Triple superphosphate, potassium sulfate, and nitrogenous fertilizers effects on fitness and aggressiveness of *Fusarium culmorum* inducing wheat crown rot

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Abstract

This study examined the effects of N, P, and K fertilizers on *Fusarium culmorum* on *in-vitro* mycelial growth and biomass, and aggressiveness on wheat plants. Urea, phosphorus, potassium sulfate, and the mixture of similar amounts of these last two ingredients increased fungal biomass. Rearing mycelia on urea, the mixture, or phosphorus increased aggressiveness by 56%, 120%, and 130%, respectively, without affecting inoculated plants dry biomass. However, the inoculum that was reared on ammonium sulfate increased infected plant biomass. These findings infer that the management of this disease may rely on a proper type of fertilizers application. First, farmers are advised to avoid urea and use ammonium nitrate instead; second, place phosphorus and potassium sulfate under the seedbed away from any inoculum in the soil. Therefore, surveying disease development, its past events, soil health and soil fertility is a prerequisite for any successful control of this disease.

Keywords: *Fusarium culmorum*, Aggressiveness, Fitness, Nitrogen forms, Phosphorus, Potassium sulfate

INTRODUCTION

Worldwide, *Fusarium culmorum* W.G. Smith Sacc. induces wheat crown rot (WCR) disease in arid and semi-arid regions (Burgess *et al*., 2001; Pettitt *et al*., 2003). This pathogen dominates the etiology of this disease, especially within spring cereal crops (Wallace, 1978; Smiley *et al*., 2016; Voigt, 2002), while in other areas it is *F. Pseudograminearum* that is the causative agent of crown rot (Alahmad *et al*., 2018; Kazan and Gardiner, 2018). Gargouri *et al*. (2003) gave evidence that the genetic variability of this pathogen has no geographic origin. However, this variability only occurs within the pathogen's population and stems from large spore dispersal (Mishra *et al*., 2003).

Weather and crop management tools such as crop type, tillage, rotation, soil fertility, weather conditions and their interactions highly affected pathogen infection and WCR disease development (Colhoun *et al*., 1968; Scherm *et al*., 2013; Smiley, 2016). Humidity and temperature affected mycelia biomass and pathogenicity consequently contributing to the development of this disease (Cook and Christen, 1976; Brennan *et al*., 2003).

In water-stressed environments, infected cereal plants develop severe disease symptoms in comparison to those grown under irrigated conditions (Smiley *et al*., 1996; Chekali *et al*., 2011). Also, any cause that impels a decreasing state in soil water potential predisposes small grain cereals to this disease by increasing *F. culmorum* colonization of wheat seedlings (Papendick and Cook, 1974; Beddis and Burgess, 1992).

For disease control, scientists mostly look for a decrease of soil inoculum, through crop residue management and rational use of nitrogenous fertilizers (Cook, 1974; Chakraborty *et al*., 2006). Swan *et al*. (2000) promoted residue decomposition practices that mitigate disease incidence in the following crop through lowering *F. culmorum* survival (Warren and Kommedahl, 1973). In this later work, nitrogen application, in the form of ammonium nitrate, speeded wheat residues decomposition and thus reduced populations of *Fusarium* spp. (Warren and Kommedahl, 1973; Pereyra *et al*., 2004). Also, applying *Trichoderma harzianum* and nitrogen to infected plant residues displaced the pathogen from its niche, but this displacement depended on the form of nitrogen applied (Lakhesar *et al*., 2010). However, the interaction of the host and the pathogen in disease development is modulated by environmental factors and crop management tools (Voigt, 2002).

Most studies that dealt with nitrogen effect on disease development pointed out that overuse of nitrogenous fertilizers intensified the disease (Papendick and Cook, 1974; Rowaished, 1981; Kommedahl, 1984). Furthermore, the form of nitrogen has a greater effect on host resistance or disease severity than does its amount. With no specification of nitrogen form used, Hemissi *et al*. (2018) pointed out that nitrogen applied at 150 to 200 kg ha^{-1} increased disease severity and whiteheads development. In contrast, urea and ammonium sulfate increased WCR severity, when they were applied as a baseline, or as top-dressing, but ammonium nitrate application at pre-sowing or at tillering and/or at stem elongation, significantly reduced the disease (Baha Eddine *et al*., 2019).

To our knowledge, limited studies have dealt with pathogen fitness and aggressiveness under nitrogen forms and/or other macronutrients action (Garrett, 1976; Huber, 1990). In any pathosystem (Huber and Haneklaus, 2007), nutrients play a greater role in altering the pathogen's

virulence (Thomas and Elkinton, 2004). Therefore, the nutrients' effects on the pathogen biology and the host, through temporal interaction among disease components are a prerequisite to the control of this disease (Agrios, 2005; Chakraborty *et al*., 2006). The general use of fertilizers (N, P, and K) in cereal production (FAO, 2006) gave a reason for this study to test them as treatment factors. Therefore, we examined the effect of nitrogenous, phosphorus and potassium fertilizers on *F. culmorum* fitness and aggressiveness.

