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Mycotoxins and Crop Yield in Maize as Affected by Irrigation Management and Tillage Practices

Marta Herrera ¹, José Cavero ², Samuel Franco-Luesma ², Jorge Álvaro-Fuentes ², Agustín Ariño ^{1,*}
and Susana Lorán ¹

¹ Facultad de Veterinaria, Instituto Agroalimentario de Aragón-IA2 (Universidad de Zaragoza-CITA), 50013 Zaragoza, Spain

² Soil and Water Department, Estación Experimental de Aula Dei (EEAD), Spanish National Research Council (CSIC), 1005 Montañana Ave., 50059 Zaragoza, Spain

* Correspondence: aarino@unizar.es; Tel.: +34-876-554142

Abstract: In addition to the weather conditions, agronomic practices can have a major influence on maize crop yield and contamination with mycotoxins. In this work, the effect of different irrigation systems (flood vs. sprinkler irrigation), sprinkler irrigation management (low vs. high frequency, daytime vs. nighttime irrigation) and tillage practices (conventional tillage, no tillage with or without crop stover) on crop yield and the contamination with aflatoxins (AFs), fumonisins (FUM) and deoxynivalenol (DON) were evaluated in the maize grain from two experimental maize fields. No aflatoxins were detected in any of the samples analyzed. DON and FUM levels were significantly higher when the sprinkler irrigation was performed at nighttime (0.54 and 1.21 mg kg⁻¹, respectively) as compared to daytime (0.38 and 0.45 mg kg⁻¹). Likewise, DON and FUM were greater when irrigation frequency was low (0.61 and 1.09 mg kg⁻¹, respectively) in comparison with high frequency (0.30 and 0.57 mg kg⁻¹). DON concentrations were significantly higher in fields with sprinkler irrigation (0.53 mg kg⁻¹) as compared to flood irrigation (0.19 mg kg⁻¹), while the levels of FUM were very similar regardless of the maize irrigation system. Mycotoxin concentrations were not affected by the different soil tillage practices. This highlights the importance of implementing crop management practices to minimize the risk of mycotoxin contamination in maize.

Keywords: fumonisins; deoxynivalenol; maize; sprinkler irrigation; flood irrigation; tillage methods



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1. Introduction

Maize (*Zea mays* L.) is an important crop worldwide, that is frequently colonized with pathogenic and mycotoxigenic fungal species. In this regard, *Fusarium* spp. are of great concern. These molds are ubiquitous in nature so that *Fusarium* ear rot is one of the most problematic diseases in maize fields. The infection with *Fusarium* spp. not only produces maize yield and quality losses but also may produce mycotoxins. Due to the range of toxic syndromes associated with exposure to these contaminants, they may pose a significant risk to human and animal health when entering the food and feed chain [1].

Fumonisin are mycotoxins mainly produced by *Fusarium verticillioides* and *F. proliferatum*, and fumonisins B1 (FB1) and B2 (FB2) are the most significant in terms of toxicity and occurrence [2]. FB1 has been categorized by the International Agency for Research on Cancer (IARC) as possibly carcinogenic (Group 2B) [3]. A high prevalence of fumonisins has been found in maize and maize-based food in many parts of the world [4–6] including Spain [7–9], and co-occurrence of fumonisins with other *Fusarium* toxins, such as deoxynivalenol, is regularly observed.

Deoxynivalenol (DON) is primarily produced by *Fusarium graminearum* and *F. culmorum*. Although it is not classified as being carcinogenic to humans (Group 3) [10], DON can induce acute gastrointestinal symptoms such as vomiting, diarrhea and food refusal in

both humans and animals [11], so this mycotoxin is also known as vomitoxin. A high prevalence of DON has been found in European maize crops with more than 30% positive samples [6,9,12].

Moreover, aflatoxins (AFs) are a group of closely related mycotoxins (B1, B2, G1 and G2) that may also contaminate maize grains. They are produced by *Aspergillus* species, particularly *A. flavus* and *A. parasiticus*. Aflatoxins have been classified as carcinogenic to humans in Group 1 of IARC [13]. Maize is an important route of exposure to aflatoxins due to its high prevalence: 65% of samples in Sub-Saharan Africa [14] and 94% and 90% in maize crops from Serbia in 2012 and 2015, respectively [6]. However, both prevalence and amounts are highly variable depending on factors such as geographical origin or climate and storage conditions.

