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# Effect of nitrogen doses on disease severity and watermelon yield

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## ABSTRACT

Nitrogen fertilization is an important step for watermelon (*Citrullus lanatus*) production due to its influence over yield, fruit quality, and disease severity. Currently, the gummy stem blight (*Didymella bryoniae*) and the downy mildew (*Pseudoperonospora cubensis*) can be taken as the most important watermelon diseases, since they impose severe impairment to the crop. Furthermore, studies focusing on plant responses to nitrogen fertilization regarding fruit yield and quality, and disease resistance are rare. Hence, the present study aimed at evaluating the effect of nitrogen doses on fruit yield and quality, and on disease prevalence in watermelon. Two experiments were carried out at the Universidade Federal de Tocantins, employing sprinkler irrigation, in an area previously used to grow watermelon. The experimental design was blocks at random, with five treatments (N doses, applied twice as side-dressing, in kg ha$^{-1}$, as follows: T1= control treatment without N, T2= 20, T3= 40, T4= 80, and T5= 120), and four replications. Urea (45% N) was used as the N source. In the first assay, the highest fruit yield and average weight were observed when 40 kg ha$^{-1}$ of N were applied. The highest severity of the gummy stem blight was observed when the highest nitrogen doses were applied (80 and 120 kg ha$^{-1}$). In the second assay, the highest severity levels of the gummy stem blight, as well as of mildew, were observed again when N doses were the highest (80 and 120 kg ha$^{-1}$). The lowest severity levels for both diseases were observed in the control treatment.

**Keywords:** *Citrullus lanatus*, *Didymella bryoniae*, *Pseudoperonospora cubensis*, mineral nutrition, production.

## RESUMO

**Influência de doses de nitrogênio na severidade de doenças e na produtividade da melancia**

A adubação nitrogenada é importante para a cultura da melancia (*Citrullus lanatus*) por afetar diretamente a produtividade, a qualidade dos frutos e a severidade de doenças. Atualmente, o crestamento gomoso (*Didymella bryoniae*) e o míldio (*Pseudoperonospora cubensis*) podem ser consideradas as doenças mais importantes da melancia devido aos prejuízos que podem causar na lavoura. Estudos envolvendo a aplicação de doses de nitrogênio e seus efeitos na produtividade, qualidade de frutos e manifestação de doenças são escassos. Desta forma, o presente trabalho teve como objetivo avaliar diferentes doses de nitrogênio e seus efeitos na produtividade, características dos frutos e na prevalência de doenças da melancieira. Foram conduzidos dois ensaios na Universidade Federal do Tocantins, sob irrigação por aspersão, em área anteriormente cultivada com melancia. Foi utilizado o delineamento experimental de blocos ao acaso, com cinco tratamentos (doses de nitrogênio, aplicados em duas vezes em cobertura, em kg ha$^{-1}$, sendo T1= testemunha sem nitrogênio em cobertura, T2= 20, T3= 40, T4= 80 e T5= 120) e quatro repetições. A uréia foi utilizada como fonte de nitrogênio (45% de N). No ensaio I, verificou-se maior produtividade e maior peso médio de frutos no tratamento que recebeu 40 kg ha$^{-1}$. Níveis mais altos de severidade do crestamento gomoso foram verificados nos tratamentos que receberam maiores dosagens de nitrogênio (80 e 120 kg). No ensaio II, nos tratamentos com 80 e 120 kg ha de N foram observados os níveis mais altos de míldio e crestamento gomoso. Menores níveis dessas doenças foram verificados no tratamento testemunha.

**Palavras-chave:** *Citrullus lanatus*, *Didymella bryoniae*, *Pseudoperonospora cubensis*, nutrição mineral, produção.

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Watermelon (*Citrullus lanatus* (Thunb.) Matsum. & Nakai) is the leading crop worldwide within the family Cucurbitaceae. In Brazil, watermelon is among the most important vegetable crops, both for the excellent economic return, as for the labor-intensive demand (Alencar, 2001). In 2006, Brazil has planted 80,641 ha with watermelon and harvested 1,505,133 tons, reaching an average yield of 18.7 t ha$^{-1}$. Currently, the country produces the world's fifth largest volume of fruits, second only to China, Turkey, Iran, and the U.S., and has the fifth largest cultivated acreage. However, the country ranks only 47$^{th}$ worldwide in terms of yield (FAO, 2007).

