



# *Lumbricus terrestris* regulating the ecosystem service/disservice balance in maize (*Zea mays*) cultivation

Christine van Capelle · Friederike Meyer-Wolfarth · Torsten Meiners · Stefan Schrader

Received: 25 November 2020 / Accepted: 9 February 2021 / Published online: 20 February 2021  
© The Author(s) 2021

## Abstract

**Background and aim** Plant pathogenic and mycotoxin-producing *Fusarium* species are globally widespread and lead to large annual yield losses in maize production (ecosystem disservice). Systems with reduced tillage and mulching are particularly under threat. In the present study, the bioregulatory performance (ecosystem service) of the common earthworm species *Lumbricus terrestris* was analysed regarding the suppression of three economically relevant *Fusarium* species, and the reduction of their mycotoxins in the maize mulch layer, taking into account the size of maize residues.

**Methods** A mesocosm field experiment was conducted in a reduced tillage long-term field trial on loam soil. Artificially *Fusarium*-infected maize residues of two size classes were used as a mulch layer. Impacts of the earthworm species on DNA amounts of *Fusarium graminearum*, *F. culmorum*, and *F. verticillioides* and concentrations of the mycotoxins deoxynivalenol

(DON), 3-acetyldeoxynivalenol (3-AcDON), and zearalenone (ZEN) were analysed.

**Results** The results reflect that *Fusarium* regulation by *L. terrestris* was species-specific and covered the whole spectrum from suppression (*F. graminearum*) to slight promotion (*F. verticillioides*). Regarding the mycotoxins, a significant acceleration of the degradation of all three toxins was detected. Fine chopping of the chaff (< 2 cm) did not significantly alter the earthworms' regulatory capacity.

**Conclusion** While *L. terrestris* can shift the ecosystem service/disservice balance in both directions with respect to *Fusarium* regulation, it shifts it towards ecosystem services with respect to mycotoxin degradation. In synergy with adapted agricultural management, this natural bottom-up effect can help to keep soils healthy for sustainable production in the long run.

**Keywords** *Fusarium* · Earthworms · Bioregulation · Pathogen suppression · Mycotoxin degradation

---

Responsible Editor: Sven Marhan.

C. van Capelle (✉) · S. Schrader  
Institute of Biodiversity, Johann Heinrich von Thünen Institute,  
Braunschweig, Germany  
e-mail: christine.vancapelle@thuenen.de

F. Meyer-Wolfarth  
Institute for Plant Protection in Field Crops and Grassland, Julius  
Kühn Institute, Braunschweig, Germany

T. Meiners  
Institute for Ecological Chemistry, Plant Analysis and Stored  
Product Protection, Julius Kühn Institute, Berlin, Germany

## Introduction

*Fusarium* species are omnipresent in soils and on plants (Wenda-Piesik et al. 2017) and, as plant pathogenic fungi of cereals, play an essential economic role worldwide (Ferrigo et al. 2016). While in temperate latitudes especially the species *Fusarium graminearum* and *Fusarium culmorum* led to high yield losses in the past (Bottalico and Perrone 2002; Leplat et al. 2013), the distribution of *Fusarium verticillioides* (formerly

*Fusarium moniliforme*), as most common species in maize, was limited to warmer regions (Aguín et al. 2014; Bottalico 1998). However, due to rising temperatures in the context of climatic change, this species has increasingly spread even in originally colder, humid regions in recent years (Czembor et al. 2015; Pfordt et al. 2020). As the temperature rise continues and a further increase in maize cultivation is predicted (EC (European Commission) 2018; Pavlik et al. 2019), an increase in infestation rates by *F. verticillioides* in temperate latitudes is expected in the near future (Oldenburg et al. 2018).

Farmers in these regions are therefore confronted with the risk of infestation by a rising number of *Fusarium* species and face the major challenge of preventing and effectively controlling infections to keep their plants and soils healthy and to ensure sustainable yields. In this context, two aspects of infestation must be taken into account, in both, prevention and control measures. First, an infestation leads to various plant disease patterns such as *Fusarium* head blight, ear rot, or stem rot, usually associated with reduced crop yields (Ferrigo et al. 2016). Second, many *Fusarium* species can produce toxic metabolic products such as trichotecenes, fumonisins, or zearalenone. These mycotoxins pose a health risk to humans and animals (Ferrigo et al. 2016) and considerably restrict the harvested crop's usability in food and feed production. In Europe, corresponding maximum levels for *Fusarium* mycotoxins in unprocessed cereals and foodstuffs as well as recommendations for animal feed are laid down in respective EU regulations (EC (European Commission) 2006a, b, 2007).

The fact that *Fusarium* and its mycotoxins are present not only in the harvested crop but also in the plant residues remaining on the soil surface after harvesting, has received little attention so far. Agricultural systems with reduced tillage combined with mulching techniques are particularly at risk (Dill-Macky and Jones 2000; Wang et al. 2020). The plant material in the mulch layer serves as a suitable growth substrate that promotes the survival, development, and spread of *Fusarium* (Champeil et al. 2004; Leplat et al. 2013) over a more extended period than buried residues (Pereyra et al. 2004). As *Fusarium* can survive saprotrophically for several years (Champeil et al. 2004; Leplat et al. 2013), host plants can still be attacked years after the initial infestation. During the long saprotrophic survival phase of *Fusarium*, mycotoxins can be produced continuously (Perincherry et al. 2019). Their introduction

into the soil system is nearly impossible to prevent. Since there are hardly any studies on the long-term effects of mycotoxin inputs, their fate is still unclear, and the possible risks, e.g., for soil life or groundwater quality, cannot be foreseen (Elmholt 2008; Kolpin et al. 2014). Consequently, a long-term impairment of soil health, yield capacity and resilience of soils can therefore not be excluded.

The principals of Good Agricultural Practices (GAP) request farmers to reduce the inoculum and limit the risk for *Fusarium* incidence and spread in the field (EC (European Commission) 2006c; Joint FAO/WHO Codex Alimentarius Commission 2017). However, no specific maximum levels for the input of mycotoxins into soils have been set. The effectiveness of *Fusarium* control and mycotoxin reduction by agronomic top-down measures is limited (Joint FAO/WHO Codex Alimentarius Commission 2017), especially concerning maize cultivation (Li et al. 2019). Large quantities of crop residues remaining on the soil surface (Champeil et al. 2004; Morel 1996); current trends towards maize mono-cropping, especially for biofuel and feed production (Fargione et al. 2009; Tissier et al. 2016); the lack of highly resistant maize varieties (Ortiz et al. 2015; Zhang et al. 2012); and the limited effectiveness of available fungicides (Masiello et al. 2019; Wegulo et al. 2015) hamper the efforts to combat *Fusarium* infestation and prevent its spread. Linked to maize cultivation's economic value as essential crop production, its predicted increase in upcoming years (EC (European Commission) 2018), and the increasing implementation of reduced tillage as a contribution to sustainable production intensification (Kassam et al. 2009) also in maize cultivation systems (Claassen et al. 2018), it becomes apparent that *Fusarium* infections currently are, and in particular will be a challenge for securing high-quality yields, now and in the future.

