

# RAMULARIA LEAF SPOT AND BOLL ROT ARE AFFECTED DIFFERENTLY BY ORGANIC AND INORGANIC NITROGEN FERTILIZATION IN COTTON PLANTS

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## Abstract

Interaction among nitrogen fertilization using bovine manure, poultry manure, *Jatropha curcas* seed cake and urea, and the diseases Ramularia leaf spot (RLS) and Boll rot (BR), caused by *Ramulariopsis pseudoglycines* and *Diplodia gossypina*, respectively, in cotton plants (*Gossypium hirsutum* L.), was studied under field conditions. Intensity (incidence and severity in percentage) of RLS and incidence (%) of BR were evaluated over time, starting in reproductive stage B1 (first visible flower bud). A randomized complete block design with a 4x4 factorial arrangement was used (fertilizers x doses), totaling 16 treatments with four replications. Disease progress was analyzed with the nonlinear Logistic and Gompertz models, obtaining the epidemiological parameters amount of initial disease ( $Y_0$ ) and progress rate ( $r$ ). Plants fertilized with 50 kg N ha<sup>-1</sup>, presented an incidence twice greater than those obtained with other fertilizers. The Logistic model better fits RLS, but no model could represent BR. Only the epidemiological parameters of RLS were affected differently in this experiment compared to BR disease. The possible role of organic and inorganic nitrogen fertilization in the RLS and BR management is discussed.

**Keywords:** Disease progress. *Gossypium hirsutum* L. Logistic and Gompertz models. Nitrogen dose. Nitrogen fertilization.

## 1. Introduction

Cotton (*Gossypium hirsutum* L.) is currently one of the main crops worldwide (Fang 2018), producing a fiber constituted by complex structures almost exclusively composed of cellulose molecules and D-glucose residues (French and Kim 2018). Although this species had a significant participation in Ecuador's agricultural sector between 1970-1990, mainly in the Guayas and Manabí provinces, the sown area was affected by climatic and economic factors and by the lack of certified seeds (García et al. 2019). Between the end of the last century and the last decade in the country, cotton fiber production (t) and planted area (ha) decreased 21 and 29 times, respectively, with Manabí province representing 79% of national total production (Vivero 2017). Experimentally, some cotton varieties have been studied in this province, such as Coker and DP-ACALA 90 (Cañarte-Bermúdez et al. 2020).

One of the most important tasks in cotton cultivation management is fertilization (Teixeira et al. 2008). Nitrogen (N) is the most required nutrient by the plant since it is a limiting factor for growth and a fundamental element for crop production (Bondada and Oosterhuis 2001). Although cotton plants respond positively to N incorporation, this response can be affected by genotype, soil type, and humidity conditions during its development (Singh 1970). Nitrogen application has a significant impact on physiological parameters, cotton growth, boll development, staple yield, and fiber quality (Bondada and Oosterhuis 2001; Teixeira et al. 2008).

Although the use of nitrogen sources of synthetic origin mainly based on ammonia (NH<sub>3</sub>) increases crop yield, this can be detrimental to an agricultural system. The loss of N as NH<sub>3</sub> causes a hostile situation between the use of this resource and the conservation of the environment, driving even a low efficiency of this type of fertilizer when applied to a crop (Zheng et al. 2018). In addition, atmospheric pollution, soil acidification, eutrophication, and biodiversity reduction may be observed (Scudlark et al. 2005). As an alternative to this type of management, more environmentally-friendly practices emerged, such as using organic nitrogen sources rich in ammonium (NH<sub>4</sub>).

Inorganic and organic fertilizers could exert suppressive effects on plant pathogens, the type of nitrogen source, and the plant-pathogen interaction that may be influenced. In fact, N applications in a crop can increase or decrease the resistance of plants to pathogens, showing differences in the strategies of pathogens to infect plant tissues (Mur et al. 2016). Likewise, these fertilizers can introduce biocontrol agents into the soil, providing nutrients for its establishment and activity (Artavia et al. 2010), improving root condition, promoting adequate plant growth and boll quality, making them more resistant against diseases (Mur et al. 2016; Chen et al. 2018; 2019; 2020). Rich N sources are known to negatively affect various plant pathogens (Blachinski et al. 1996; Artavia et al. 2010; Veromann et al. 2013). This element also affects physical defenses and antimicrobial phytoalexin production in plants but has a positive effect on defense-related enzymes and proteins that induce local protection and systemic resistance (Sun et al. 2000). However, effect of these sources effect on diseases that attack foliar tissues in cotton plants is unknown, especially in their epidemiological parameters such as initial disease (*Y<sub>0</sub>*) and progress rate (*r*). Nevertheless, in this context, understanding how pathogens with different infection strategies respond to N levels is of fundamental importance (Mur et al. 2016).

On the Ecuadorian coast, the cotton crop is affected by diseases such as damping off (*Rhizoctonia solani*), cotton anthracnose (*Colletotrichum gossypii*), and Boll rot (*Diplodia gossypina*) (Si3n et al. 1992). Other diseases, like *Ramularia leaf spot* (*Ramulariopsis pseudoglycines*), present in neighboring countries (Aquino et al. 2008) and not yet reported in Ecuador, can negatively affect the leaf area of cotton plants and fiber yield (Ascari et al. 2016; da Silva et al. 2019). Despite the importance of these diseases, there is little bibliography about them in Ecuadorian cotton crops. Even studies evaluating the effect of organic fertilizers on cotton cultivation are scarce. Thus, this study evaluated the interaction of cotton crop (variety DP ACALA 90) above ground diseases with organic fertilization (bovine manure, chicken manure, or *Jatropha curcas* seed cake), and synthetic (rich in N) fertilization (urea).