The State of Tocantins produces about 3,500 ha of watermelon, grown mainly between rice seasons in low land areas under sub-irrigation (Santos *et al*., 2005b). The production is concentrated in the southern region of the State, especially in the town of Formoso do Araguaia, where the average yield reaches 30 t ha$^{-1}$ (Santos *et al*. 2001). There are also some fields in the highlands, where pressurized irrigation, sprinkler or localized, is employed.

Watermelon is highly demanding in nitrogen, which is essential to plant development and boosts both plant growth and crop yield. However, N in excess affects fructification and confers an aqueous texture to fruits (Carmello, 1999). Moreover, high concentration of N in the plant reduces the production of phenolic compounds (fungistatic) and lignin in leaves, lessening the resistance to pathogens. Nitrogen also increases the concentration of amino acids and amines in the apoplast and leaf surface, which apparently has more influence than sugars in conidia germination, therefore favoring the development of fungal diseases (Marschner, 1986). Consequently, if the N dose applied to watermelon in one hand may increase the yield, in the other hand, it may enhance losses caused by diseases. The search for a balance in N application enhances the need for more investigation on the subject.

Dozens of pathogens, with a broad range of symptoms, can infect watermelon. In addition to diseases, there are also abiotic stresses that can cause crop total loss if preventive measures are not taken (Santos *et al.*, 2005b). According to these authors, the main watermelon diseases are the gummy stem blight, mildew, fruit rots, and viruses. The gummy blight is caused by the fungus *Didymella bryoniae* (Aversw) Rehm and produce stem lesions that exudates gum, in addition to inducing necrotic spots on leaves and damping-off and death of young plants. The gummy blight can reduce yield in up to 19.2% (Santos *et al.*, 2005a). The downy mildew, caused by the fungus *Pseudoperonospora cubensis* (Berck. et Curt.) Rostov., has major relevance when cooler temperatures prevail, especially at night, particularly if it happens in association with leaf wetness and relative humidity above 90%. The downy mildew can be responsible for crop total loss (Santos *et al.*, 2005c).

Studies on plant response to nitrogen and its effects on fruit yield and quality, as well as on disease resistance, are scarce for watermelon. Thus, the present work was designed to evaluate the effect of different doses of nitrogen on yield and other fruit characteristics and also on disease severity in watermelon.

## MATERIAL AND METHODS

The experiments were carried out at the Universidade Federal de Tocantins, Experimental Station of the *Campus* of Palmas, 280 m of altitude. The experiments were planted in an area where watermelon has been previously grown for three consecutive years, to ensure natural inoculum. Data on precipitation and temperature were collected at the meteorological station of the *Campus* of Gurupi. The soil in the experimental area was classified as Typic Hapludox, according to EMBRAPA (1999).

About two months before soil preparation, 2 t ha$^{-1}$ of lime were incorporated into the soil. The soil was prepared by one subsoiling, one plowing and two harrowing operations. Subsoiling was applied to break through an impediment layer and thus improve water infiltration. The fertilization consisted of 800 kg ha$^{-1}$ of the formulation 05-25-15 (NPK) applied in the hole at planting.

We used the cultivar Crimson Sweet which, despite being susceptible to the major diseases, is largely cultivated in Brazil and well accepted in the market. The spacing between rows and plants was 2 x 2 m, respectively. Each plot consisted of two 6-plant 12-m rows; in total twelve plants per plot. Seeds were treated with the fungicide Thiram, 1.5 g kg$^{-1}$ of seeds, to minimize potential problems caused by soil pathogens during and immediately after germination. The experimental design was randomized blocks with five treatments and four replications. Treatments consisted of different nitrogen doses (kg ha$^{-1}$), applied on side-dressing, using urea as the N source (45% N), where: T1= control without nitrogen, T2= 20, T3= 40, T4= 80, and T5= 120. Side dressing was split in two fertilizations, the first, 28 days after planting (DAP), and, the second, 35 DAP. Along with the fertilization with urea, 104 kg ha$^{-1}$ of potassium chloride (58% $K_2O$) were applied, divided in two equal doses. Four to five seeds per hole were sown at 2 cm deep.