Against this background and considering that soils treated by reduced tillage usually show higher functional soil biodiversity than ploughed soils (Pelosi et al. 2014), soil self-regulation and intrinsic biocontrol mechanisms as natural bottom-up effects have increasingly come into the focus of farmers and consultants in recent years. In order to make recommendations on how best to support the provision of the ecosystem service 'bioregulation' and thus benefit from it in the long-term, knowledge of the key organisms involved and a deeper understanding of the regulation by external factors are needed.

Several studies have been carried out on different groups of organisms, covering various size classes from microorganisms to macrofauna (e.g., Goncharov et al. 2020; Schrader et al. 2013). Concerning soil fauna, the results suggest that in particular the anecic primary decomposers within the earthworm community, which show a food preference for fungal-infected plant material, are suitable antagonists with high bioregulatory potential (Meyer-Wolfarth et al. 2017; Schrader et al. 2013; Wolfarth et al. 2011). In the agroecosystems of temperate regions, the earthworm species *Lumbricus terrestris* is a particularly promising representative, as it occurs in high densities in unploughed arable soils (Briones and Schmidt 2017; van Capelle et al. 2012) and prefers *Fusarium*-infected plant material as a food source (Bonkowski et al. 2000; Goncharov et al. 2020). Furthermore, there are indications that this species also promotes the degradation of *Fusarium* mycotoxins in plant residues (Oldenburg et al. 2008; Schrader et al. 2009; Wolfarth et al. 2016).

However, all these studies relate exclusively to wheat and, concerning mycotoxins, to deoxynivalenol (DON). Studies on *L. terrestris* in suppressing *Fusarium* and degrading various *Fusarium* mycotoxins in maize residues are lacking so far. Recommendations to farmers often emphasise the importance of chaff size in suppressing *Fusarium* infections. In this context, the general rule applies: the finer, the better. However, the relevance of size ranges for pathogen suppression and mycotoxin degradation has not yet been investigated.

Against the background of these gaps in knowledge, the objective of the present field study was to analyse the effectiveness of the bioregulatory potential of the earthworm species *L. terrestris* in suppressing the three *Fusarium* species *F. graminearum*, *F. culmorum*, and *F. verticillioides* and in accelerating the degradation of the main mycotoxins DON, 3-acetyledoxynivalenol (3-AcDON) and zearalenone (ZEN) in the maize mulch layer. Two different size classes of maize remains (fine straw and coarse straw) were considered to detect and assess residue size-specific differences.

The present study aims at testing the following overall hypothesis: earthworms are crucial bioregulators within the dynamic of the ecosystem service/disservice balance in maize residue mulching systems.

## Materials and methods

### Environmental conditions and site description

The experiment was conducted in the reduced tillage plots of a long-term soil tillage field trial located near Göttingen in Germany (study site: ‘Garte Süd’). Details on geographic location, climate, soil conditions, and set up of the field trial are given in Table 1.

The experiment was carried out in late summer 2018 after the harvest of rape. During the experimental time of six weeks, the mean air temperature was about  $13.7 \pm 0.6$  °C; the total precipitation was  $68.1 \text{ l m}^{-2}$ .

### Soil

Topsoil (Haplic Luvisol) was collected from the reduced tillage plots of the field trial. The soil was air-dried and stored at 4 °C until further processing. Shortly before the experiment started, the soil was macroscopically cleared of organic residues, sieved using a mesh size of 2 mm, and defaunated by three consecutive freezing (−18 °C) and thawing (room temperature) cycles of 24 h each. Finally, the soil was moistened to 17.34% (w/w), which corresponds to a water holding capacity (WHC) of about 60%.

### Maize residues

In 2017, silage maize (*Zea mays*, cultivar ‘Werena’) was cultivated at an experimental site of the Julius Kühn Institute in Braunschweig (Germany). Maize plants were artificially infected by *Fusarium* spp. injection in early August at the principal growth stage 6 of flowering and anthesis (BBCH 65) (Meier 2018) to receive *Fusarium*-infected and mycotoxin-contaminated residues for the experiment. Each maize plant was inoculated by injecting 0.5 ml spore suspension directly into the stem between the first and the second node. The spore suspension contained spores of three strains of the species *F. graminearum* in equal proportions. The total spore concentration was  $250,000 \text{ spores ml}^{-1}$ . The preparation of the spore suspension was carried out according to Oldenburg and Ellner (2015), who give a detailed description of the spore suspension preparation.

The maize stalks were chopped and divided into two size classes after harvesting: coarse straw (5–6 cm) and fine straw (1–2 cm). The material was air-dried until further processing. Although the maize plants were

artificially infected only with the species *F. graminearum*, the maize stalks contained DNA from *F. graminearum* and, additionally, *F. culmorum* and *F. verticillioides* after harvest (Table 2). Initially, the mycotoxins DON and 3-AcDON were found in both maize residue size classes (Table 2).

### Earthworms

Individuals of the primary decomposing anecic earthworm species *Lumbricus terrestris* were purchased from a commercial provider (Superwurm e.K., Düren, Germany). Two weeks before the start of the experiment, 24 adult individuals (clitellate) were adapted to the soil from the field trial at 17 °C ( $\pm 1$  °C). During this adaptation period, earthworms were kept in plastic containers and fed with non-infected control maize material. Before individuals were inserted into the experimental units (mesocosms), they were transferred into tap water to remove adherent organic material and mucus. Their biomass was determined by weighing ( $\pm 0.01$  g).