Mycotoxin prevention is achieved by controlling producing molds, which requires the application of pre- and post-harvest strategies [15]. In this regard, it is important to consider the predisposing conditions leading to fungal growth and mycotoxin production, such as weather conditions, soil fertility, drought and insect damage or unseasonal rains during harvest [16]. Apart from conducive environmental conditions, maize contamination with mycotoxins is influenced by agronomic management practices [17,18]. In a recent study on the impact of agronomic practices on *Fusarium* mycotoxin accumulation in maize grain, the main driver for mycotoxin accumulation was stress induced by plant competition rather than environmental conditions [19]. Selecting varieties adapted to the growing area and resistant to fungal diseases can help to reduce mycotoxin production in the field [20]. Cropping practices, management of crop residues, chemical control or biocontrol of plant diseases, irrigation, and harvest date are some of the agricultural practices that have an impact on the prevention and control of mycotoxin contamination of crops [21]. Particularly, water stress has been identified as a very important agronomic factor influencing *Aspergillus* and *Fusarium* growth and mycotoxin production. While drought might result in an increased risk of aflatoxin and fumonisin contamination of maize [22,23], irrigation has proven to have different effects on mycotoxin contamination [24,25].

The predictions of climate change indicate an increased risk of mycotoxins in European maize due to periods of drought and high temperatures. Unlike other European countries, maize in Spain is usually grown under irrigation due to the semiarid Mediterranean climatic conditions. Two irrigation systems are currently used to irrigate maize: flood and sprinkler irrigation. There are main differences between them. First, flood irrigation applies high irrigation rates (>100 mm) at low frequency (every 10–14 days), while sprinkler irrigation usually applies low irrigation rates (<30 mm) and at high frequency (1 to 4 day interval). Second, sprinkler irrigation wets the maize canopy at each irrigation event, while flood irrigation does not wet the maize canopy. These differences between the two irrigation systems could affect the occurrence of mycotoxins in maize grain. Moreover, different soil tillage practices also affect the soil water content and the presence of crop stovers in the soil, which could affect the occurrence of mycotoxins in maize grain. Some studies have found that the management of sprinkler irrigation affects the microclimatic conditions (temperature and relative humidity) of the maize crop [26,27], which could result in different occurrences of mycotoxins in maize grain. Accordingly, the objective of this work was to study the effect of different irrigation systems and tillage systems, and different sprinkler irrigation management on crop yield and the presence of fumonisins, deoxynivalenol and aflatoxins in maize.

2. Materials and Methods

Two field experiments were conducted during four maize seasons (2015, 2016, 2017 and 2018) to evaluate the following: (1) field #1 for the effect of irrigation time and irrigation frequency under sprinkler irrigation, and (2) field #2 for the effect of irrigation system (sprinkler vs. flood) and soil tillage.

2.1. Sampling and Experimental Conditions

Conventional maize (*Zea mays* L.) Pioneer P1758 was grown in two experimental fields located in Zaragoza, Spain. The coordinates of field #1 site were 41°42' N, 0°49' W, 225 m altitude, and those for field #2 site were 41°43' N, 0°48' W, 225 m altitude. Maize was planted in April and harvested in September/October each year. All the samples collected from the same field were subjected to equal weather conditions. The weather data of the experimental fields is listed in Supplementary Table S1. This area is characterized by a Mediterranean semiarid climate with an annual mean air temperature of 14.1 °C, annual precipitation of 298 mm and an annual mean reference crop evapotranspiration of 1243 mm. As a reference, the climatological year in Spain in 2015 was extremely hot (average air temperature 16 °C) and very dry (500 mm rain), during 2016 it was very hot (15.8 °C) and humid (682 mm), extremely hot (16.2 °C) and very dry (474 mm) in 2017, and it was hot (15.5 °C) and very humid (808 mm) during 2018 (Climatological Summaries from the State Meteorological Agency (AEMET) available at: <https://www.aemet.es/en/portada> (accessed on 3 March 2023)).