Thinning was performed 22 DAP, when plants had three true leaves (Carvalho, 1999), leaving two plants per hole. Two hand hoeing, 20 and 30 DAP, respectively, were performed to remove weeds. We employed sprinkler irrigation, with 12-hour irrigation shifts (one irrigation in the morning and another in the afternoon), to avoid the hours of strong winds. The average water depth was 7.9 mm day$^{-1}$, measured by collectors placed in the center of each plot.

To control pests, such as the Cucurbit beetle, aphids, and leaf miners, we used insecticides, sprayed in the late afternoon, not to coincide with bee visitation. In total there were five sprays, three with Deltamethrina (100 mL 100 L$^{-1}$ of water) and two with Acephate (100 g 100 L$^{-1}$ of water). Fungicides were not used.

Fruits were harvested manually from 85 DAP ahead. The commercial harvest was indicated by the drying of the first tendril, located at the insertion of the fruit peduncle into the stem and/or by the color of the side of the fruit which was in contact with the ground that changes from white to cream, in association with the sound of hollow produced by the ripe fruit when tapped with a firm hand. Only fruits considered commercial, weighing at least 5.0 kg, were taken into account (weighing in digital scale) to estimate the commercial yield and fruit average weight.

Assessments of the gummy blight in leaves (GBL) and of the downy mildew were carried out after the first harvest, according to methodology described by Santos *et al*. (2005a). To this end, we used the midpoint of the grading scale from 0 to 9, where 0= healthy tissue, 1= less than 1% of damaged tissue, 3= between 1 and 5% of damaged tissue, 5= between 6 and 25% of damaged tissue, 7= 26 to 50% of damaged tissue, and 9= more than 50% of damaged tissue. In total, we carried out four disease evaluations, 50, 60, 70, and 80 DAP. For the statistical analysis, we used the average severity among evaluations.

**Assay I -** The experiment was carried out from July to October 2006, when the monthly precipitations were 0, 3, 50, and 0.8 mm, respectively. The monthly maximum/minimum temperatures were 33.6/15.4, 35.1/17.1, 35.1/20.4, and 35.1/23.4°C, respectively. The results of the soil analysis for the samples collected at 0-20 cm deep were: pH (CaCl2) 5.0, 0.17 cmolc dm$^{-3}$ of Al, 2.79 cmolc dm$^{-3}$ of H+Al, 2.93 cmolc dm$^{-3}$ of Ca+Mg, 10.1 mg dm$^{-3}$ of P, 10.17 mg dm$^{-3}$ of K, 5.75 cmolc dm$^{-3}$ of cation exchange capacity, 2.3 g dm$^{-3}$ of organic matter, and 51.4 of saturation, according to the method recommended by EMBRAPA (1997). In this assay, there was no occurrence of mildew. Therefore, only the gummy blight in leaves was evaluated.

**Assay II -** The experiment was set in the same location of assay I, from March to June 2007. In March and April, the monthly precipitations were 151 and 20.8 mm respectively. In the following two months there was no rainfall. In this experiment, it was possible to assess the severity of both the gummy blight and the downy mildew. However, due to high level of infection in plants, the number and average fruit weight were not assessed and, consequently, neither yield.

Regression models were adjusted for the two trials. We performed also a correlation analysis for the variables studied in assay I, using the program Sisvar (Ferreira, 1999).