### Mesocosms as experimental units

Cylindrical mesh-bags made of nylon-gauze (diameter: 12 cm, height: 30 cm) were used as experimental units (mesocosms). A mesh size of 15  $\mu\text{m}$  ensured an exchange of air, water, and soluble nutrients with the surrounding soil in the field, but prevented other soil fauna from immigrating and earthworms and ascospores from escaping. Shortly before the experiment started, 24 mesocosms were filled with 1500 g (dw) moistened soil. The soil was compacted to a soil column of about 10 cm height, resulting in a bulk density similar to field conditions (Table 1). Two adult *L. terrestris* individuals were put into one half of the mesocosms (earthworm treatment), whereas the other half represented a non-faunal control (control). The weight-based allocation of individuals to mesocosms ensured nearly similar earthworm biomasses per experimental unit and treatment. Mean earthworm biomass per mesocosm was  $8.7 \pm 0.2$  g. When earthworms burrowed completely into the soil, 10 g of artificially infected, air-dried, and chopped maize stubbles were added to each mesocosm's soil

**Table 1** Site description and environmental conditions (climate, soil) of the field site 'Garte Süd' as well as experimental design of the long-term field trial. TOC = total organic carbon, TN = total nitrogen, CEC = cation exchange capacity

'Garte Süd'	
<u>Site</u>	
Biogeographic region	continental/atlantic
Geographic location	9 km south of Göttingen (DE)
GPS coordinates	51°29' N, 9°56' E
Altitude	163 m a.s.l.
<u>Climate</u>	
Mean annual air temperature	9.5 °C
Mean annual precipitation	621 mm y <sup>-1</sup>
<u>Soil</u>	
Type	Haplic Luvisol
Texture	Ut4 (19% clay, 68% silt, 13% sand)
pH	7.7
TOC	0.94%
TN	0.10%
CEC	11.5 cmol <sup>+</sup> kg <sup>-1</sup>
Bulk density	1.30 Mg m <sup>-3(a)</sup>
<u>Long-term field trial</u>	
Start	1970
Plot size	20 m × 40 m
Tillage	conventional tillage (plough, n=8), reduced tillage (rotary harrow, n=8)
Crop rotation	cereal-legume based (no repeated rotation) (2018: rape)

<sup>(a)</sup>Jacobs et al. (2009)

surface (about 113 cm<sup>2</sup>) and moistened by spraying with tap water (about 3 ml). This amount of plant material is equivalent to 8.8 t ha<sup>-1</sup>, representing standard mulching conditions for maize residues under reduced tillage (Morel 1996). Half of the mesocosms of each soil fauna treatment (earthworm treatment and control) received maize residues of one size class each (fine straw or coarse straw). Finally, mesocosms were closed by plastic clips.

### Experimental design

The experiment was carried out during six weeks from September 5 to October 10. In total, 24 mesocosms were established in the reduced tillage plots of the long-term field trial. The experiment comprised mesocosms of four different treatment combinations with  $n = 6$  per treatment: control/coarse straw, control/fine straw, earthworm treatment/coarse straw, and earthworm treatment/fine straw. Mesocosms were arranged in blocks of four (one per treatment with a distance of 1 m). One block of four mesocosms was inserted into each of six randomly selected reduced tillage plots, resulting in a replication of six per treatment. Mesocosms were buried in close contact with the surrounding soil, with the soil surface within and around mesocosms being at the same level.

**Table 2** Initial *Fusarium* species DNA amounts (*F. graminearum*, *F. culmorum*, *F. verticillioides*) and mycotoxin concentrations (DON, 3-AcDON, ZEN, NIV, FB<sub>1</sub>, and FB<sub>2</sub>) ± standard errors [ $\mu\text{g kg}^{-1}$ ] in both maize residue size classes (coarse straw, fine straw) at the beginning of the field experiment. ND = not detected

Cultivar	'Werena'	
	Coarse straw	Fine straw
DNA amount [ $\mu\text{g kg}^{-1}$ ]		
<i>F. graminearum</i>	10±2	18±9
<i>F. culmorum</i>	19,929±6956	2685±951
<i>F. verticillioides</i>	96,072±39,224	110,175±36,290
Mycotoxin concentration [ $\mu\text{g kg}^{-1}$ ]		
DON	3717±490	6405±650
3-AcDON	4233±352	8661±2745
ZEN	ND	ND
NIV	ND	ND
FB <sub>1</sub>	ND	ND
FB <sub>2</sub>	ND	ND

Due to Germany's dry weather conditions in 2018, mesocosms and surrounding soil were irrigated weekly by adding 10.4 l m<sup>-2</sup> of tap water per mesocosm. This measure was necessary to prevent the earthworms from entering a dormant stage to survive the unfavourable weather conditions.

### Determination of soil surface cover

Photographs of each mesocosm's soil surface area were taken at the beginning and the end of the field experiment. By scanning these top view photographs, relative shares [%] of uncovered soil or casts and areas covered with maize residues were evaluated using a colour analysis software specially developed to determine the degree of soil surface coverage (Programm zur Analyse von Bodenbedeckungsgraden aus Digitalfotos © 2005–2007 by Ulf Böttcher (CAU Kiel, Germany)).

### Sampling and sample processing

After six weeks of field exposure, mesocosms were removed from the field plots and taken to the laboratory. Mesocosms were opened, and the straw remaining on the soil surface was removed macroscopically. Maize residues were mechanically cleaned from adhesive soil (to prevent potential leaching of mycotoxins (Gautam and Dill-Macky 2012), washing was avoided), dried at 30 °C and finely ground using a batch mill (A10 basic, IKA®-Werke GmbH & Co.KG, Staufen, Germany). The ground plant material was further used to determine *Fusarium* species DNA amounts and to analyse mycotoxin concentrations.

Earthworms were removed from the soil columns, transferred to cold tap water to wash off adhering soil and organic material, and weighed individually.

Finally, two soil samples were taken from the well-mixed material of each soil column. The samples of one subset were used for the gravimetric determination of soil moisture after 24 h drying at 105 °C. The samples of the second subset were dried at 30 °C and manually ground (< 0.5 mm particle size) using a mortar and pestle. These samples were used for the determination of mycotoxin concentrations.

### Determination of *Fusarium* species DNA amounts

The DNA amounts ( $\mu\text{g kg}^{-1}$ ) of *F. graminearum*, *F. culmorum*, and *F. verticillioides* in maize residues

were determined using a quantitative real-time PCR (qPCR) based on TaqMan® technology to quantify the abundances of the three *Fusarium* species. For this purpose, the methods described in Brandfass and Karlovsky (2008) and Hogg et al. (2007) were modified. DNA was extracted from 1.0 g finely ground sample material. The qPCR was performed by an analytical laboratory specialized in the diagnosis of phytopathogens using biomolecular technologies (IDENTXX GmbH, Stuttgart, Germany). Detailed information on the methodological procedure can be obtained from IDENTXX GmbH, Stuttgart.