MATERIALS AND METHODS

Fertilizers' effect on *in-vitro* mycelium growth and biomass

This experiment used phosphorus as triple superphosphate (45% P_2O_5), potassium as potassium sulfate (48% K_2O), phosphorus plus potassium sulfate (1/2-1/2 for each), and nitrogen as ammonium sulfate (21% N), ammonium nitrate (33.5% N) and urea (46% N) as fertilizers. From each fertilizer, six quantities were independently added to flasks that contained one liter of distilled water and 10 g of glucose. These quantities were rated 1 as control (unfertilized treatment), and 2: 0.2 g, 3: 0.4 g, 4: 0.8 g, 5: 1.2 g, and 6: 1.6 g as the remaining levels. All the flasks were sterilized for 20 minutes at 120 °C and a pressure of 3 bars. Once cooled, each flask was inoculated with a PDA fragment (approximately 1 cm^2) of a fresh local virulent *F. culmorum* isolate. *Fusarium* isolation and identification followed the procedure described previously (Baha Eddine *et al.,* 2019). The isolate used herein was obtained from infected crown wheat. Inoculated flasks were incubated for one month under 12 h of photoperiod. Afterward, the content of each flask was separately filtered through sterilized cheesecloth and the collected mycelia biomass was independently dried out in a laminar flow cabinet and then weighed.

This experiment was repeated once. The flasks of the first experiment were incubated at 20–25 °C whereas those of the second one were incubated at 15–20 °C. Additionally, for the first trial, a mycelium sample from each treatment (fertilizer by dose interaction) was transferred to two Petri dishes containing PDA medium. This fungal transfer is meant to assess the carryover effect of fertilizers and their rates on *in-vitro* mycelia growth.

Colony growth measurements in each Petri dish were initiated on the 3rd day and resumed on the sixth day of post-inoculation. The evolution of the growth colony (cm), in four perpendicular directions of a plate, was monitored. The colony size was estimated by the following formula:

$$colonie\ size = 1/4 \sum_{I=1}^{4} \pi \times R_i^2$$

Where R_i is the radial length in direction *i*, and the mean of colony size per treatment by dose and by experiment was plotted against post-inoculation dates (Baha Eddine *et al.,* 2020). This monitoring of the growth is intended to assess the fungal fitness (Pringle and Taylor, 2002).

Fertilizers' effect on Aggressiveness

The experiment was carried out in a greenhouse in the 2017–2018 growing season and was laid out in the first week of November and ended in late April. Six plastic trays of 77 cavities (7 × 11) were used and each cavity had a dimension of 64 cm^3 (4 × 4 × 4 cm). Each tray contained one fertilizers' treatment level: phosphorus, potassium sulfate, phosphorus plus potassium sulfate, ammonium sulfate, ammonium nitrate, or urea. Within each plastic tray, six columns that were vertically arranged represented fertilizers' rates, each of which was separated by a column of 7 voids. And within each fertilizer's column, the seven cavities represented repetitions (mycelium samples as batches). All the cavities, except those separating the rate's factor, were filled to half of their capacity with a natural soil (Tirs) brought from the INRA experimental station of Sidi El Aïdi (Settat, Morocco). This soil was previously sieved and sterilized, for one hour, three times in a row at 120 °C and a pressure of 3 bars. Five seeds of a sensitive durum wheat cultivar "Ourgh" were sown in each cavity, and then a thin layer of the same soil was broadcast to cover the seeds. Thereafter, 12 ml of water was poured onto each seeded cavity to initiate germination.

Two days later, the dried mycelia biomass, collected from all liquid media, were separately grounded and homogeneously distributed over the 7 vertical cavities which represented batches. The inoculum was then covered with a thin layer of sterile soil. Irrigation with distilled water was provided according to the plant's need.

Pathogen aggressiveness, defined as the pathogen's ability to invade and establish in the host (Thomas and Elkinton, 2004), was quantified herein by the severity of symptoms induced by the inoculum reared on different growth media. Disease evaluation started at the heading growth stage in line with our previous work (Baha Eddine *et al.,* 2019). Any plant with a disease progression that exceeded the third node had been given a score of 3 as the highest severity score. Subsequently, evaluated plants per treatment were separately left to dry in the open air of a shutdown greenhouse. Then, the dry weight of plant biomass affected by each treatment was weighted. This experiment was immediately carried out after each trial that dealt with mycelia biomass production and repeated once.

Statistical Analysis

All statistical computations were carried out using the SPSS statistical package (version 20) for Windows. A combined analysis, of the two experiments, was used to assess treatment effect (fertilizers and rates) on fungal biomass. The Leven test for homogeneity of variance between the two experiments gave a basis to separate analysis for each experiment. Therefore, fertilizers' effect and their rates, within each experiment, were evaluated by an analysis of variance (ANOVA) using the GLM procedure. Significant differences among fertilizers were tested at 5% probability with the Waller-Duncan test statement, and doses with a polynomial contrast statement (West *et al.,* 2015). To analyze the strength of this

relationship, either linear or logarithmic, a regression analysis was performed. On the other hand, to evaluate the carryover effect, a mixed model with colony size as a dependent variable was adopted with fertilizers and rates as fixed factors and plates as random. The experimental design was a longitudinal model with a repeated statement as post-inoculation days and subjects as plates. We selected a covariance matrix as arima 1 with the covtype (ar1) statement and everything else is kept as default. The experiment about disease severity was analyzed as a nested design. Fertilizers were taken as a fixed factor while doses were nested within fertilizers because they were assumed to have different nitrogen concentrations and then were taken as experimental errors for fertilizers. The batches (samples of inoculum) within doses by fertilizers were experimental error for doses nested within fertilizers. Significant differences between the control and fertilizers' effect were based on Bonferroni test at 5% probability. Finally, the two levels of this later experiment were taken as random (Berger *et al.*, 2018).