## RESULTS AND DISCUSSION

**Assay I -** The nitrogen doses directly influenced yield and the average weight of marketable fruits, adjusting to a quadratic model. There was an increase in yield from the first N dose (20 kg ha$^{-1}$) up to 40 kg ha$^{-1}$ (Figure 1A). The use of 40 kg ha$^{-1}$ of N increased the watermelon marketable fruit yield by 14 tons ha$^{-1}$ when compared to the control, without nitrogen fertilization. From 40 kg ha$^{-1}$ of N ahead, according to the regression function, yield started declining, possibly agreeing with the Law of Maximum, and probably also due to high severity levels of the gummy blight in the treatments in which larger N doses were applied (Figure 1C). According to Fageria *et al*. (1999), the quadratic model has been the one that best represents crop response to nitrogen. The adjustment of our results to a quadratic function suggests that the N doses used in this study were appropriate, since it was observed an increase both in fruit yield and average size in the initial doses (20 kg ha$^{-1}$ of N), reaching a saturation point (40 kg ha$^{-1}$ of N) to decrease in the highest doses (120 kg ha$^{-1}$ of N). All $R^2$ values were significant ($p<0.001$) according to the table of Little & Hills (1978).

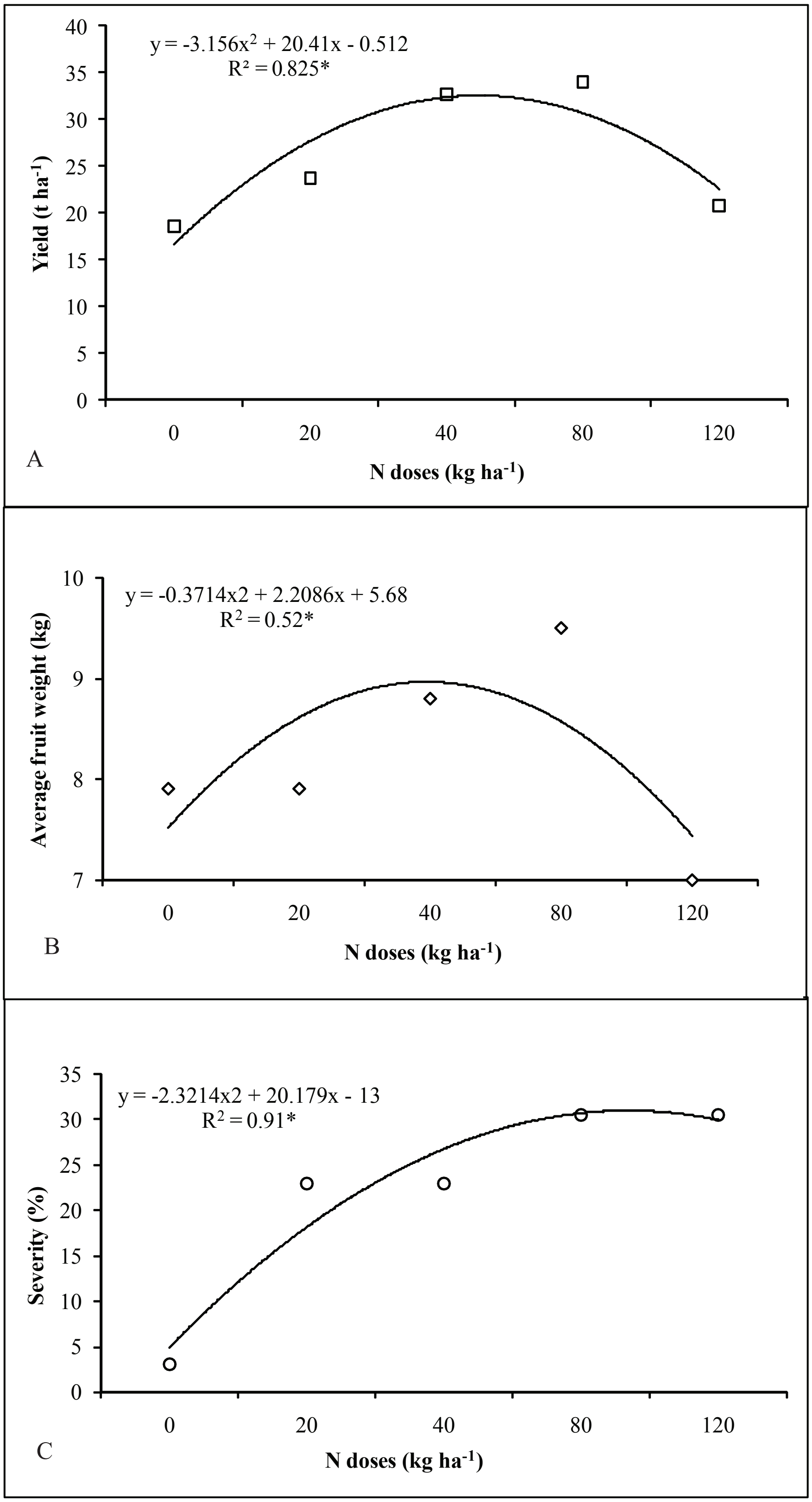


**Figure 1.** Yield (**A,** kg ha$^{-1}$), average fruit weight (**B,** g), and the gummy stem blight severity (**C, %**) in watermelon as affected by increasing nitrogen doses (produtividade (**A,** t ha$^{-1}$), peso médio de fruto (**B,** kg), severidade de crestamento gomoso do caule da melancia (**C,** %) em função de doses crescentes de nitrogênio). Gurupi, UFT, 2006.
*$R^2$ significant, Little & Hills (1978), p<0.01, 22 df (significativo, Little & Hills (1978), p<0,01, 22 graus de liberdade).