## 2. Material and Methods

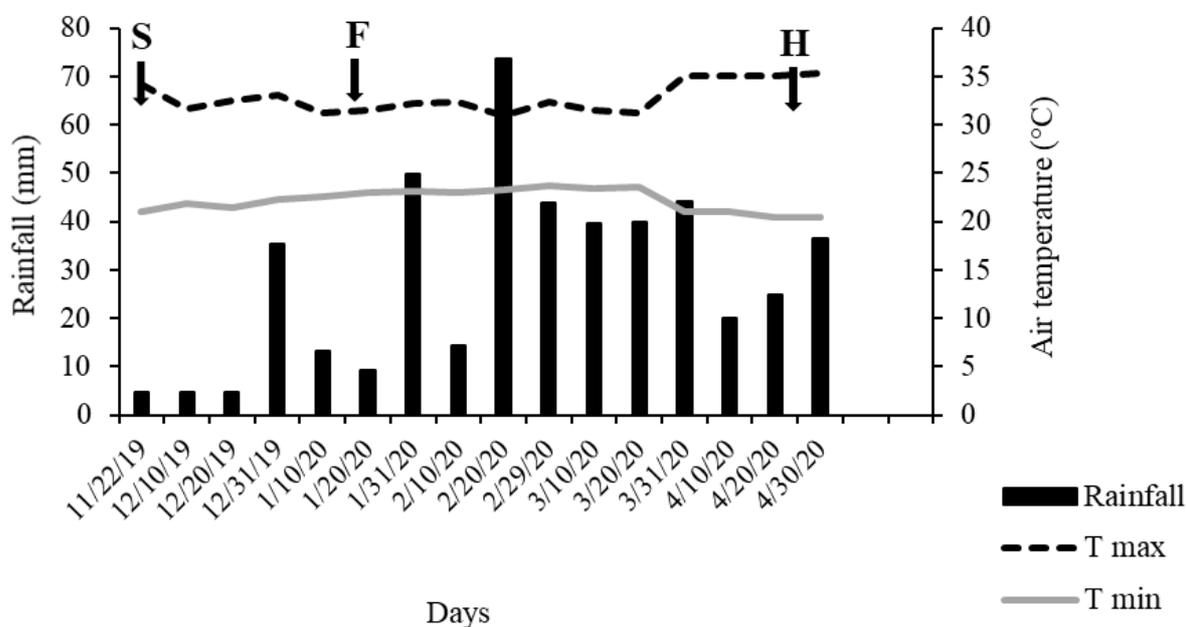
### Study area and Field experiment

This research was done between November/2019 and April/2020, at the La Teodomira experimental campus, of the Faculty of Agronomic Engineering, Technical University of Manabí (UTM), Ecuador (01° 09' 51 S, 80° 23' 24" W, 60 m altitude) (INAMHI 2015).

Cotton DP ACALA 90 was sown under field conditions, 0.40 m between plants, 1 m between rows, and 2 m between blocks, in an area where cotton was grown in the previous cycle. Soil type was clay loam, according to the USDA soil taxonomy (Soil Survey Staff, 2014), obtained by soil analysis in the Laboratory of Soils, Plant Tissues, and Water of the National Institute of Agricultural Research (INIAP), Tropical Experimental Station Pichilingue (Table 1). Rainfall and maximum and minimum air temperatures (T max and T min), recorded in Lodana, Manabí, Ecuador during the experiment are shown in Figure 1.

**Table 1.** Physical characteristics (soil type and pH: hydrogen ionic potential) and chemical (OM: organic matter, N: Nitrogen, P: Phosphorus, K, Potassium, Ca: Calcium, Mg: Magnesium, H: Hydrogen, Mn: Manganese, Co: Cobalt, and Z: Zinc).

Soil	pH	MO %	N %	P	K	Ca	Mg	H	Mn	Co	Z
				mg kg <sup>-1</sup>	mg kg <sup>-1</sup>	cmol kg <sup>-1</sup>	cmol kg <sup>-1</sup>	mg kg <sup>-1</sup>			
Clay loam	7.5	0.90	0.04	17.4	1.06	15.25	5.27	26.7	5.55	2.19	<2.60



**Figure 1.** Rainfall and maximum and minimum air temperatures (T max and T min) of Lodana, Manabí, referring to the experimental period (November 22, 2019 to April 30, 2020) with indication of sowing (S), beginning of flowering (F) and harvest (C).

The total experimental area was 2688 m<sup>2</sup> (48 m x 56 m), where each experimental plot consisted of 36 m<sup>2</sup> (6 m x 6 m) with a useful area of 20.8 m<sup>2</sup> (5.20 m x 4 m). Fertilizers used as N sources were bovine manure, *Jatropha curcas* seedcake, poultry manure, or urea (the latter used as a control). The first fertilization was performed 20 days after emergence (DAE), placing half the dose that corresponded to it in each treatment, the second fertilization was done at flowering (50 DAE), completing the exact dose of nitrogen from its sources (50, 100, 150, 200 kg of N ha<sup>-1</sup>). The concentrations of nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), and magnesium (Mg) of each organic fertilizer are shown in Table 2.