The following parameters related to agronomical practices were evaluated in a factorial design to explore the influence of the independent variables (factors) and their interactions on the dependent variables (mycotoxin levels and grain yield). The first experiment studied the effect of the variables of irrigation time (daytime or nighttime) and irrigation frequency (low and high frequency) when irrigating with a solid-set sprinkler system (Table 1). The starting time was 10:00 h Greenwich Mean Time (GMT) for daytime irrigations and 22:00 h GMT for nighttime irrigations. The experimental fields were divided into irrigation sectors (324 m² in size) that were irrigated independently. Each of the four treatments (F1 to F4) was replicated three times, so there were 12 irrigation sectors where grain samples were taken in each harvest year. The same amount of irrigation water was applied to all the treatments and was calculated weekly [27] and measured using an electromagnetic flow meter (Promag 50, Endress + Hauser, Reinach, Switzerland). As a reference, the irrigation water supply in field #1 ranged between 580 and 620 mm per season.

Table 1. Variables and treatments studied in field experiment #1.

Irrigation Frequency (IF)	Irrigation Time (IT)	
	Daytime: starting at 10:00 h Greenwich Mean Time (GMT)	Nighttime: starting at 22:00 h GMT
Low frequency: two irrigation events per week	F1: daytime & low frequency	F2: nighttime & low frequency
High frequency: daily irrigation	F3: daytime & high frequency	F4: nighttime & high frequency

In the second experiment, the effect of two variables was evaluated: irrigation system (sprinkler vs. flood) and tillage practices (conventional tillage, no tillage without crop stover and no tillage with crop stover) (Table 2). Each of the 6 treatments (S1 to S6) was replicated 3 times, so 18 experimental plots (108 m² in size) were sampled in each harvest year. In this case, sprinkler irrigation events occurred two times per week (Monday and Wednesday), whereas flood irrigation events occurred every 10–14 days. All tillage treatments under the same irrigation system received the same amount of irrigation water. Per season, irrigation water in field #2 ranged between 550 and 700 mm for sprinkler irrigation and from 700 to 870 mm for flood irrigation, respectively.

Table 2. Variables and treatments studied in field experiment #2.

Tillage Practices (TP)	Irrigation System (IS)	
	Sprinkler irrigation	Flood irrigation
Conventional tillage (CT)	S1: sprinkler & CT	S2: flood & CT
No tillage without crop stover (NT)	S3: sprinkler & NT	S4: flood & NT
No tillage with crop stover (NTr)	S5: sprinkler & NTr	S6: flood & NTr

Fertilizer operations were the same in the different treatments of every experiment. The management and conditions of these agronomic assays have been described in detail elsewhere [27–29].

The complete experimental plots of the two experiments were harvested with a commercial combine to obtain the maize yield (megagrams per hectare, Mg ha⁻¹). Samples analyzed for mycotoxins were taken from a global subsample of 2 kg of maize grains obtained after harvesting with the combine. Samples were dried at 60 °C. Grains were ground in a mill (IKA Labortechnik M20, Staufen, Germany), thoroughly homogenized, and kept at –21 °C until analysis.

2.2. Mycotoxin Analysis

The analyses were carried out with quantitative lateral flow immunoassays from VICAM[®] (Waters Corporation, Milford, MA, USA) according to the instructions of the manufacturer. The AFLA-V AQUA is a rapid flow lateral strip test for the quantification of aflatoxins (AFB1 + AFB2 + AFG1 + AFG2) in the range of 2–40 µg kg⁻¹ (limit of detection, LOD = 2 µg kg⁻¹). The FUM-V AQUA measures total fumonisins (FB1 + FB2 + FB3) in the range of 0.2–20 mg kg⁻¹ (LOD = 0.2 mg kg⁻¹). DON-V AQUA is a rapid flow lateral strip test for the quantification of deoxynivalenol in the range of 0.2–16 mg kg⁻¹ (LOD = 0.2 mg kg⁻¹). These methods are validated procedures for performing the analytical test for official inspections [30]. All analyses were carried out in accordance with the manufacturer's instructions using an aqueous-based extraction solution. The test strips were analyzed on a VICAM's Vertu lateral flow reader (Waters Corporation, Milford, MA, USA), which provides fast and quantitative results.

Certified reference material (CRM) is available to the authors' laboratory, and it regularly takes part in proficiency testing. With every analytical sample batch, certified reference materials purchased from Biopure (Romer Labs, Tulln, Austria) were analyzed to check the ongoing precision and recovery. In so doing, maize flour containing aflatoxins (AFB1 at 7.3 ± 2.4 µg kg⁻¹; AFB2, AFG1 and AFG2 at <1 µg kg⁻¹ each), maize flour containing 1232 µg kg⁻¹ fumonisin B1 and 282 µg kg⁻¹ fumonisin B2 and maize flour containing 1077 ± 73 µg kg⁻¹ of DON were used. The performance values obtained were within the acceptable margins outlined in Commission Regulation (EC) No. 401/2006 [31].