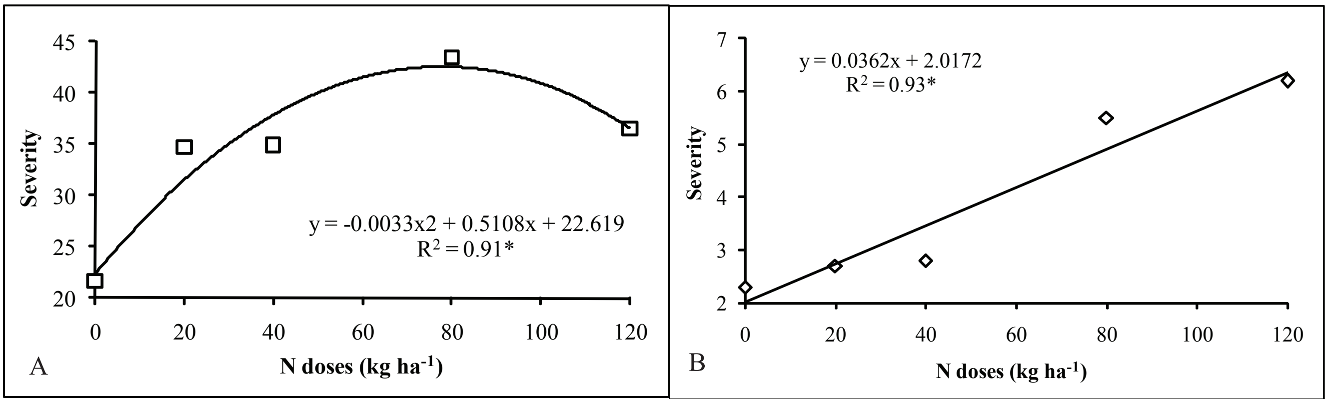


**Figure 2.** Severity of the downy mildew (**A**) and the gummy stem blight (**B**) in watermelon as affected by increasing nitrogen doses (severidade do míldio (**A**) e do crestamento gomoso do caule (**B**) em melancia em função de doses crescentes de nitrogênio). Gurupi, UFT, 2007. *$R^2$ significant, Little & Hills (1978), $p<0.01$, 22 df (significativo, Little & Hills (1978), p<0,01, 22 graus de liberdade).

Similar results were obtained by Andrade Junior *et al*. (2006), who showed that the watermelon commercial production increased with the application of a nitrogen fertilizer. The same authors found that, contrary to yield, fruit quality was not significantly affected by nitrogen. These authors however did not comment on disease incidence. Therefore, we believe that the conditions of the experimental areas with regard to inoculum sources were different between our work and that reported by Andrade Junior *et al*. (2006). Consequently, the results of both reports can not be compared without reservation, since it is well known that diseases directly influence yield.

According to the regression equation obtained, the N mounting doses resulted in increases in fruit weight up to 40 kg $ha^{-1}$ of N, with fruits reaching an average weight of 8.8 kg (Figure 1B). When 120 kg $ha^{-1}$ of N were used, the average weight of marketable fruits was merely 7 kg.