### Disease assessment

Disease intensity was evaluated for RLS (*R. pseudoglycines*) and BR (*Diplodia* sp.), at the beginning of the reproductive phase (B1) (76 DAE) (first visible flower bud) (Marur and Ruano 2004), in the lower, middle, and upper canopy (a plant organ for each) of four cotton plants from the useful plot, totaling six evaluations over time. Disease incidence was measured by counting each of the symptomatic organs (leaves and capsules) present in plants, employing a visual observation, and transforming the average value of each stratum into a percentage. Disease severity was only estimated for RLS, using the diagrammatic scale proposed by Aquino et al. (2008).

### Experimental design and statistical analyses

A randomized block design with a 4x4 factorial arrangement was used (fertilizers x dose), totaling 16 treatments with four replications. After confirming the normality and homoscedasticity of the values obtained, variance analysis was performed, and the averages were compared using the Tukey test ( $P < 0.05$ ).

Furthermore, Pearson's analysis was performed to investigate the correlation among all the variables evaluated.

For the analysis of plant disease progress, the primary inoculum ( $Y_0$ ) and the progress rate ( $r$ ) were estimated using Logistic (Eq. 1) and Gompertz (Eq. 2) model equations (Berger 1981; Tjørve and Tjørve, 2017):

$$y=1(1+\exp(-a+rt))^{-1} \quad (1)$$

where  $y$  = disease proportion ( $0 < y < 1$ ),  $a$  = logit ( $y_0$ ),  $r$  = rate, and  $t$  = time

$$y=\exp(-B \cdot \exp(-kt)) \quad (2)$$

where  $B$  is a position parameter,  $k$  = rate, and  $t$  = time.

Averages were compared when the probability for all the values of a factor were significant, using the Tukey test ( $P < 0.05$ ). Progress curves were plotted using the epifitter package (Alves and del Ponte 2020). All analyzes were performed with Rstudio (RStudio Team 2017).

**Table 2.** Concentrations (%) of Nitrogen (N), Phosphorus (P), Potassium (K), Calcium (Ca), and Magnesium (Mg) of organic fertilizers Bovine manure, Seedcake, and Poultry manure.

Fertilizers	Concentrations (%)				
	N	P	K	Ca	Mg
Bovine manure	3.9	0.6	1.6	1.4	0.5
<i>Jatropha curcas</i> seedcake	2.1	0.9	2.6	0.9	0.6
Poultry manure	3.0	0.7	2.3	2.7	0.6

### 3. Results

Factorial analysis showed a significant interaction between fertilizers and doses only for RLS incidence ( $P < 0.0194$ ). In contrast, for Boll rot incidence, only differences between doses were observed ( $P < 0.02519$ ) (Table 3).

**Table 3.** Results of two-way analysis of variance for average incidence and severity of Ramularia leaf spot and incidence of Boll rot.

Variable	Factors	GL <sup>1</sup>	F <sup>2</sup>	P – value
Ramularia leaf spot ( <i>Ramulariopsis pseudoglycines</i> )				
Incidence	Fertilizers	3	1.67050	0.17420
	Dose	3	0.62240	0.60121
	Interaction	9	2.25960	0.01941*
Severity	Fertilizers	3	0.45900	0.71110
	Dose	3	0.36110	0.78120
	Interaction	9	0.71340	0.69650
Boll rot ( <i>Diplodia</i> sp.)				
Incidence	Fertilizers	3	1.08210	0.35747
	Dose	3	3.16910	0.02519*
	Interaction	9	2.48790	0.06998