RESULTS

The two trials about fungal biomass production had a highly significant different variance (Levene statistic = 7.09; df1 = 1; df2 = 70; P = 0.010). This significant test was the basis for separately analyzing them. Therefore and within both experiments, fertilizers and their rate were found to significantly affect *F. culmorum* biomass ($P \leq 0.001$). Also, the polynomial contrast test gave evidence that the increase of biomass production with an increase in fertilizers' rates had a significant cubic profile ($P \leq 0.001$) in both trials.

The use of ammonium sulfate, ammonium nitrate, urea, or phosphorus as a substrate in growth media, induced a similar increase of fungal biomass, but this increase was different within the two temperature essays (Figures 1a, 1b, 1c, and 1d). In contrast, potassium sulfate, or the mixture of phosphorus, and potassium sulfate mostly produced similar biomass production profiles nearly smaller between the two experiments (Figures 1e and 1f). The significant effect

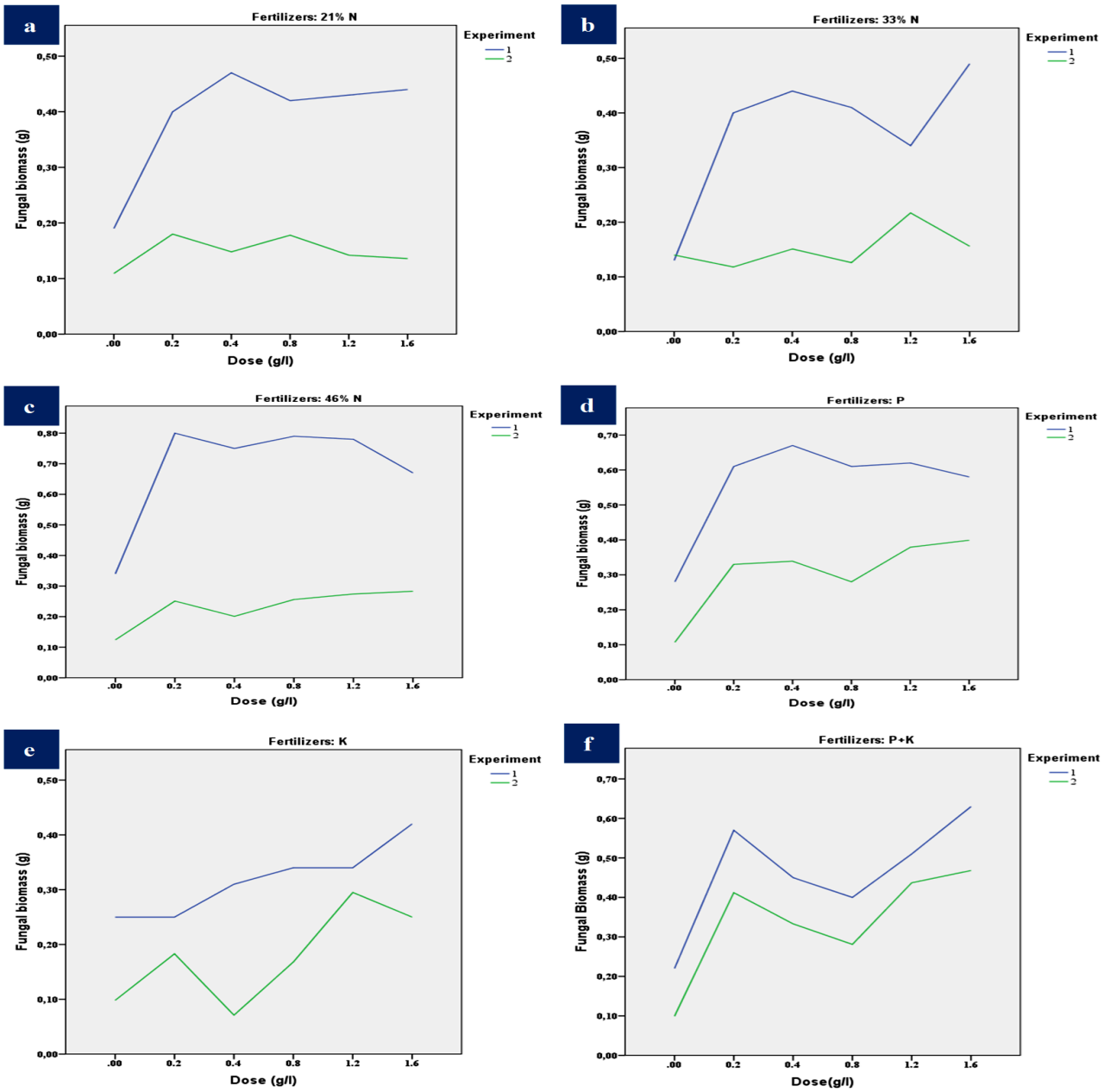

Figure 1: Effect of fertilizer's type and rates added to growth media on Fusarium culmorum fungal biomass production under two experimental conditions (experiment 1 : 20–25 °C; experiment 2: 15–20 °C). Note: 21% N: Ammonium sulfate (a); 33% N: Ammonium nitrate (b); 46% N: Urea (c); P: Phosphorus (d) and K: Potassium sulfate (e); P+K (f)

of the growth media on fungal biomass showed that ammonium sulfate, ammonium nitrate, or potassium sulfate gave the lowest production in both experiments (Table 1). Whereas, those containing urea, phosphorus, or the phosphorus mixture and potassium sulfate significantly stimulated *F. culmorum in-vitro* growth, and therefore the fungal biomass (Table 1).