#### Determination of *Fusarium* mycotoxin concentrations

Initial maize residues as well as maize and soil samples from all mesocosms at the end of the experiment were analysed for the presence of the *Fusarium* mycotoxins DON, 3-AcDON, ZEN, nivalenol (NIV), fumonisin B1 (FB<sub>1</sub>), and fumonisin B2 (FB<sub>2</sub>). For the determination of mycotoxin concentrations, 1.0 g finely ground maize residues and 5.0 g homogenised soil material were extracted by turbulent shaking for 30 min. An acetonitrile/water mix (50:50) was used as an extraction solution. The extracts were then diluted 1:10 with 30% methanol. 10 µl of the purified filtrates were analysed by a Thermo scientific DIONEX UltiMate 3000 HPLC system. The column was a Phenomenex Kinetex C18 (2.6 µm, 100 mm, 3 mm i.d.). The mobile phase consisted of solvent A (methanol +0.5% acetic acid +5 mmol ammonium acetate) and solvent B (water +0.5% acetic acid +5 mmol ammonium acetate). A gradient procedure was used as followed: starting with 2% of A: up to 98%. The flow rate was 300 µl/min, and the column temperature was set at 40 °C. The HPLC was coupled with the mass spectrometer QTRAP 5500 (AB SCIEX) used in electrospray ionization mode.

The detection limit for DON, 3-AcDON, ZEN, NIV, FB<sub>1</sub>, and FB<sub>2</sub> was 1 µg kg<sup>-1</sup>. For more information on mycotoxin determination, see Oldenburg and Ellner (2015).

#### Statistics

Changes in *Fusarium* DNA levels, mycotoxin concentrations, and accompanying parameters (soil moisture, soil surface cover, earthworm biomass) during the experimental period were calculated using the formula  $\log(X(t_1)/X(t_0))$ , where X is the respective value of a

parameter at the start time  $t_0$  and the end time  $t_1$  of the experiment (Crawley 2007), to ensure relative comparability between increases and decreases. In cases where either initial or final values were zero (ZEN concentration), one was added to both concentrations to determine comparable value shifts. Impacts on the log-rates of change were analysed using linear mixed-effect models. Due to the nested experimental design, plots were generally considered as a random factor.

Changes of all parameters were analysed for an effect of the residue size class. In terms of soil moisture, pathogen suppression, and mycotoxin degradation, moreover, the soil fauna treatment (control vs. earthworm treatment) and the two-way interaction between both factors were integrated into the model.

Since the incorporation of maize residues into the soil presupposes earthworms' presence and activity, the statistical evaluation of the decrease in soil surface cover refers exclusively to the mesocosms with earthworms.

Model residuals were checked visually (normal quantile-quantile plots (QQ plots), residual plots, boxplots) and by testing procedures (Shapiro-Wilk normality test, Levene test) to evaluate normality and homogeneity of variance. In cases in which the assumption of the normal distribution of residuals or the homogeneity of variances was violated, data were transformed by either exponential transformation ( $e^x$ ) (data sets: *F. graminearum* DNA amount, 3-AcDON and ZEN concentrations) or square root transformation (data set: earthworm biomass) to fulfill the model requirements.

Analysis of Deviance (Type II Wald chi-square test) was performed to analyse the impacts of explanatory variables and their interaction. Correlations between parameters were analysed by use of Pearson correlation.

To make comparative statements on the relevance and regulation of individual species and mycotoxins, changes of relative proportions [%] within the overall spectrum were analysed. To investigate whether percentages of the three *Fusarium* species and the different mycotoxins in maize residues changed under field conditions and depending on treatment, a PERMANOVA was performed (number of permutations: 9999). Two different comparisons were carried out for each of the two residue size classes: firstly, a comparison of the initial material with the control material after completion of the experiment, and secondly, a comparison between the control material and the material from the earthworm treatment at the end of the experimental period. In terms

of the first comparison, time (start vs. end) and residue size class; in terms of the second comparison, fauna treatment, residue size class, and plot were considered as explanatory variables.

All statistics were performed using R version 3.5.0 (R Core Team 2018). The packages lme4 (Bates et al. 2015), MASS (Venables and Ripley 2002), and car (Fox and Weisberg 2019) were used for linear mixed modelling. The package corrplot (Wei and Simko 2017) was used to analyse Pearson correlations, the package vegan (Oksanen et al. 2019) for analysis of relative changes of DNA amounts and mycotoxins via PERMANOVA. The figure was created using the ggplot2 package (Wickham 2016).

## Results

### Soil moisture

In the course of the experiment, the soil moisture in the mesocosms decreased by about  $9.3 \pm 0.6\%$ , resulting in an average water holding capacity of  $58.56 \pm 0.36\%$  at the end of the experimental period (Supplementary Table S1). Soil moisture changes differed significantly between control ( $-10.8 \pm 0.7\%$ ) and earthworm treatment ( $-7.8 \pm 0.7\%$ ) (Table 3), resulting in higher average soil moisture in mesocosms with earthworms compared to the control (Supplementary Table S1). No significant residue size class or interaction effects were detected (Table 3).

### Soil surface cover

During the experimental period, earthworms incorporated plant material into the soil. Accordingly, in mesocosms with *L. terrestris*, the surface area of soil

covered by maize residues decreased, whereas it remained unchanged at 100% in the control (Supplementary Table S1). Since fine straw was incorporated into the soil more effectively than coarse straw, the decrease of soil surface cover significantly differed between residue size classes (fine straw:  $-16.0 \pm 1.8\%$ , coarse straw:  $-5.1 \pm 1.3\%$ ) (Supplementary Table S1, Table 3). No correlation between changes in soil moisture and soil surface cover was detected (Supplementary Table S2).

### Recapture rate and biomass of earthworms

After completion of the experiment, the loss of one of a total of 24 *L. terrestris* individuals was recorded. The recapture rate was thus 96%. All recaptured individuals were active. None of them entered a dormant stage.

Mean earthworm biomass decreased during the experimental period by about  $-7.3 \pm 1.4\%$  (Supplementary Table S1). Reduction rates did not significantly differ depending on residue size class (Table 3) and were not correlated with changes in soil moisture or soil surface cover (Supplementary Table S2).

### *Fusarium* DNA amounts

DNA amounts of all three *Fusarium* species in maize residues increased during the duration of the experiment, independent of treatment (Fig. 1, Supplementary Table S3). Average rates of increase ranged from eight (*F. verticillioides* in the coarse straw of the control) to 17,200 (*F. culmorum* in the fine straw of the control) times the initial value.