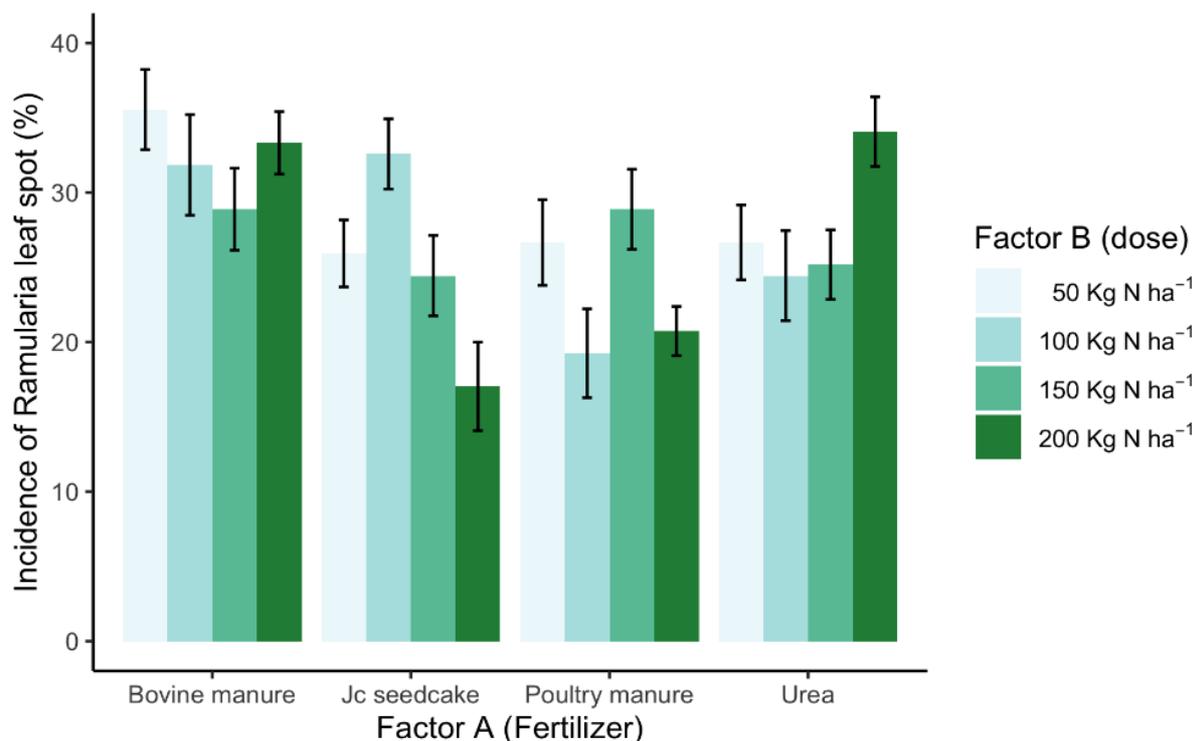
<sup>1</sup> GL: degrees of freedom; <sup>2</sup> F: Fisher's calculated value; \* statistical significance  $P < 0.05$ .

In the RLS incidence interaction analysis (Figure 2), no response pattern was observed for the doses of each fertilizer. However, cotton plants fertilized with 100 kg N ha<sup>-1</sup> of *J. curcas* seedcake and 100 and 200 kg N ha<sup>-1</sup> of poultry manure showed an RLS incidence between 16 and 21%, which were lower than the other treatments. In general, these results indicate that, regardless of the dose used the plants fertilized with bovine manure had the highest incidence of RLS (33%) compared to the other fertilizers used.

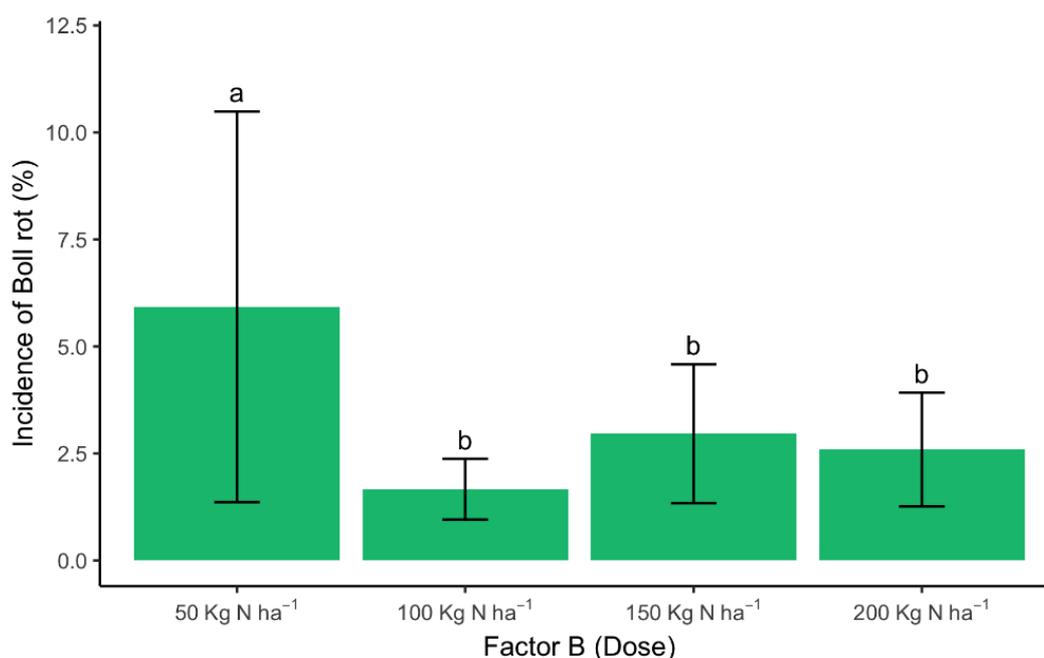
Regarding doses, cotton plants fertilized with 50 kg N ha<sup>-1</sup> presented higher BR incidence than any other fertilizer and doses (Figure 3).

No correlation was observed between incidence, severity and, intensity, with probabilities between 0.002 and 0.892 (data not shown).