2.3. Statistical Analysis

A sample was determined as positive when the result was above the limit of detection (LOD). For descriptive statistics (mean and standard deviation SD) a value equal to one-half the LOD was assigned to the samples below this limit. First, the Shapiro-Wilk test was used to check if the observations fit the normal distribution model and the Levene test was conducted to examine the homogeneity of variances. The distribution of mycotoxins did not meet the requirements of normality and homoscedasticity except for FUM in the experiment of irrigation system and tillage practices. Therefore, all statistical differences of DON and FUM mycotoxin data were analyzed by Generalized Linear Mixed Models (GLMMs). For the experiment on irrigation time and frequency (field #1), the mycotoxin data was modeled using the Poisson distribution. For the experiment on the irrigation system and tillage practices (field #2), DON data were fitted using the Poisson distribution, while FUM data was treated as a normal (Gaussian) distribution. In both experiments, the year was considered as a random effect, while irrigation time and irrigation frequency as well as the irrigation system and soil tillage were considered as fixed factors for field #1 and field #2, respectively.

On the other hand, grain yield under both field experiments met the normality and homoscedasticity assumptions and significant differences between treatments were evaluated by ANOVA test. In both field experiments, the year was considered a random factor, while fixed factors varied between field experiments. Irrigation time and irrigation frequency and their interaction and irrigation type, and soil tillage and their interaction were considered as fixed factors under field experiment #1 and field experiment #2, respectively. When

significant, differences between treatments were identified at 0.05 probability level using the Tukey test. All statistical analyses were performed with JMP[®], Version 12 (SAS Institute Inc., Cary, NC, USA, 1989–2021).

3. Results

Agronomic recommendations usually emphasize maximum crop yield; however, their impact on mycotoxin accumulation needs consideration. This is why controlling mycotoxins together with high-yield performance could provide a significant guideline for maize producers. Therefore, the results of maize yield as affected by the variables studied are reported and analyzed in the present paper, although these data have been already discussed in two previously published works [27,29]. As regards mycotoxins, aflatoxins were not detected in any of the samples analyzed, so no results are shown. The concentrations of fumonisins and deoxynivalenol found in maize samples are described in detail below.

3.1. Effect of Irrigation Time and Irrigation Frequency

Regarding the first experiment (field #1), Table 3 shows the concentrations of DON and FUM and the grain yield (mean \pm standard deviation) as affected by irrigation time and irrigation frequency. DON levels were significantly greater at nighttime irrigation (0.54 mg kg^{-1}) than at daytime irrigation (0.38 mg kg^{-1}) and at low irrigation frequency (0.61 mg kg^{-1}) as compared to high frequency (0.30 mg kg^{-1}). Likewise, FUM levels were significantly higher when the sprinkler irrigation was performed at nighttime (1.21 mg kg^{-1}) as compared to daytime (0.45 mg kg^{-1}) and when irrigation frequency was low (1.09 mg kg^{-1}) in comparison with high frequency (0.57 mg kg^{-1}). However, interactions between irrigation time and frequency of irrigation showed no statistically significant differences for either mycotoxin.

Table 3. Deoxynivalenol (DON) and fumonisin (FUM) content in maize grain and crop yield as affected by the irrigation time (IT) and irrigation frequency (IF).

Effect	DON (mg kg^{-1})	FUM (mg kg^{-1})	Yield (Mg ha^{-1})
<i>Irrigation time (IT)</i>			
Day	$0.38 \pm 0.64 \text{ a}^1$	$0.45 \pm 0.90 \text{ a}$	$14.7 \pm 1.3 \text{ a}$
Night	$0.54 \pm 0.87 \text{ b}$	$1.21 \pm 1.33 \text{ b}$	$16.3 \pm 1.2 \text{ b}$
	$p = 0.0302$	$p = 0.0165$	$p = 0.0143$
<i>Irrigation frequency (IF)</i>			
Low	$0.61 \pm 1.00 \text{ a}$	$1.09 \pm 1.21 \text{ a}$	15.5 ± 1.2
High	$0.30 \pm 0.36 \text{ b}$	$0.57 \pm 1.14 \text{ b}$	15.5 ± 1.8
	$p = 0.0002$	$p = 0.0461$	$p = 0.6285$
<i>IT \times IF</i>			
Day \times Low	0.52 ± 0.88	0.80 ± 1.21	15.0 ± 1.1
Day \times High	0.23 ± 0.23	0.10 ± 0.00	14.3 ± 1.5
Night \times Low	0.70 ± 1.16	1.38 ± 1.21	16.0 ± 1.2
Night \times High	0.37 ± 0.45	1.04 ± 1.50	16.6 ± 1.2
	$p = 0.5743$	$p = 0.1230$	$p = 0.0838$

¹ For each variable and effect, values followed with different letters are significantly different at $p < 0.05$.