A significant relationship between fertilizers' rates and *F. culmorum* fungal biomass production is evident in table 2, except for ammonium sulfate and ammonium nitrate at a temperature of 15 to 20 °C (Table 2). Most of the other rates had a logarithmic relation between doses and biomass production. Potassium rates showed a significant linear effect even stronger at 20 to 25 °C when comparing its standardized coefficient beta of 0.95 to 0.76 at 15-20 °C. Also, the use of urea, phosphorus, or the mixture of phosphorus and potassium induced similar logarithmic strength on the production of fungal biomass under both temperature conditions (Table 2).

Overall, the two experimental conditions had a different effect on fungal biomass (Figure 1) and within each trial, the main effect of fertilizers' rate level off at 0.2 g L^{-1}.

The inoculum reared on different sources of fertilizers and rates presented similar growth patterns on PDA medium (Table 3) and the only significant effect (P = 0.001) on *in-vitro* mycelial growth was relative to days of post-inoculation (Table 3). Therefore, the growth media continent did not have any carryover effect on the fitness cost of this pathogen.

Table 1: Mean fertilizers' effect on *Fusarium culmorum* fungal biomass (g) production per experiment

Fertilizers	Experiment 1 (20–25 °C)	Experiment 2 (15–20 °C)
Control (0% N)	0.24	0.11
21% N	0.43*	0.16
33% N	0.42*	0.15
46% N	0.76*	0.25*
P	0.62*	0.35*
K	0.33*	0.19*
P+K	0.51*	0.39*

Note: 21% N: Ammonium sulfate; 33% N: Ammonitrate; 46% N: Urea; P: Phosphorus and K: Potassium sulfate. Values of the same column followed by a star are significantly different from the control (0% N) at 5% probability (simple contrast).

Table 2: Linearization of the fertilizers' effect on *Fusarium culmorum* fungal biomass production under two temperature conditions

Fertilizer	Temperature (°C)	Dependent	Intercept	Slope	S. C. Beta	R^2	p. value
21% N	15-20	N	-	-	-	-	-
	20-25	Ln (Y)	-0.80 (0.049)	0.118 (0.017)	0.96	0.93	0.002
33% N	15-20	N	-	-	-	-	-
	20-25	Ln (Y)	-0.82 (0.096)	0.182 (0.036)	0.93	0.87	0.007
46% N	15-20	Ln (Y)	-1.33 (0.052)	0.119 (0.020)	0.95	0.90	0.004
	20-25	Ln (Y)	-0.25 (0.078)	0.121 (0.029)	0.90	0.81	0.015
P	15-20	Ln (Y)	-1.00 (0.077)	0.189 (0.029)	0.96	0.91	0.003
	20-25	Ln (Y)	-0.446 (0.067)	0.121 (0.025)	0.92	0.85	0.009
K	15-20	Y	0.101 (0.039)	0.109 (0.043)	0.79	0.62	0.060
	20-25	Y	0.249 (0.014)	0.099 (0.016)	0.95	0.91	0.003
P+K	15-20	Ln (Y)	-0.89 (0.117)	0.216 (0.044)	0.93	0.86	0.008
	20-25	Ln (Y)	-0.638 (0.098)	0.132 (0.037)	0.88	0.77	0.023

Note: 21% N: Ammonium sulfate; 33% N: Ammonitrate; 46% N: Urea; P: Phosphorus and K: Potassium sulfate. N and - refer to no relationship; Ln (Y) is a natural logarithm relation of the dependent variable (biomass) and log dose; S. C. Beta is a standardized coefficient beta.

Table 3: Variance analysis of fertilizers' carryover effect and their rates on *Fusarium culmorum* growth on PDA medium

Source	Numerator df	Denominator df	F	Probability
Evaluation	2	44.01	2019.24	0.001
Fertilizers	5	22.35	0.78	0.575
Rate	5	22.35	1.08	0.401
Rate * Evaluation	10	44.01	1.03	0.435
Fertilizers * Evaluation	10	44.01	1.635	0.128

Note: df= degree of freedom; Fertilizers and Doses represent factors of different inoculum sources that were reared in the growth media.

Fertilizers significantly affected disease severity ($P \leq 0.001$) and therefore the aggressiveness of the pathogen (Table 4). Compared with the unfertilized check, the overall use of the mycelia reared either on urea, the mixture of phosphorus and potassium sulfate, or phosphorus stimulated disease severity by 56%, 120%, and 130%, respectively (Figure 2). In contrast, amended media with ammonium sulfate and ammonium nitrate did not change this characteristic (Figure 2).

All fertilizers rates within fertilizers and batches significantly affected the dry weight of inoculated plants at $P \leq 0.001$, $P \leq 0.001$, and $P \leq 0.048$, respectively (Table 5). The

Table 4: Variance analysis of the effect of fertilizers and their rates on disease severity induced by *Fusarium culmorum* inoculum that was reared on fertilizers

Source	Type III Sum of Squares	df	Mean Square	F	Probability
Experiment	66.5	1	66.5	107.6	0.001
Fertilizers	75.6	6	12.6	26.4	0.001
Rates (Fertilizers)	11.5	24	0.48	0.91	0.584
Batch (Rates (Fertilizers))	97.2	186	0.52	0.85	0.892
Error	173.1	28	0.62		

Note: df= degree of freedom; Note that fertilizers' factor and doses within Fertilizers represent the inoculum that was reared on media containing the used fertilizers, but none of the fertilizers was added to the inoculated plants.