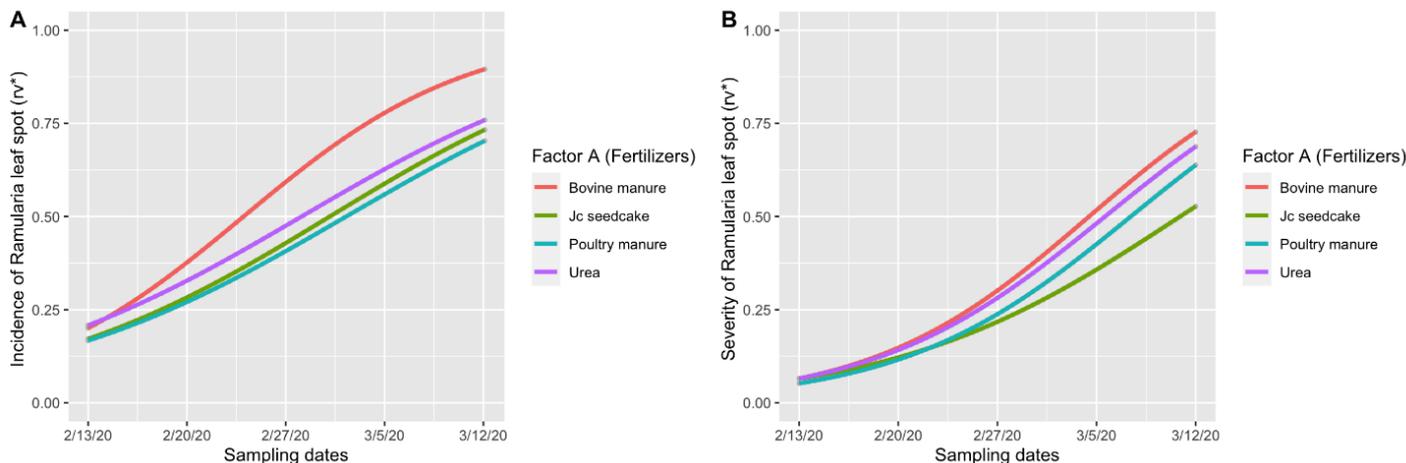
The RLS incidence progress curve in cotton plants fertilized with *J. curcas* seedcake and poultry manure started lower than in the other fertilizer treatments. However, the curve that presented the greatest proportion of final disease was in plants fertilized with bovine manure (Figure 4A). Although all the curves had similar starting points in the case of RLS severity, the endpoint of the disease found in plants fertilized by *J. curcas* seedcake was the smallest compared to the rest (Figure 4B).



**Figure 2.** Factorial interaction (fertilizers x dose) for incidence (%) of Ramularia leaf spot, on cotton plants under organic fertilization (bovine manure, *Jatropha curcas* seed cake, and poultry manure) and synthetic (urea), using four doses (50, 100, 150 y 200 kg N ha<sup>-1</sup>) for each source.