Research carried out under various cropping conditions has revealed that the adequate commercial yield is obtained in watermelon when 50 to 120 kg $ha^{-1}$ of N are applied. Singh & Naik (1989) tested 50, 100, 150, and 200 kg $ha^{-1}$ of N and concluded that N doses above 50 kg $ha^{-1}$ resulted in excessive vegetative growth and decrease in fruit yield. In our study, N application resulted in an increase in both fruit yield, and weight only up to 40 kg $ha^{-1}$. From these doses, both yield, and the average weight of marketable fruits decreased (Figure 1A), probably due to (1) the profuse vegetative growth observed when higher doses of N were used and (2) and the higher disease severity observed when the largest N doses were used. Malavolta *et al*. (1997) stated that although the symptoms of nitrogen excess are not well identified, reduction in fruit set can definitely take place. In watermelon, nitrogen deficiency decreases the number of hermaphrodite flowers and determines fruit shape, as well as the fruit color hue that becomes lighter. In addition, it causes a collapse in plant metabolism at the time of fruit growth, limiting plant growth as a whole (Garcia & Souza, 2002).

Positive and significant correlation ($r= 0.88$, $p<0.01$) was found between fruit weight and commercial yield. In contrast, the correlation between the gummy blight severity and yield was not significant. This result indicates that other factors besides those investigated in this work may be directly involved in yield.

The evaluation of the gummy blight severity indicated that the use of increasing doses of N favored the watermelon infection by *Didymella bryoniae*. Higher severity levels were observed in treatments with the highest N doses (80 kg and 120 kg $ha^{-1}$). The control treatment, which did not receive N, showed the lowest disease severity on leaves (Figure 1C). Where the infection by *D. bryoniae* was more severe, i.e., in the treatments with 80 kg $ha^{-1}$ of N or more, there was reduction in yield and in the average weight of marketable fruits (Figures 1A and 1B, respectively). It is known that greater variation between day and night temperatures result in leaf wetness. In our study, there was a thermal gradient of around 15°C between the maximum and minimum temperatures, which favored the infection, despite the low precipitation.

**Assay II -** As we increased the N dose, the attack of the downy mildew (*Pseudoperonospora cubensis*) also tended to become more severe. The treatments using 80 and 120 kg $ha^{-1}$ of N showed the highest values for disease severity (Figure 2A). On contrary, the lowest severity was found in the control, where no N was added. There were no significant differences in mildew severity when we compared plants that received 20 and 40 kg $ha^{-1}$ of N.

For the gummy blight in leaves (*D. bryoniae*), there was also a trend of linear increase of disease severity with N doses. Higher severity levels occurred in the treatments in which 80 and 120 kg $ha^{-1}$ of N were applied, while the lowest severity occurred in the control, without N addition (Figure 2B). As observed for mildew, no significant differences were observed in the gummy blight severity when we compared plants receiving 20 and 40 kg $ha^{-1}$ N.

In this experiment, rainfalls occurred only in March and April, respectively 151 and 20.8 mm. In the following months, we used sprinkler irrigation, which also creates a microclimate favorable to the pathogen.

As far as we know, there were no reports in literature about the effect of nitrogen over the gummy blight and the downy mildew in watermelon. However, Santos *et al*. (2005a), in conditions similar to ours of restricted rainfall, an ecological situation that can be considered unfavorable to the disease, found 26.5% of leaf area damaged due to infection only by *D. Bryoniae*, which resulted in losses of 19.2% in yield. In the current study, in assay II, due to heavy attack of mildew and gummy, losses went up to 100%, since no fruits fit the commercial standard size, i.e., at least 5.0 kg.

It is important to note that we should not make a straight comparison between the results obtained in this work and the routine of watermelon commercial fields. In our study, to make it possible to assess N influence over the severity of the downy mildew and the gummy blight, there has been no fungicide spraying. Conversely, in commercial areas, fungicides are sprayed up to seven times during the crop season, depending on the environmental conditions and disease severity.

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