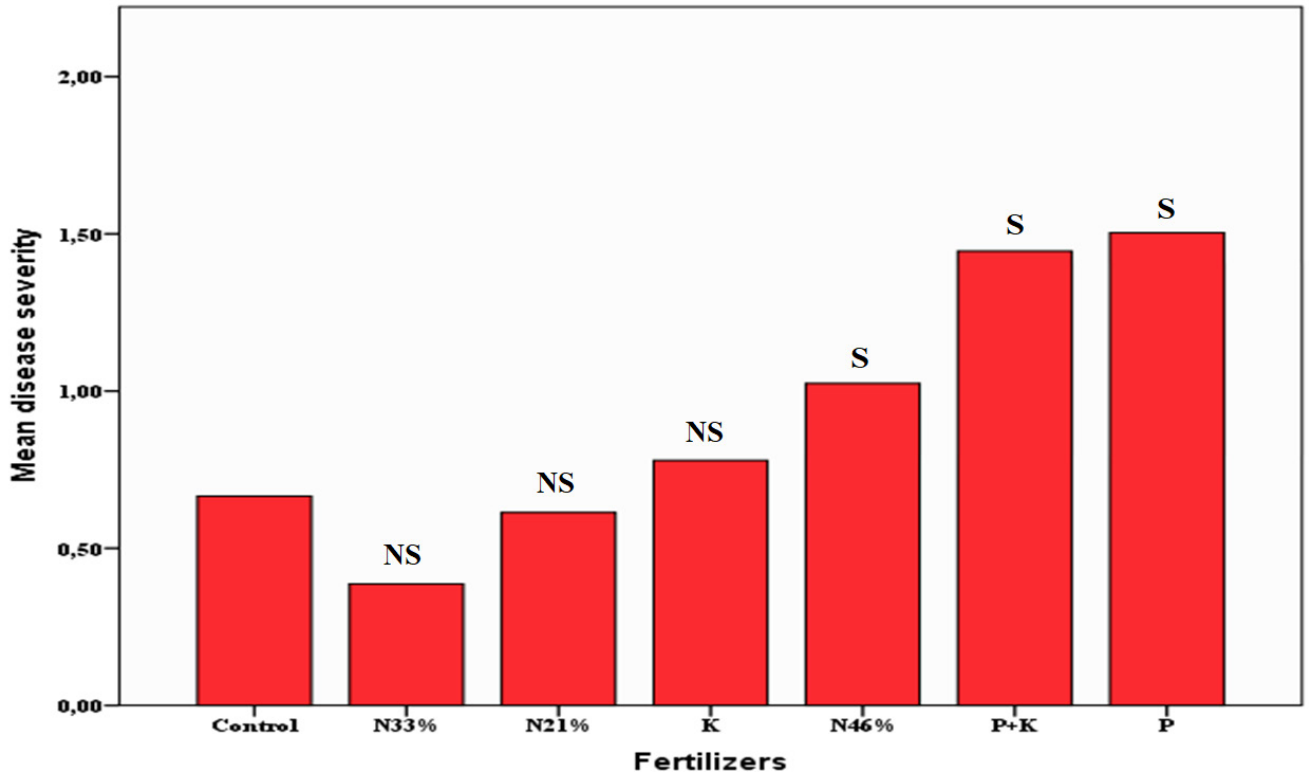

Figure 2: Simple mean fertilizers' effect on inoculum aggressiveness assessed as wheat crown rot disease severity. Note: S= significantly different from the control based on a Bonferroni test at 5% probability. NS=non significant; The simple mean effects represent the inoculum that was reared on fertilizers, and none of the fertilizers was added to the inoculated plants

Table 5: Variance analysis of the effect of the inoculum that was reared on fertilizers and their rates on biomass dry weight of inoculated plants (g)

Source	Type III Sum of Squares	df	Mean Square	F	Probability
Experiment	62.4	1	62.4	676.7	0.001
Fertilizers	5.29	6	0.88	6.73	0.001
Doses (Fertilizers)	3.15	24	0.13	1.58	0.048
Batch (Doses(Fertilizers))	15.4	186	0.08	0.90	0.786
Error	25.8	280	0.09		

Note: df= degree of freedom; Fertilizers' factor and doses within Fertilizers represent the inoculum that was reared on media containing the used fertilizers, but none of the fertilizers was added to the inoculated plants

greatest inoculated plant dry weight was induced by the inoculum cultured on ammonium sulfate media (Figure 3). The significant effect of rates within fertilizers on infected plant biomass, depicted in Figure 4, revealed an interaction between fertilizers and their rates. This interaction was mostly fueled by differences between the effects of ammonium nitrate and ammonium sulfate (Figure 4).