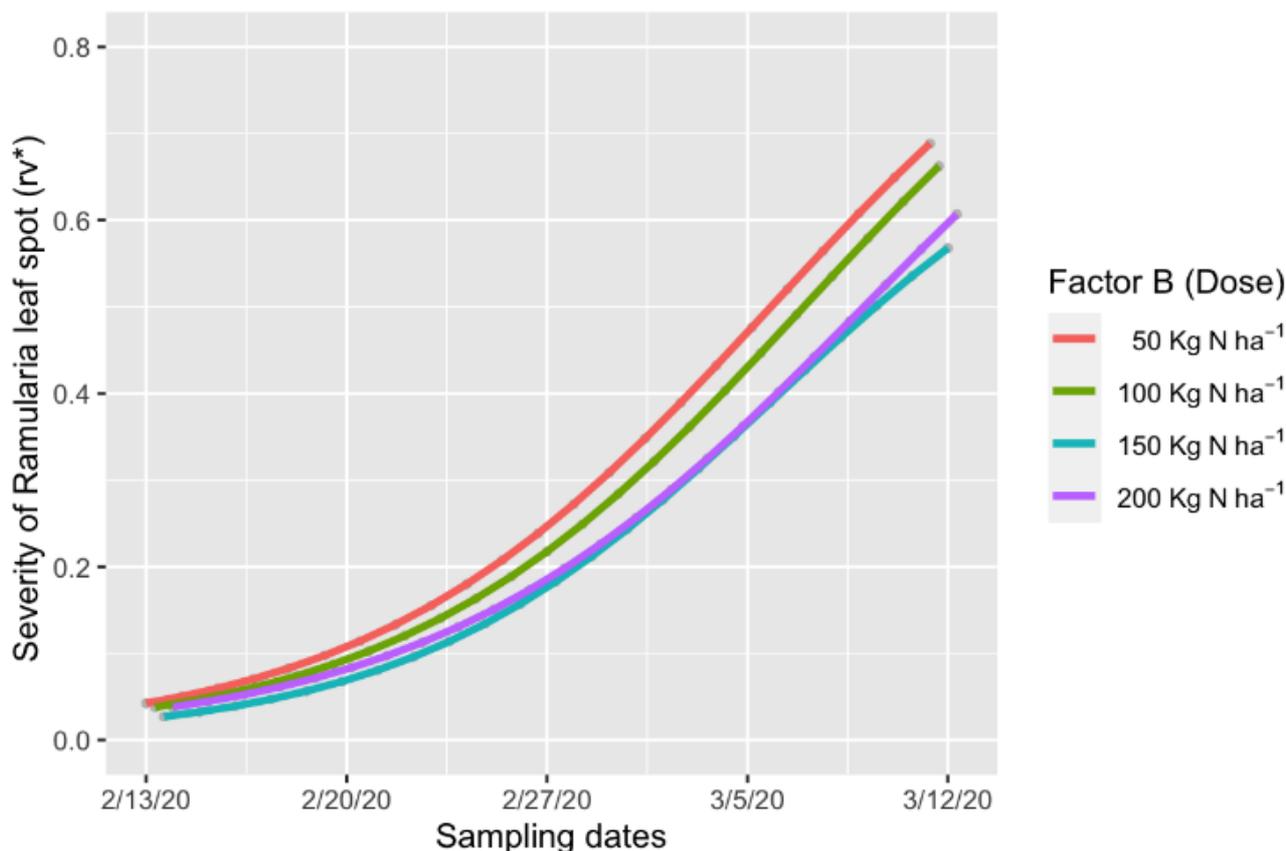


**Figure 3.** Incidence values (%) of Boll rot in cotton plants, grouped by fertilizer (bovine manure, *Jatropha curcas* seed cake, and poultry manure), using four doses (50, 100, 150 y 200 kg N ha<sup>-1</sup>) for each source. Lowercase letters indicate a significant difference by Tukey's test ( $P < 0.05$ ).



**Figure 4.** A - Temporal progress of incidence and B - severity of Ramularia leaf spot in cotton plants under organic nitrogen fertilization (bovine manure, *Jatropha curcas* seed cake, and poultry manure) and synthetic (urea), using a Logistic model.

The RLS severity (B) progress curve temporarily increased similarly, observing that plants fertilized with greater doses reached a smaller final disease proportion (Figure 5).



**Figure 5.** Temporal progress of the severity of Ramularia leaf spot in cotton plants under organic and synthetic nitrogen fertilization, using 50, 100, 150, and 200 kg N ha<sup>-1</sup>, using a Logistics model.

Concerning the modeling of epidemics using non-linear Logistics and Gompertz, the first model better represented the RLS, except for doses, where none of the curves were significant (Table 4). A lower amount of initial disease ( $Y_0$ ) for RLS was found using the Logistic model, both in the incidence (0.259) and in severity (0.081), in plants fertilized with *J. curcas* seed cake or poultry manure, compared to the other treatments. In contrast, a lower disease progress rate ( $r$ : 0.012) in plants fertilized with urea was observed, compared to the others (0.016). This same parameter for the variable severity, no difference was observed between

fertilizers. Finally, 150 (0.076) and 200 (0.028) kg N ha<sup>-1</sup>, negatively affected RLS  $Y_0$  and positively the  $r$  compared to the other doses.

**Table 4.** Amount of initial disease ( $Y_0$ ) and disease progress rate ( $r$ ) obtained from the quantification of incidence and severity (%) of *Ramularia* leaf spot, over time in cotton plants under organic fertilization (bovine manure, *Jatropha curcas* seed cake, and poultry manure) and synthetic (urea), using four doses (50, 100, 150 and 200 kg N ha<sup>-1</sup>) for each source. From the analysis obtained for the models Logistic and Gompertz, the coefficient of determination was obtained ( $R_2$ ) and the probability ( $P$ ).

Fertilizer	Logistic				Gompertz			
	$Y_0$	$r$	$R_2$	$P$ -value	$Y_0$	$r$	$R_2$	$P$ -value
Incidence (%) of <i>Ramularia</i> leaf spot								
Factor A (Fertilizers)								
Bovine manure	0.354 a <sup>ε</sup>	0.015 a	0.34	0.00001	0.867	0.040	0.39	0.07078
<i>J. curcas</i> seedcake	0.263 b	0.016 a	0.31	0.00004	0.615	0.074	0.37	0.03309
Poultry manure	0.254 b	0.016 a	0.32	0.00003	0.583	0.079	0.39	0.02156
Urea	0.327 a	0.012 b	0.29	0.00032	0.648	0.064	0.28	0.09145
Factor B (Dose)								
50 kg N ha <sup>-1</sup>	0.487	0.000	0.00	0.59825	0.533	0.511	0.05	0.40871
100 kg N ha <sup>-1</sup>	0.485	0.000	0.00	0.98839	0.502	0.951	0.06	0.47845
150 kg N ha <sup>-1</sup>	0.429	0.004	0.02	0.31305	0.506	0.333	0.06	0.34263
200 kg N ha <sup>-1</sup>	0.473	0.000	0.00	0.99526	0.469	0.073	0.00	0.95881
Severity (%) of <i>Ramularia</i> leaf spot								
Factor A (Fertilizers)								
Bovine manure	0.099 a	0.026 <sup>ns</sup>	0.59	0.00000	13.600	0.007	0.59	0.00737
<i>J. curcas</i> seedcake	0.077 b	0.022	0.42	0.00000	0.652	0.028	0.45	0.09016
Poultry manure	0.074 b	0.027	0.58	0.00000	8.870	0.008	0.58	0.46556
Urea	0.102 a	0.024	0.54	0.00000	1.158	0.022	0.56	0.08271
Factor B (Dose)								
50 kg N ha <sup>-1</sup>	0.113 a	0.023 b	0.57	0.00000	0.804	0.032	0.61	0.03292
100 kg N ha <sup>-1</sup>	0.096 ab	0.024 b	0.54	0.00000	1.266	0.019	0.56	0.11966
150 kg N ha <sup>-1</sup>	0.074 b	0.028 a	0.60	0.00000	3.330	0.012	0.60	0.01596
200 kg N ha <sup>-1</sup>	0.078 b	0.027 a	0.48	0.00000	11.020	0.007	0.48	0.57280

<sup>ε</sup> An analysis of variance with its respective comparison of means was performed only when the probability for all the factor values was significant ( $P < 0.05$ ); <sup>γ</sup> Lowercase letters in the column indicate the difference between means by Tukey's test ( $P < 0.05$ ); <sup>ns</sup> There is no difference between column means by Tukey's test ( $P < 0.05$ ).