The highest maize yield was reached by irrigation at nighttime, which produced 16.3 Mg ha^{-1} , a significant average increase ($p = 0.0143$) of yield of +11% as compared to daytime irrigation. Nonetheless, the frequency of sprinkler irrigation did not affect grain yield, nor did the interactions between the factors analyzed.

3.2. Effect of Irrigation System and Tillage Practices

Regarding the second experiment (field #2), the effect of the irrigation system (flood system vs. sprinkler irrigation) and tillage practices (conventional tillage vs. no tillage with or without crop stover) on the levels of DON and FUM and the yield of grain

(mean \pm standard deviation) can be seen in Table 4. DON content of maize grain was affected by the irrigation system, with a significantly ($p = 0.0244$) higher concentration with sprinkler irrigation (0.53 mg kg^{-1}) as compared to flood irrigation (0.19 mg kg^{-1}). However, the fumonisin content of maize grain was not significantly affected by the irrigation system.

Table 4. Deoxynivalenol (DON) and fumonisin (FUM) content in maize grain and crop yield as affected by the irrigation system (IS) and soil tillage (ST).

Effect	DON (mg kg^{-1})	FUM (mg kg^{-1})	Yield (Mg ha^{-1})
<i>Irrigation system (IS)</i>			
Sprinkler (S)	$0.53 \pm 0.49 \text{ a}^1$	5.94 ± 3.26	15.3 ± 1.9
Flood (F)	$0.19 \pm 0.18 \text{ b}$	5.52 ± 2.89	13.6 ± 1.9
	$p = 0.0244$	$p = 0.6082$	$p = 0.1273$
<i>Soil tillage (ST)</i>			
Conventional tillage (CT)	0.36 ± 0.35	5.09 ± 3.17	15.1 ± 1.7
No tillage without crop stover (NT)	0.39 ± 0.52	5.84 ± 3.09	13.9 ± 1.8
No tillage with crop stover (NTr)	0.33 ± 0.32	6.25 ± 2.97	14.2 ± 2.6
	$p = 0.6443$	$p = 0.4415$	$p = 0.4353$
<i>IS \times ST</i>			
CT \times S	0.53 ± 0.42	5.74 ± 4.14	15.5 ± 1.6
NT \times S	0.55 ± 0.68	5.00 ± 2.83	14.7 ± 1.9
NTr \times S	0.51 ± 0.37	7.07 ± 2.62	15.6 ± 2.3
CT \times F	0.19 ± 0.14	4.44 ± 1.80	14.7 ± 1.7
NT \times F	0.23 ± 0.26	6.68 ± 3.26	13.2 ± 1.3
NTr \times F	0.14 ± 0.09	5.44 ± 3.22	12.8 ± 2.2
	$p = 0.4758$	$p = 0.1506$	$p = 0.1209$

¹ For each variable and effect, values followed with different letters are significantly different at $p < 0.05$.

Soil tillage did not affect the concentration levels of the mycotoxins evaluated. This result was unexpected since no tillage increases the amount of crop residue on the soil surface and generally leads to an increase in the inoculum of mycotoxin-producing fungi during subsequent harvests. There were no significant interactions between the irrigation system and soil tillage for DON or FUM in maize. Maize yields were higher than 15 Mg ha^{-1} under sprinkler irrigation and conventional tillage; however, the differences between treatments were not significant, nor were their interactions.

4. Discussion

Maize can be contaminated with fumonisins, deoxynivalenol, aflatoxins and other mycotoxins because of infection by toxigenic fungi, primarily in the genera *Fusarium* and *Aspergillus*. In the present study, the influence of some crop management practices on mycotoxin risk is discussed.