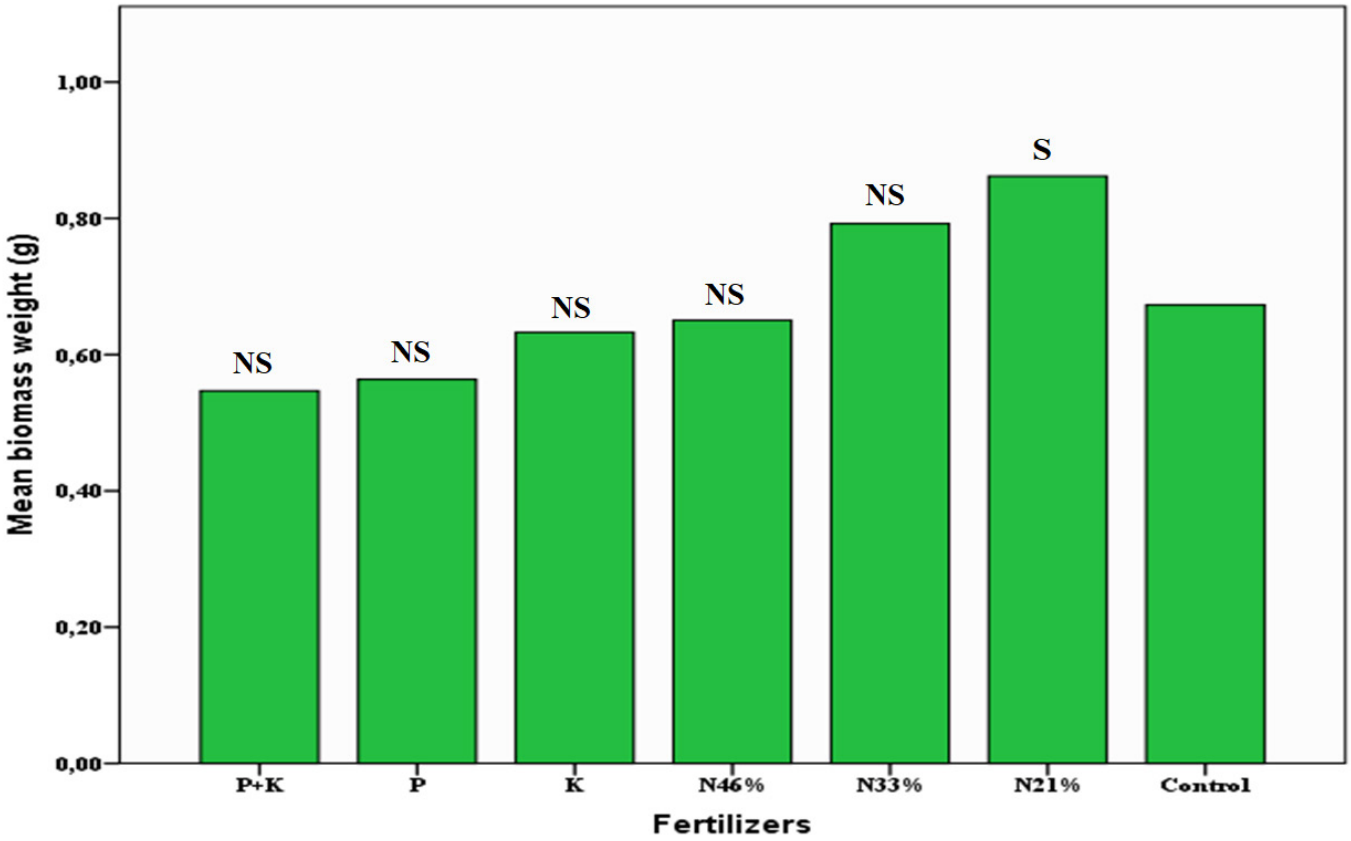

Figure 3: Simple mean fertilizers' effect on biomass dry weight of inoculated plants. Note: S= significantly different from the control based on Bonferroni test at 5% probability. NS=non significant; The simple mean effects represent the inoculum that was reared on fertilizers, and none of the fertilizers was added to the inoculated plants.