For BR incidence (%), none of the non-linear models used fitted well the parameters (Table 5). Furthermore, both  $Y_0$  and the  $r$  for 100 kg N ha<sup>-1</sup> dose could not be calculated.

**Table 5.** Amount of initial disease ( $Y_0$ ) and disease progress rate ( $r$ ) obtained from the quantification of incidence (%) of Boll rot, over time in cotton plants under organic fertilization (bovine manure, *Jatropha curcas* seedcake, and poultry manure) and synthetic (urea), using four doses (50, 100, 150 and 200 kg N ha<sup>-1</sup>) for each source. From the analysis obtained for the models Logistic and Gompertz, the coefficient of determination was obtained ( $R_2$ ) and the probability ( $P$ ).

Fertilizer	Logistic				Gompertz			
	$Y_0$	$r$	$R_2$	$P$ -value	$Y_0$	$r$	$R_2$	$P$ -value
Incidence (%) of Boll rot								
Factor A (Fertilizers)								
Bovine manure	0.026	0.040	0.09	0.16337	0.706	0.026	0.09	0.73157
<i>J. curcas</i> seedcake	0.022	0.040	0.10	0.19940	0.218	0.114	0.12	0.51944
Poultry manure	0.136	0.015	0.02	0.56079	0.271	0.114	0.05	0.40132
Urea	0.052	0.033	0.19	0.08442	0.340	0.095	0.22	0.27887
Factor B (Dose)								
50 kg N ha <sup>-1</sup>	0.083	0.033	0.23	0.26119	0.829	0.030	0.25	0.38284
100 kg N ha <sup>-1</sup>	0.049	0.013	0.01	0.62627	*	*	*	*
150 kg N ha <sup>-1</sup>	0.025	0.045	0.22	0.10295	5.900	0.001	0.23	0.77601
200 kg N ha <sup>-1</sup>	0.052	0.024	0.03	0.51379	0.188	0.088	0.05	0.58102

\* The data did not allow us to calculate the epidemiological parameters studied in the present work.

#### 4. Discussion

Currently, countless studies have shown the positive effect generated by N-rich fertilizers in cotton plants. For example, applications between 55 and 240 N kg ha<sup>-1</sup> increase canopy photosynthesis and leaf weight, thus inducing a greater number of nodes and capsules, hundred seed's weight, and fiber production (Bondada and Oosterhuis, 2001; Teixeira et al. 2008; Chen et al., 2018; 2019; 2020). However, little is known about the consequence of N fertilization in cotton plant diseases. In this study, conducted under field conditions, the performance of Ramularia leaf spot (RLS) and Boll rot (BR) is presented for the first time with the application of four sources and increasing doses of N in DP ACALA 90 cotton plants.

The cotton crop sown in the previous cycle seems to have provided a sufficient primary inoculum for RLS or BR to appear in the vegetative stages, but with greater intensity in the first disease. Indeed, RLS can accumulate a primary inoculum available in the early stages of cotton (da Silva et al. 2019). In contrast, the genotype used is susceptible to the *Cotton leafroll dwarf virus* (CLDV) transmitted by the aphid *Aphis gossypii*, and to bacterial blight caused by *Xanthomonas citri* pv. *malvacearum*. For the first time and under field conditions, this genotype is shown to be susceptible to RLS and BR diseases.

Although nutrients can affect the tolerance or resistance of plants to pathogens (Mur et al. 2016; Artavia et al., 2010; Veromann et al. 2013), regardless of the disease, plants respond differently to each fertilizer and dose of N. N-rich sources can reduce *Alternaria macrospora* spore germination and mycelial growth under *in vitro* conditions and lesion diameter and Alternariosis percentage under controlled conditions, but none of the parameters were reduced under field conditions (Blachinski et al. 1996). However, the same authors mention that this effect was generated only by some nitrogen sources, such as potassium nitrate (KNO<sub>3</sub>), which negatively affects the diameter of lesion but not the severity of disease (%). In addition, applications of different forms of N, such as nitrate (NO<sub>3</sub>) and ammonium (NH<sub>4</sub>), can act biochemically, physiologically, and molecularly in plants against pathogens (Sun et al., 2020). In this sense, it is inferred that both external factors and an effect at the cellular level of each fertilizer may have influenced the observed behavior in the present study.

Regardless of the dose used, plants fertilized with bovine manure had a greater incidence of RLS. Plants fertilized with 100 kg N ha<sup>-1</sup> of *J. curcas* seedcake and 100 and 200 kg N ha<sup>-1</sup> of poultry manure showed a lower incidence of this disease than the other treatments. According to laboratory analysis results, Bovine manure has 33 and 47% more N than Poultry manure and *J. curcas* seedcake, respectively. Conversely, this pair of fertilizers have a large amount of K compared to Bovine manure and urea. Thus, K could have affected RLS. Indeed, K-rich fertilizers can reduce bacterial blight incidence (*Xanthomonas citri* pv. *malvacearum*) in cotton, decreasing its effect when the plants reach the optimum growth level (Huber and Graham 1999).