Average increases in amounts of *F. graminearum* DNA were significantly higher in the control (200-fold) compared to the earthworm treatment (about 40-fold) and in fine straw (approximately 200-fold)

**Table 3** Chisquare ( $\chi^2$ ) values and significance levels of Analysis of Deviance (Type II Wald Chisquare Test) on the effects of fauna treatment (control vs. earthworm treatment) and residue size (coarse straw vs. fine straw) on changes of soil moisture, soil

surface cover and earthworm biomass [%]. Significant results are printed in bold. Degrees of freedom (Df) for all terms equal 1. NA = not applicable/no data available

	Soil moisture	Soil surface cover	Earthworm biomass
Fauna treatment (F)	<b>9.017**</b>	NA	NA
Residue size (R)	0.255	<b>21.083***</b>	1.252
F x R	0.404	NA	NA

Significance codes: < 0.001\*\*\*; < 0.01\*\*; < 0.05\*; < 0.1(\*)

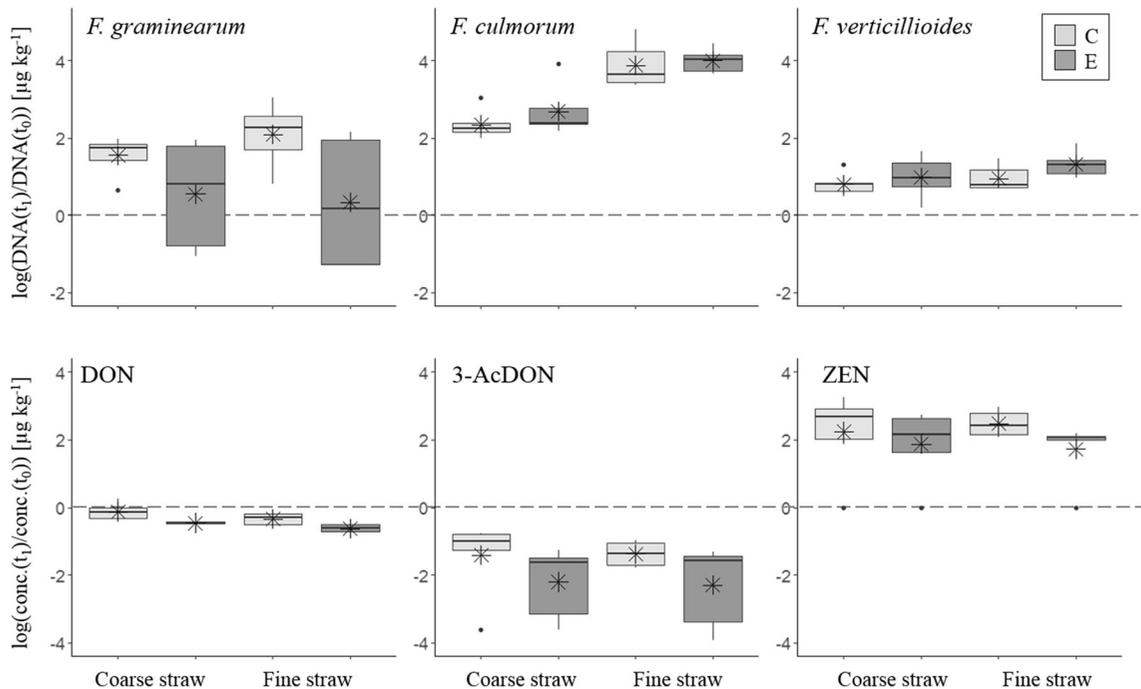
compared to coarse straw (about 50-fold) (Fig. 1, Table 4). The earthworm-induced suppression of fungal growth, quantified as smaller increases in *F. graminearum* DNA levels compared with the control, tended to be more pronounced in fine straw than in coarse straw (Fig. 1, Table 4). Average increases in *F. culmorum* DNA amounts were significantly higher in fine straw (about 15,000-fold) than in coarse straw (about 1000-fold), but showed no differences depending on the presence of earthworms (Fig. 1, Table 4). DNA amounts of *F. verticillioides*, by contrast, showed a tendency of a stronger increase in the earthworm treatment (about 20-fold) compared to the control (10-fold) but did not differ depending on residue size class (Fig. 1, Table 4).

The shifts in DNA quantities of *F. culmorum* and *F. verticillioides* were positively correlated ( $r=0.473$ ,  $p=0.020$ ) (Supplementary Table S2). In the earthworm treatment, *F. culmorum* DNA amounts increased with decreasing soil surface cover ( $r=-0.595$ ,  $p=0.041$ ) (Supplementary Table S2), indicating increased growth

rates of this species after incorporation of maize residues into the soil.

#### Mycotoxin concentrations

At the end of the experiment, the three *Fusarium* mycotoxins DON, 3-AcDON, and ZEN were detected in the maize residues (Fig. 1, Supplementary Table S4). FB<sub>1</sub> was found in low concentration in only one sample (coarse straw in the control) (Supplementary Table S4). Contamination of maize residues with the mycotoxins NIV and FB<sub>2</sub> has not been detected at any time. While DON and 3-AcDON were already present in the initial material, ZEN was newly formed during the field experiment. Average concentrations of DON and 3-AcDON decreased during field exposure. Rates of decrease were significantly higher in the earthworm treatment (DON:  $-70.0 \pm 2.5\%$ , 3-AcDON:  $-97.7 \pm 0.5\%$ ) compared to the control (DON:  $-33.0 \pm 11.6$ , 3-AcDON:  $-92.4 \pm 1.6\%$ ) (Fig. 1, Table 4). Analogously, average rates of new ZEN formation were also significantly lower in mesocosms with earthworms



**Fig. 1** Changes in DNA amounts (DNA) of the species *F. graminearum*, *F. culmorum*, and *F. verticillioides* (shown above) and in concentrations (conc.) of the mycotoxins DON, 3-AcDON and ZEN (shown below)  $\pm$  SE [ $\mu\text{g kg}^{-1}$ ] during the experimental runtime, presented as  $\log(X(t_1)/X(t_0))$ , with  $t_0$  = start

time and  $t_1$  = end time of the experiment, in the control (C) and the earthworm treatment (E), shown for both residue size classes (coarse straw, fine straw). Stars indicate arithmetic means. Values  $<0$  indicate a decrease, values  $>0$  an increase

compared to the control (Fig. 1, Table 4). An effect of residue size class, with a significantly higher decrease in fine straw ( $-62.6 \pm 5.6\%$ ) than in coarse straw ( $-40.3 \pm 12.1\%$ ), was only detected for DON (Fig. 1, Table 4). No significant effect of the interaction between earthworm treatment and residue size class was detected for any of the three toxins (Table 4).

Changes in 3-AcDON contamination were positively correlated with shifts in both, DON ( $r = 0.580$ ,  $p = 0.003$ ) and ZEN ( $r = 0.424$ ,  $p = 0.039$ ) concentrations (Supplementary Table S2).

No mycotoxins could be detected in the soil samples, as concentrations were generally below the detection limit.

The results of the linear mixed-effect models on the effects of soil fauna treatment (control vs. earthworm treatment), maize residue size (coarse straw vs. fine straw), and the interaction between them on the (partially transformed) log-rates of changes in accompanying parameters (soil moisture, soil surface cover and earthworm biomass), *Fusarium* species DNA amounts, and mycotoxin concentrations are given in the supplementary Tables S5 and S6.