To begin with and concerning the aflatoxins, no conclusions could be drawn about the effect of the agronomic conditions on their contamination levels since these mycotoxins were not detected in any of the samples analyzed. In fact, the contamination with aflatoxins had not been normally perceived as a matter of concern for the primary production of maize in irrigated fields. However, with the advent of climate change, aflatoxins may become a problem in European maize, especially under coincident drought conditions and high temperatures. In addition, species of the genus *Aspergillus* are generally considered storage fungi as they usually contaminate products in the post-harvest stages, although they are sometimes also present in the field [32].

Nonetheless, different studies have reported significant occurrence of AFs in maize from some European countries over recent decades, even exceeding the EU maximum limits set for AFs in maize [6,33,34]. High temperatures and water stress can favor the growth of *Aspergillus* and subsequent aflatoxin production [9]. Indeed, AFB1 has been predicted to become a food safety issue in maize in Europe in the future due to climate change [35]. Therefore, it will be necessary to develop prevention and control strategies.

Maize crops under drought stress are believed to be more susceptible to contamination by both aflatoxins and fumonisins, and, consequently, research on irrigation and mycotoxin risk has been conducted. Water stress seems to be particularly critical for *A. flavus* during silk emergence and kernel ripening, and it has been identified as one of the most significant risk factors influencing *Fusarium* growth and fumonisin production [20,36]. Specifically, Marín et al. [22] observed that drought stress might cause induction of FUM1 expression of *F. verticillioides*, which might result in a high risk of fumonisin contamination of maize, particularly if it occurs during early maize reproductive growth [17]. Furthermore, excess moisture also may promote *Fusarium* infection. It has been reported that rain before harvest may intensify the contamination of fumonisins in maize [20]. Regarding this concern, maize seems to be more susceptible to *Fusarium* infection at the anthesis. Environmental humidity and rainfall during this stage of growth and in the course of the ripening of the crops are of great importance since they can favor the development of *Fusarium* infection, thus increasing the risk of mycotoxin contamination [21,37].

Deoxynivalenol mycotoxin in cereal grains is typically associated with high moisture levels after flowering. Thus, irrigation could increase the risk of DON contamination in maize, particularly sprinkler irrigation as compared to flood irrigation. In fact, as regards mycotoxin contamination of maize, flood irrigation is preferable to overhead systems so that the silks do not become excessively wet. Irrigation according to water needs is one of several good agricultural practices (GAP) that could be applied in the pre-harvest stages in order to improve field yields while mitigating the pathogens. It is even more important considering that maize yield could be adversely affected by projected climate change [38]. Studies carried out in this field reported that appropriate irrigation reduced *F. verticillioides* infection and fumonisin accumulation in Italian maize [24]. Higher yields have also been obtained by reducing drought conditions [39], although this also depends on the irrigation systems. However, in this cited study, there were no consistent significant differences in fumonisin and aflatoxin levels among different irrigation regimes and years in maize grains from crops with high, moderate and no irrigation. Alvarado-Carrillo et al. [40] observed better water use (>56% efficiency) and an increased yield (>20% efficiency) in maize when drip irrigation was used as compared to flood irrigation; however, the aflatoxin levels were not influenced by the irrigation system (16.4 $\mu\text{g kg}^{-1}$ vs. 15.6 $\mu\text{g kg}^{-1}$, respectively).

Irrigation is needed for growing maize in many parts of Spain, except for the humid northwestern regions. Thus, irrigation management should be optimized in terms of the amount and timing of irrigation to prevent drought stress and avoid excessive moisture. Quantifiable values for FUM and DON found in this study allowed us to draw some conclusions about the effect of different irrigation systems on the mycotoxin contamination of maize. According to the concentrations found, the sprinkler irrigation system significantly increased DON contamination of maize as compared to flood irrigation. This could be due to the fact that irrigation with sprinklers wets the maize canopy at each irrigation event (two irrigation events per week in this experiment), while flood irrigation does not wet the maize canopy (it only gets wet due to rainfall). Anyway, the levels of DON were below the EU maximum level set for unprocessed maize (1.75 mg kg^{-1}), and sprinkler irrigation can increase maize yield by 14 to 20% as compared to flood irrigation [29].

Regarding the management of sprinkler irrigation, both the irrigation time and frequency influenced the mycotoxin content. Thus, the concentration of DON and FUM in maize grain was significantly higher when irrigating at night and with low irrigation frequency. However, it has been reported that sprinkler irrigation at nighttime resulted in higher maize yield (+10%) than irrigation at daytime [27], and yield is a primary grower criterion. Therefore, in areas where deoxynivalenol and fumonisin levels are generally low (far below the EU thresholds set at 1.75 mg kg^{-1} for DON and 4 mg kg^{-1} for FUM), the potential increase in mycotoxin content with nighttime sprinkler irrigation should not cause the increase in maize yield obtained with this timing of irrigation to be overlooked.