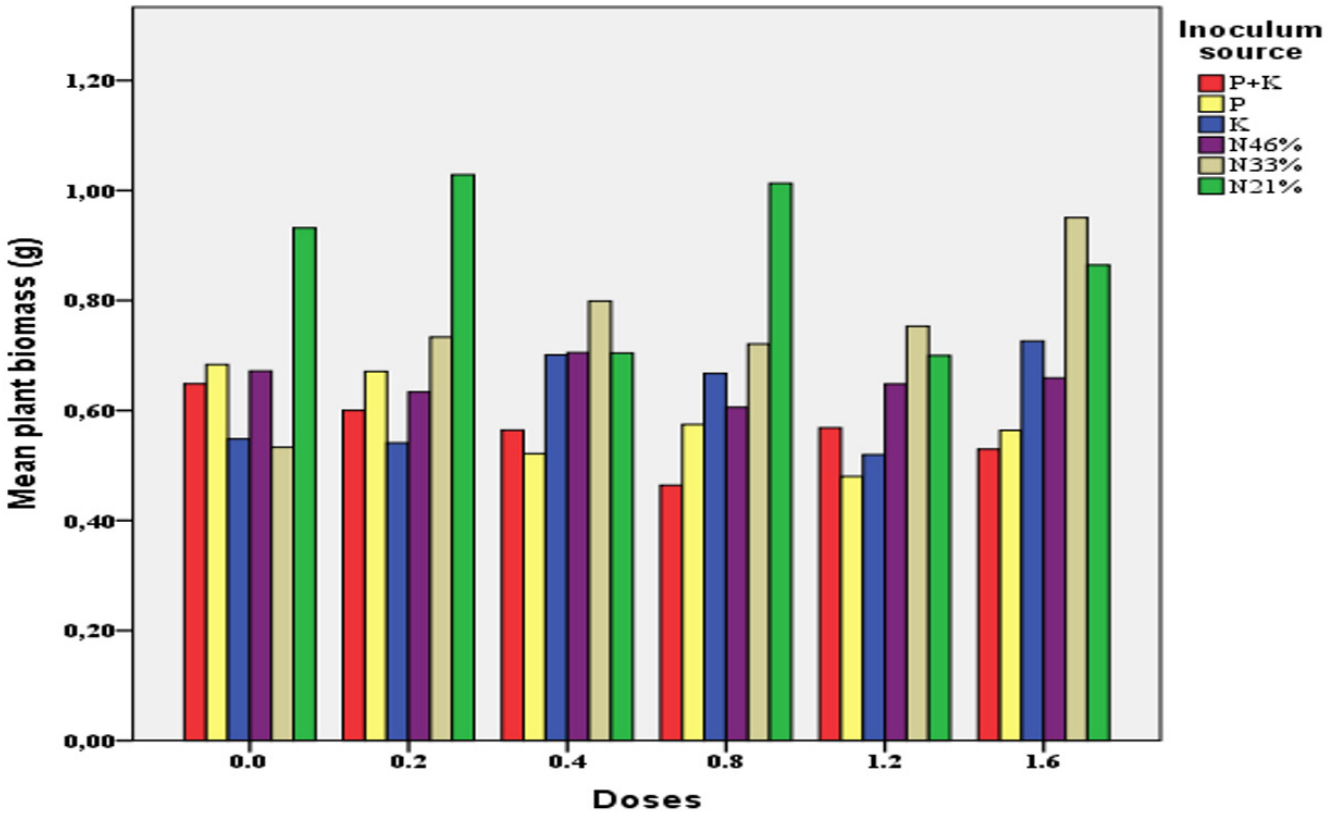

Figure 4: Differential effect of inoculum sources (fungus reared on media containing) fertilizers) and rates within fertilizers on dry weight biomass of inoculated plants. The inoculum source represent the fungus that was reared on the fertilizers used, and none of the fertilizers was added to the inoculated plants.

DISCUSSION

Levels of yield losses caused by WCR disease urge scientists and farmers to seek sustainable control measures based on the knowledge of disease biology and epidemiology (Duveiller *et al.,* 2007; Paulitz *et al.,* 2010; Chekali *et al.,* 2013). Thus, we studied management of this disease with major mineral nutrients (nitrogen, phosphorus, and potassium), applied at different rates, on the fitness features (growth, biomass production, and aggressiveness) of *F. culmorum*.

This work was set up as a continuation of a preliminary study that dealt with *F. culmorum* growth on solid PDA (Damir, 2014). The solid media were independently amended with 0.1, 0.2, 0.4, and 0.8 g L^{-1} of phosphoric, potash, and nitrogenous fertilizers. Though, media containing phosphorus at 0.4 g L^{-1}, 0.6 g L^{-1}, and 0.8 g L^{-1} remained liquid, although we increased the amount of agar. These findings justified our use of liquid media instead.

Monitoring *F. culmorum* biomass production, within the two-temperature conditions, showed that the mycelia production was dependent on the setup of the two experiments. The fungal biomass obtained at 20-25 °C was greater than the one obtained at 15-20 °C by 104% (Table 1). This finding is supported by the optimum growth at 25 °C of the European *Fusarium culmorum* isolates on PDA, irrespective of their geographic origin, and these later isolates were the fastest-growing of the fusaria tested (Brennan *et al.,* 2003). Under these two temperature conditions, fertilizers affected the fitness and aggressiveness of *F. culmorum*. Phosphorus and the mixture (phosphorus and potassium sulfate) improved these two features, and explain in part Greaney's (1938) observation at Winnipeg who noticed an increase in disease severity when soil phosphorus was in excess or a deficient state. Damir (2014) also pointed out that phosphorus added at post-emergence to inoculated plants, grown in pots filled with natural soil, enhanced disease development. Further, she found that the application of phosphorus and potassium sulfate as a solution at post-emergence increased disease severity on the same variety herein used. These reported studies infer that close contact of this pathogen with phosphorus acts directly upon the fitness components, and therefore impacted disease development.

Even though the application of phosphorus and its addition to potassium sulfate increased the fitness, aggressiveness, and disease development, they did not affect the infected plant biomass (Table 1, 4, and 5, Figures 2 and 3). Baha Eddine *et al.* (2019) showed that WCR disease development has a close association with nitrogen fertilizers. Thus, whenever infected plants were fertilizers with either form of ammonium sulfate or urea, the disease intensity as the incidence and severity took severe proportions. In this study, we did not add any fertilizer to the natural soil, and levels of disease severity recorded may infer that the disease stems from a nitrogen pool (mycotoxins promoters) of the pathogen. Therefore, adding nitrogen shifted this pathogen from a non-dominant to a host-dominant disease in this pathosystem (Kommedahl and Windels, 1979).

Although potassium sulfate had shown some effect on fitness and aggressiveness it did not induce any significant disease severity (Table 1 and Figure 2). Hofgaard *et al.* (2010) also noticed a reduction *in-vitro* growth of *F. culmorum* and *F. graminearum* when the fertilizer containing potassium phosphate was added into the growth medium. Even a decrease in head blight was observed when this fertilizer was applied as a foliar application on wheat plants. Also, Onuorah (1969) gave evidence that an application of potassium sulfate decreased WCR severity on adult plants. Moreover, high potassium application, especially if associated with high nitrogen, reduced the disease even further (Dixon *et al.,* 2010).