#### Proportional analysis

In the initial maize residues applied to the mesocosms, *F. verticillioides* represented the dominant *Fusarium*

species in both residue size classes with mean proportions of about 71% in coarse straw and about 96% in fine straw (Table 5). Accordingly, the relative proportion of *F. culmorum* was higher in coarse straw (about 29%) than in fine straw (about 4%). Less than 0.1% of the *Fusaria* present in both residue size classes belonged to *F. graminearum* (Table 5).

During the experiment, this distribution shifted significantly (Table 6) in favour of *F. culmorum*, which accounted for more than 80% in the control at the end of the experiment (Table 5). This shift was significantly higher in fine straw than in coarse straw (Tables 5 and 6). The comparison of the *Fusarium* community in maize residues from the control with that in residues from the earthworm treatment affirms a tendency towards higher proportions of *F. culmorum* and lower percentages of *F. verticillioides* in fine straw compared to coarse straw (Tables 5 and 6). The presence of earthworms did not significantly affect the shifts in species' relative abundances (Table 6).

Regardless of the residue size class, about 54% of the mycotoxin contamination in the initial material was 3-AcDON and about 46% DON. ZEN and FB<sub>1</sub> were not detected (Table 5). In the course of the experiment, this ratio shifted significantly (Table 6), resulting in a final contamination in which DON accounted for the highest toxin content at over 76% (Table 5). About 10% of the toxin load was 3-AcDON and between 11 and 14% the

amounts and mycotoxin concentrations [ $\mu\text{g kg}^{-1}$ ]. Significant results are printed in bold. Degrees of freedom (Df) for all terms equal 1

**Table 4** Chisquare ( $\chi^2$ ) values and significance levels of Analysis of Deviance (Type II Wald Chisquare Test) on the effects of soil fauna treatment (control vs. earthworm treatment) and residue size (coarse straw vs. fine straw) on changes of *Fusarium* DNA

<i>Fusarium</i> DNA amount		Mycotoxin concentration	
<i>F. graminearum</i>		DON	
Fauna treatment (F)	<b>10.529**</b>	Fauna treatment (F)	<b>17.204***</b>
Residue size (R)	<b>4.562*</b>	Residue size (R)	<b>6.283*</b>
F x R	3.395(*)	F x R	0.061
<i>F. culmorum</i>		3-AcDON	
Fauna treatment (F)	1.262	Fauna treatment (F)	<b>11.358***</b>
Residue size (R)	<b>48.243***</b>	Residue size (R)	0.551
F x R	0.300	F x R	0.543
<i>F. verticillioides</i>		ZEN	
Fauna treatment (F)	3.096(*)	Fauna treatment (F)	<b>4.572*</b>
Residue size (R)	2.369	Residue size (R)	0.419
F x R	0.333	F x R	0.054

Significance codes: < 0.001\*\*\*; < 0.01\*\*; < 0.05\*; < 0.1(\*)

**Table 5** Relative amounts  $\pm$  standard errors [%] of *Fusarium* species DNA (*F. graminearum* (*F. gram.*), *F. culmorum* (*F. culm.*), *F. verticillioides* (*F. vert.*)) and *Fusarium* mycotoxins (DON, 3-AcDON, ZEN, FB<sub>1</sub>) in initial maize residues (Start) and

at the end of the experiment in the control (Con.) and the earthworm treatment (Earthw.) in both residue size classes (coarse straw, fine straw). Relative amounts >50% are listed in bold. ND = not detected

		<i>Fusarium</i> DNA amount [% $\pm$ SE]			<i>Fusarium</i> mycotoxins [% $\pm$ SE]			
		<i>F. gram.</i>	<i>F. culm.</i>	<i>F. vert.</i>	DON	3-AcDON	ZEN	FB <sub>1</sub>
Coarse straw	Start	0.07 $\pm$ 0.05	29.06 $\pm$ 10.05	<b>70.88<math>\pm</math>10.10</b>	46.41 $\pm$ 1.31	<b>53.59<math>\pm</math>1.31</b>	ND	ND
	Con.	0.01 $\pm$ 0.00	<b>82.60<math>\pm</math>6.29</b>	17.38 $\pm$ 6.29	<b>76.04<math>\pm</math>5.21</b>	9.64 $\pm$ 2.79	14.23 $\pm$ 5.23	0.09 $\pm$ 0.08 <sup>(a)</sup>
	Earthw.	0.01 $\pm$ 0.00	<b>84.60<math>\pm</math>6.03</b>	15.39 $\pm$ 6.03	<b>80.73<math>\pm</math>5.28</b>	5.78 $\pm$ 1.95	13.49 $\pm$ 4.90	ND
Fine straw	Start	0.02 $\pm$ 0.01	4.03 $\pm$ 1.97	<b>95.95<math>\pm</math>1.97</b>	45.69 $\pm$ 4.74	<b>54.31<math>\pm</math>4.74</b>	ND	ND
	Con.	0.02 $\pm$ 0.01	<b>94.72<math>\pm</math>1.04</b>	5.26 $\pm$ 1.04	<b>78.96<math>\pm</math>3.41</b>	10.37 $\pm$ 1.62	10.67 $\pm$ 3.95	ND
	Earthw.	0.01 $\pm$ 0.00	<b>92.08<math>\pm</math>0.36</b>	7.91 $\pm$ 0.36	<b>85.75<math>\pm</math>2.88</b>	8.60 $\pm$ 2.67	5.66 $\pm$ 1.58	ND

<sup>(a)</sup> detected in one sample

**Table 6** Results (sums of squares (Sums of Sqs.), R<sup>2</sup> and *p* values) of multivariate permutation analysis of variance using distance matrices to analyse the homogeneity of the distribution of *Fusarium* species DNA amounts and mycotoxins [%] in maize residues. Comparison of the distribution in initial residues and in the control at the end of the experiment (shown above), and between control and earthworm treatment at the end of the experiment (shown below). Number of permutations: 9999. Significant results are printed in bold

	Sums of Sqs.	R <sup>2</sup>	<i>p</i> value
Initial residues vs. Control			
DNA amount [%]			
Time (T)	2.0801	0.8455	<b>0.0002***</b>
Residue size (R)	0.0002	0.0001	0.9155
T x R	0.1383	0.0562	<b>0.0158*</b>
Mycotoxins [%]			
Time (T)	0.7187	0.7648	<b>0.0001***</b>
Residue size (R)	0.0007	0.0008	0.9431
T x R	0.0002	0.0003	0.9741
Control vs. Earthworm treatment			
DNA amount [%]			
Fauna treatment (F)	0.0001	0.0002	0.9345
Residue size (R)	0.0577	0.1703	0.0505 <sup>(*)</sup>
Plot	0.0556	0.1642	0.6198
F x R	0.0032	0.0096	0.5915
Mycotoxins [%]			
Fauna treatment (F)	0.0177	0.0454	0.3321
Residue size (R)	0.0160	0.0410	0.3570
Plot	0.1084	0.2774	0.2726
F x R	0.0020	0.0052	0.8690

Significance codes: < 0.001\*\*\*; < 0.01\*\*; < 0.05\*; < 0.1<sup>(\*)</sup>

newly formed ZEN (Table 5). These shifts within the toxin spectrum were independent of the residue size (Table 6). Relative proportions of *Fusarium* toxins did not differ significantly between maize residues from the control and those from the earthworm treatment (Table 6).