Our results are consistent with other studies that have also stated that irrigation systems can have an influence on mycotoxin contamination of maize crops [17,25,40].

Additionally, it has been suggested that factors other than drought, such as insect damage, would be also responsible for determining mycotoxin levels in harvested grain [39].

Tillage practices are an important pre-planting factor as *Fusarium* and *Aspergillus* spores survive in crop stover through infected debris, and the chances for infection by these fungi are greatest if the crop stover is left on the soil surface. The risk posed by the inoculum levels of these fungi in crop stover can be reduced by tillage practices that bury the stover; however, the impacts can vary substantially by pathogen and by location. Specifically, Blandino et al. [41] have described that DON contamination in winter cereals is more severe if the preceding crop is maize, particularly when minimum tillage or no tillage is applied. So much importance has been given to tillage practices that some authors have prioritized land preparation such as tillage, cover crop and crop rotation when describing the fundamental GAP to control mycotoxin contamination in maize [21]. Nevertheless, it has been reported that these practices have a bigger impact on deoxynivalenol producers than on the fumonisin and aflatoxin-producing fungi [23].

However, in our study, tillage practices did not affect the incidence of deoxynivalenol or fumonisins. In agreement with our results, Marocco et al. [42] reported that tillage practices did not significantly affect the incidence of *F. verticillioides* and FB1 in maize fields. Nevertheless, other studies have found that fields with previous crop stovers had a higher presence of *F. verticillioides* and FB1 compared to fields without stovers, confirming that leaving crop stovers in maize fields should be avoided [25,43]. Notwithstanding, following these practices in individual fields may not have a significant impact on mycotoxin accumulation because of long-distance dispersal of spores from other fields. In fact, the role of reduced tillage in the content of such contaminants in maize grain is still debated [44]. Considering this, the potential benefits of tillage practices in maize must be weighed against their costs and their potential to promote soil erosion. In any case, it must be remembered that not only removing crop debris but also intercropping with other crops than maize is recommended GAP to minimize mycotoxin contamination. In truth, non-hosting crops on *F. verticillioides* should be selected when applying intercropping systems.

Insects act as vectors for fungal spores, and they cause damage to maize kernels, creating infection sites for toxigenic fungi, and inducing plant stress that may predispose the plants to infection. Particularly, fumonisins are most closely associated with insect injury [23]. In addition, the mono-cropping system and the favorable weather conditions promote the attack of insects [45]. Therefore, local information about pests and available scouting methods should be used to determine if insect populations would be high enough to warrant an insecticide application and determine the best time and method for the application to avoid mycotoxin risks.

This is sufficiently important that the European Commission pointed out that it is necessary to apply preventive measures to minimize fungal infection and insect damage to the crop, and, if necessary, to use approved and registered insecticides and fungicides under the conditions recommended by the manufacturers [46].

5. Conclusions

Proper irrigation and field management can be particularly relevant for mycotoxin control. Deoxynivalenol and fumonisins increased with nighttime sprinkler irrigation and at low sprinkler irrigation frequency indicating the relationship between these *Fusarium* toxins and water management in maize crops. In addition, deoxynivalenol increased in maize fields with sprinkler irrigation as compared to flood irrigation. In this regard, despite these higher levels of DON, and considering that the levels of this toxin were below the maximum value allowed by the EU, the sprinkler irrigation system should be recommended. This is because of the need to apply more sustainable agronomic practices (lower irrigation water and fertilizer rates due to higher water and fertilizer efficiency) and the higher maize yield under sprinkler irrigation. In contrast, the soil tillage treatments were not determinant in the mycotoxin concentrations detected in the present work, so reduction of tillage is possible without affecting the mycotoxin content of maize. This

study can be used in the framework of mycotoxin risk assessment in other temperate regions to identify the potential benefits of agronomic modifications based on adaptation to climate change.

Supplementary Materials: The following supporting information can be downloaded at <https://www.mdpi.com/article/10.3390/agronomy13030798/s1>. Table S1: Monthly (and daily range) mean air temperature (°C) and monthly cumulative rainfall (mm) during the experimental period in fields #1 and #2.

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