed analysis laboratories, because they have the advantages of being inexpensive, and of presenting a high correlation with the vigor in the field. Nowadays, these analyses are already automated, being performed through computerized analysis of seedlings (Marcos-Filho, 2009, Rodrigues *et al.*, 2020) [11, 18].

The demand for increased productivity in the cotton crop, and the strict relationship of seed quality with crop productivity have motivated the increase in the production of high-quality seeds, and, consequently, in research in the Seed Production and Technology area.

It is known that regions of altitude above 700 meters are recommended for the production of high quality seeds (Bewley *et al.*, 2013) [4]. However, the Umuarama region, to which this experiment was implemented, is at an altitude of 401 meters. Through this experiment, it was possible to observe that it is feasible to obtain quality seeds in a low-altitude region. Since the newly harvested seeds presented high physiological quality, regardless of the water depth treatment and the nitrogen rate. In addition, even after the six-month storage, they still presented high germination (Fig 1), and vigorous seedlings (Fig 3), besides not having reduced the percentage of germination with storage in some treatments (Fig 1).

Since the demand for quality seeds has been increasing and the tendency is to explore different areas for seed cultivation, this research generates subsidies for seed producers with regard to seed production in a low-lying region.

Thus, it is possible to conclude that the water depths and nitrogen rates applied to the plants affect the productivity, but not the initial physiological quality of the seeds. In addition, the absence of irrigation significantly reduces seed yield. However, it is still possible to obtain high-quality seeds in a low-lying region.

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