## Discussion

In the present study, the ecosystem service/disservice balance provides information on the relationship between infestation pressure as well as mycotoxin contamination by phytopathogens (here: three *Fusarium* species) and the bioregulatory potential of natural antagonists (here: *L. terrestris*). The regulatory processes observed in the control treatment exclusively indicate soil microbial activity, while those in the earthworm treatment reflect single and interaction effects between earthworms (incl. associated microorganisms) and soil microorganisms. The findings help to better understand natural bottom-up bioregulation pathways in maize cultivation and to evaluate their effectiveness in the context of the synergy effect between farmer and soil fauna (Meyer-Wolfarth et al. 2017; Schrader et al. 2020). The knowledge gained about the functional relationships and interactions is of great relevance both now and in the future, as reduced tillage in combination with mulching techniques is becoming increasingly important worldwide as a contribution to sustainable agricultural production, including in maize cultivation (Claassen et al. 2018; Kassam et al. 2009).

The maize straw showed an apparent infestation with *F. culmorum* and *F. verticillioides* at the beginning of the experiment, although only the species *F. graminearum* was artificially injected. Therefore, this infestation is due to natural infection in the field during the growth phase of the maize plants. The high proportion of *F. verticillioides* DNA (over 70%) reveals that this species, which is originally native to warmer and drier regions (Aguín et al. 2014; Bottalico 1998), is already present in temperate latitudes. This result is in line with Czembor et al. (2015) and Pfordt et al. (2020), who have recently detected this species in Poland and Germany. It underlines the assumption of Oldenburg et al. (2018) that *F. verticillioides* will play an increasing role in maize cultivation in temperate latitudes in the future. The relative proportion of the initially inoculated species *F. graminearum*, by contrast, was the lowest within the *Fusarium* community (< 0.1%) at any time and in any treatment. This result supports the assumption of Leplat et al. (2013) and Pereyra and Dill-Macky (2008), who classified the species *F. graminearum* as a relatively weak competitor compared to other *Fusarium* species and within the soil fungal community. The frequently described high competitiveness of this species (Velluti et al. 2000; Xu et al. 2007) is not confirmed in the present study. It is known that several anthropogenic factors, including preceding crops, tillage system, and weed management, can alter the development of the soil biota, which in turn can change the saprotrophic development of *F. graminearum* (Leplat et al. 2013). Thus, differences in farming practice might explain differences in competitive ability besides the origin of fungal species and isolates (see below).

During the mesocosm experiment, DNA levels of all three *Fusarium* species in maize residues increased in the control treatment. The main reason for this biomass increase was probably the enhanced water availability under field conditions compared to the initial dried material, which represents the most essential factor for *Fusarium* growth besides temperature (Belizán et al. 2019). With an arithmetic mean of 14 °C, the temperature during the experimental runtime was well below the respective temperature optimum, but with a value of over 10 °C still in a range in which growth of all three species could be expected (Brennan et al. 2003; Cook and Christensen 1976). The third factor that significantly influences the growth of *Fusarium* species as a function of humidity and temperature is the origin of species

and isolates and the time available for their adaptation to specific climatic conditions (temperature ecotypes) (Brennan et al. 2003; Hudec and Muchova 2010; Pettitt et al. 1996). These different adaptation stages to the conditions of temperate latitudes are reflected in the detected growth rates, which were highest for *F. culmorum*, followed by *F. graminearum* and *F. verticillioides* (Fig. 1).

The species *F. culmorum* was the predominant *Fusarium* species in Central and Northern Europe until the 1990s as the main pathogen of maize stem rot (Bottalico 1998). Long-term adaptation to climate conditions in this part of Europe probably contributed to the high biomass increase of *F. culmorum*, which represented the dominant species at the end of the experiment with a share of over 80% in all treatments. The species *F. graminearum*, by contrast, was originally native to warmer and humid regions (Brennan et al. 2003) and has only been present in the colder regions of temperate latitudes since the 1980s (van der Lee et al. 2015; Waalwijk et al. 2003). Currently, *F. graminearum* is often considered the dominant species in most cereal growing areas worldwide (Goswami and Kistler 2004; Manstretta and Rossi 2016). It is frequently described as displacing *F. culmorum* (van der Lee et al. 2015; Waalwijk et al. 2003). Slower growth rates and lower biomasses compared to *F. culmorum*, as demonstrated in the present study, were also detected in wheat by Brennan et al. (2003) and Xu et al. (2007). The third species, *F. verticillioides*, originates from even warmer regions (Aguín et al. 2014). This species probably does not yet show a pronounced adaptation to the variable climatic conditions in late summer or autumn. Thus, its growth rates were the lowest compared to the other two *Fusarium* species in the present study. Overall, the demonstrated growth rates and relative abundances of the three species link the conclusions of Hudec and Muchova (2010) and Pfordt et al. (2020) as they indicate that species and factors of their original latitude are the key factors that determine *Fusarium* growth rates and species spectrum at a specific temperature and humidity in maize cultivation.

Regarding an effect of the maize residue size classes, the two species *F. graminearum* and *F. culmorum* showed a higher DNA increase in fine straw than in coarse straw. This effect can be explained by the fact that the finely chopped material offers a higher proportion of surfaces. This favours the direct contact of *Fusarium* fungi, which are mainly contained in the

maize stalk pith (Zhang et al. 2016), with soil water and nutrients. As the moisture requirements of *F. culmorum* and *F. graminearum* are higher compared with the drought-adapted species *F. verticillioides* (Marín et al. 1996; Torres et al. 2003), they benefited more from the splitting of the plant material. In addition, a finer crushing of the maize straw allows quick colonization by soil-borne fungi and bacteria. Diverse interactions with these soil microorganisms regulate the growth of *Fusarium* fungi through direct, indirect, and mixed-path mechanisms (Wachowska et al. 2017). Whereas in the literature, mainly antagonistic effects of the soil microbiome against *Fusarium* species are described (Wachowska et al. 2017), the present results indicate a species-specific increase. In total, these results suggest that, without soil fauna, a smaller chaff size of maize mulch may stimulate specific *Fusarium* species and even increase the infestation pressure caused by them.