Application of a $CaNO_3$ (1.21 g L^{-1}) solution on infected plants increased WCR disease development (Onuorah, 1969). Papendick and Cook (1974) further demonstrated that application of nitrogen when coupled to water stress enhanced disease incidence and severity. Rowaished (1981) used a series of Hoagland solutions containing concentrations and forms (NO_3 and NH_4), ranging from 21 ppm up to 1050 ppm and found a slight increase in disease severity with high concentration levels of nitrogen.

At the two temperatures, nitrogen, like urea, affected fungal biomass, aggressiveness, and overstimulated this biomass by one to two folds (Table 1). This significant increase induced levels of aggressiveness that were less than those of phosphorus and the mixture but did not affect plant dry biomass. In recent work, urea was also found to optimally increase pathogen fitness when it was added to solid media at a rate of 1.5 g L^{-1} (Baha Eddine *et al.,* 2020), but concentrations over 12 g L^{-1} of urea were harmful to the pathogen. However, at a fixed nitrogen concentration of 1.57 % N among nitrogenous fertilizers, this nutrient favored mycelium growth and aggressiveness by 50%. Additionally, Damir (2014) also showed that an application of urea either at 0.1 or at 0.2 g L^{-1} favored fungal growth. This nutrient was also found to increase disease development when placed either under seedbed or as a foliar application (Baha Eddine *et al.,* 2019), or as a top-dressing fertilizer (Akgül and Erkilic, 2016; Baha Eddine *et al.,* 2019).

Sulfur is known to be a good fungicide against various plant diseases (Haneklaus, 2007). But surprisingly in our study, ammonium sulfate was found to only affect growth fitness at 25°C, and on the contrary, increased dry weight of infected plants through a non-significant effect on disease severity. In another work, fertilizing infected plants, independently as top-dressing at tillering or at stem elongation, increased disease severity by 33% and 30%, respectively (Baha Eddine *et al.,* 2019), but nothing was reported on yield. Yet, a simultaneous application of this fertilizer at these two growth stages, increased disease severity by 53% (Baha Eddine *et al.,* 2019). Ammonium sulfate with 23 to 24% of sulfate also has

a fungistatic action on wheat powdery mildew (Cook, 1987). Also, Williams and Cooper (2004) gave evidence that some fusaria species were sensitive to this nutrient. This may explain in our work the noted weak fitness and aggressiveness of this pathogen, but its stimulation of the infected plant biomass needs more research.

The mitigated ammonium nitrate's effect on fitness (Table 1) and aggressiveness (Figure 2) agreed with the findings of Baha Eddine *et al.* (2020) which stated that the use of this fertilizer at 24 g L^{-1} lessened these two characteristics, and resulted in a decrease of disease severity with a reliable expression of disease resistance. Thus, these results explain why levels of this disease were low when this fertilizer was applied at pre-sowing or as a top dressing at tillering and/or at stem elongation (Baha Eddine *et al*., 2019).

The macronutrient impact on plant diseases depends on host response, previous crop sequence, rate of nitrogen applied, residual N, the period of application, soil microorganisms, the ratio of NH_4^+ to NO_3^- and disease etiology (Kommedahl, 1984). However, depending on the pathosystem, other companion ions such as Ca, K, Cl, and SO_4 may also be involved in disease development (Dordas, 2008; Lawrence and Wade, 2018, Yadav *et al.,* 2020). Thus, every pathosystem must be investigated for the right nutrient forms to use for any disease management (Fageria *et al.,* 2010). Evaluating the effect of N, P and K nutrients on this fungus may shed light on its genetic diversity characterized by phenotypic traits such as pathogenicity, aggressiveness, and mycotoxin production (Korn *et al.,* 2011).

CONCLUSION

This study added new evidence to the causal relation between nitrogen form and disease development. Therefore, it provided insight into WCR disease by assessing the interaction between the causal agent and N, P and K macronutrients. The use of urea and phosphorus alone or with potassium sulfate increased *F. culmorum* fitness and aggressiveness. In contrast, ammonium nitrate, as a nutrient, significantly lessened these two characteristics. The study also allowed us to assume that these findings may pinpoint some disease management and therefore, fulfill the control of this disease throughout soil health quality. For example, when *F. culmorum* propagules occur in soil profiles, we can speculate that the application of urea for top-dressing cereal in arid and semi-arid regions should not be recommended, whereas the use of ammonium nitrate can be a good alternative. Also, we advise cereal farmers to incorporate, into their field, phosphorus and potassium under seedbed or drilled rowed to avoid any contact of these fertilizers with *F. culmorum* and decline any furrow application. However, the use of ammonium sulfate as a baseline fertilizer is recommended to take advantage of this fertilizer's fungistatic action. But preliminary tests for soil fertility and quantification of the inoculum in cereal fields at pre-sowing frame valuable information for crop management in regions prone to WCR disease. Finally, scientists involved in screening and quantifying disease resistance may consider these findings when carrying out their selection methods. Other scientists may focus on the nutrition effect on the molecular and epigenetic status of *F. culmorum*. For functional annotation and confidence in our finding, confirmation of our results under field conditions is deemed necessary.

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