Boll rot incidence was twice as low in cotton plants fertilized with doses between 100 and 200 kg N ha<sup>-1</sup>, compared to the initial dose of 50 kg N ha<sup>-1</sup>. In cotton, N above 200 kg ha<sup>-1</sup> can increase root growth, especially in top soil layers, and physiological and biochemical processes in leaves (Chen et al. 2019). Meanwhile, in other crops, such as rapeseed (*Brassica napus* L.), an increase in N availability can produce acetic acid emission (a volatile antifungal), which could reduce the levels of Dark spot disease (*Alternaria brassicae*) (Veromann et al. 2013). Thus, perhaps some compound or substance produced by fertilizers could be absorbed by the plant and used to reduce the pathogen infection that causes BR.

None of the diseases was significantly correlated, not even between incidence and severity of RLS, which means that this disease responded differently to fertilizers and doses. Two important points need to be discussed here: the particularity of each pathogen and the tissue it affects, and the nitrogen source and dose used. In this regard, the susceptibility of tomato plants to Fusarium wilt (*Fusarium oxysporum* f. sp. *lycopersici*), Bacterial speck (*Pseudomonas syringae* pv. *tomato*), and Powdery mildew (*Oidium lycopersicum*) are dependent on the supply of N (Hoffland et al. 2000). Also, both the amount of nitrogen added (direct effect) and the competition of pathogens (indirect effect) play an important role in the plant-pathogen interaction (Liu et al. 2017). This supports the hypothesis that the response to the disease would depend on the nutritional source, each pathogen, or both.

Although the Logistic model is the most used to describe epidemic progress, the Gompertz model is also among the most used in this type of research (Berger 1981; Bergamin Filho 2018; Tjørve and Tjørve 2017). In this study, the Logistic model fitted better than Gompertz to RLS, but no model was adjusted to

BR. For the epidemiological components using the Logistic model, the smallest  $Y_0$  was observed for the incidence and severity of RLS in plants fertilized with *J. curcas* seedcake and poultry manure, when compared to the other fertilizers. In general, this result coincided with the progress curves. However, the  $r$  for RLS incidence was lower only in plants fertilized with urea, compared to the other N sources. Although the increase or reduction of disease due to N fertilization ( $\text{NH}_4$ ,  $\text{NO}_3$ , or another source), forms of N can act biochemically, physiologically, and molecularly in different ways in plants (Sun et al. 2020). Therefore, maybe that each nitrogen source is affecting some physiological process in cotton plants, resulting in that different effect on the progress of RLS.

Doses of 150 and 200 kg N ha<sup>-1</sup> negatively affected the  $Y_0$  and positively the  $r$  of RLS in cotton plants, respectively, compared to the other doses. This result coincided with the progress curves, where plants fertilized with greater doses reached a lower final proportion of the disease. It is known that increasing N can negatively affect the area under the disease progress curve (AUDPC) and severity index of *Alternaria* leaf blight (*Alternaria dauci*) and *Cercospora* leaf spot (*Cercospora carotae*) on carrot plants (Saude et al. 2014). Perhaps these differences are due to the behavior of each disease and crop. Nonetheless, the N mechanism of action reducing disease pressure is not yet fully understood. One hypothesis would be that N's additional application promotes the growth of new leaves, which would temporarily reduce the severity indices of RLS in cotton plants (Saude et al. 2014).

In this study, we analyzed no biological components. However, the organic fertilization with *J. curcas* seedcake and poultry manure in cotton could be a long-term sustainable practice by increasing the abundance of beneficial soil microorganisms, and could be an effective method to inhibit soil-borne diseases (Lin et al. 2019; Tao et al. 2020). Organic fertilizers can increase the size of the rhizosphere population, i.e., antagonists and pathogens and functional groups of rhizosphere fungi and actinomycetes, protecting cotton plants from soil-borne pathogens (Huang et al. 2006). Although more studies are needed to investigate disease responses to nitrogen fertilization, studies similar to the present one demonstrate that this option is viable. Perhaps in cotton, the inorganic fertilizer could be partially replaced by an organic source.

## 5. Conclusions

An interaction between nitrogen sources and doses was observed in RLS incidence. The incidence of BR was two times lower in cotton plants fertilized with doses between 100 and 200 kg N ha<sup>-1</sup>, than between 50 kg N ha<sup>-1</sup>. Each of the diseases responded differently to fertilizers and doses. The logistic model fitted better than Gompertz to RLS, but no model fitted to BR. The less amount of initial disease ( $Y_0$ ) was found for the incidence and severity of RLS in plants fertilized with *Jatropha curcas* seedcake and poultry manure, but a lower progress rate ( $r$ ) for the incidence of RLS in plants fertilized with urea, compared to the other sources of N. The doses 150 and 200 kg N ha<sup>-1</sup> negatively affected RLS  $Y_0$  and positively  $r$  in cotton plants, respectively, compared to others doses.

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