The bioregulatory capacity of the detritivore earthworm species *L. terrestris* on Fusaria was species-specific: *F. graminearum* was suppressed, *F. culmorum* was not affected, and *F. verticillioides* was slightly promoted. Accordingly, with regard to *Fusarium* regulation, the hypothesis of the present study was confirmed for only two out of three species, a shift of the service/disservice balance towards service (pathogen suppression) even for only one species. The present results for maize residues contradict the results for wheat straw of Meyer-Wolfarth et al. (2017), Oldenburg et al. (2008), Schrader et al. (2009), and Wolfarth et al. (2011), who demonstrated suppression of *F. culmorum* by *L. terrestris*. These different effects suggest that the cultivated plant and potentially even the respective cultivar plays an important role not only for the composition of the *Fusarium* community (Czembor et al. 2015) and their respective growth rates (Brennan et al. 2003) but also for the interaction between *Fusarium* species and soil fauna.

The chaff size of maize residues played only a minor role during earthworm-induced regulation of *Fusarium* growth. The tendency of a stronger suppression in fine straw than in coarse straw was detected for *F. graminearum* only. Since the earthworm biomass did not differ significantly depending on the residue size class, and the mean weight reduction of less than 10% over six weeks indicates an adequate nutrient supply (Fründ et al. 2010), good usability of both chaff sizes as a food source for *L. terrestris* can be assumed. The decreasing soil surface cover in the earthworm treatment

(fine straw > coarse straw) indicates incorporation of the residues into the soil and the burrow system. According to the ‘external rumen’ principle, which is based on the definition of Swift et al. (1979) and specified for earthworms by Lavelle (1988) and Brown et al. (2000), this organic material is stored in the burrows until it has been further split and pre-decomposed by the microbial community. The stronger suppression of *F. graminearum* in fine straw was probably not caused by direct feeding of earthworms, but rather by the priming effect of earthworm mucus, being highly bioavailable for soil microorganisms (Binet et al. 1998; Schrader et al. 2013). It can be assumed that due to this effect and the higher soil moisture in the earthworm treatment, competing or antagonistic soil microorganisms colonized the fine straw faster than the coarse straw. Comparable effects regarding a stronger reduction of *F. graminearum* in strongly split compared to intact maize residues were also demonstrated in field experiments of Vogelgsang et al. (2011).

During the experimental period, DON and 3-AcDON were reduced in the control due to microbial degradation and transformation (Vanhouette et al. 2016; Venkatesh and Keller 2019; Wachowska et al. 2017), which was faster for 3-AcDON compared to DON. While DON and 3-AcDON were already formed in the growing maize plant, ZEN was only produced in the chaff of the maize mulch layer. Unlike DON (Proctor et al. 1995; Snijders 1995), ZEN does not play a significant role as a virulence factor and for disease development in the living plant (Munkvold 2017), but can inhibit the formation of certain soil-borne microorganisms (Bacon et al. 2017) and thereby lead to a competitive advantage in the saprotrophic phase of Fusaria. Thus, this result supports the theory described by Müller et al. (2014) and Venkatesh and Keller (2019) of the formation of specific mycotoxins due to interactions with microorganisms and proves that the toxin composition in the living plant can differ considerably from that in the mulch layer. FB<sub>1</sub> is the most common mycotoxin produced by *F. verticillioides* in maize (Bottalico 1998; Czembor et al. 2015) and probably plays a role in suppressing the plant’s defense reaction (Galeana-Sánchez et al. 2017). However, in the present study, this mycotoxin was not formed during plant growth and was only detected in a single sample in the mulch layer. In accordance with the results of Ryu and Bullerman (1999), a correlation between growth rates of the three *Fusarium*

species and detected changes in toxin concentrations was not found in the present study.

Concerning the residue size classes, in line with the study of Vogelgsang et al. (2011), the present results reflect a significantly stronger reduction of DON in fine than in coarse straw, but no effect on the degradation of 3-AcDON or ZEN. Since mycotoxins serve as communication signals in fungal-bacterial interactions, some bacteria possess the ability to degrade or transform certain toxins or either promote or inhibit their formation (Vanhoutte et al. 2016; Venkatesh and Keller 2019). The results of the control treatment reflect that the degradation of *Fusarium* toxins in the mulch layer specifically depends on the composition of the respective microbial community.

The earthworms (*L. terrestris*) significantly accelerated the reduction of all three toxins, revealing their bioregulatory potential for mycotoxin degradation. With regard to mycotoxins, the hypothesis of the present study was confirmed and an earthworm-induced shift of the service/disservice balance towards service was demonstrated. At the end of the experiment, the maize material in the control, which was only affected by soil microorganisms, still had 1.8 times the maximum legal level of DON and 1.5 times the maximum level of ZEN for unprocessed maize (EC (European Commission) 2007). By contrast, in the earthworm treatment, maximum legal levels for unprocessed maize were significantly undercut with a 0.8-fold concentration for DON and a 0.5-fold concentration for ZEN. Overall, the earthworms reduced the DON concentration by half and 3-AcDON and ZEN concentrations by two thirds over the experimental period compared to the control. These results are consistent with the studies of Meyer-Wolfarth et al. (2017), Oldenburg et al. (2008), Schrader et al. (2009), and Wolfarth et al. (2011), who demonstrated a reduction of DON in wheat straw by *L. terrestris*. Effects of earthworms thereby resulted from a combination of direct and indirect bioregulatory processes. The feeding activity reduced the mycotoxin concentration by degradation processes in the course of intestinal passage, presumably with the participation of the intestinal flora (Schrader et al. 2013). For DON, this is shown by the studies of Oldenburg et al. (2008) and Schrader et al. (2009), in which it was demonstrated that DON concentrations in the intestine of *L. terrestris* were significantly lower than in wheat straw and even below the detection limit

in casts (Wolfarth et al. 2011). Beyond that, earthworms secrete mucus and coelomic fluid through dorsal pores in their body wall, produce casts, and create middens. Each of these earthworm products contains highly bioavailable substances that increase microbial activity (Brown 1995) and potentially promote microbial mycotoxin degradation. The promotion of these soil animals, for example through reduced or no-tillage where soil conditions permit, the reduction of soil compaction to a necessary level, and the demand-oriented application of agrochemicals, can hence make a significant contribution to reducing mycotoxin contamination of crop residues.

## Conclusion

In maize cultivation, the earthworm species *L. terrestris* represents a key species within the soil fauna community for a species-specific bioregulation of *Fusarium* species. *L. terrestris* can shift the ecosystem service/disservice balance in arable systems in both directions, depending on *Fusarium* species involved in the infestation. Since *L. terrestris* does not generally contribute to suppressing all *Fusarium* species relevant in maize cultivation, management decisions have to be made site-specifically and depending on the respective infestation. However, in this context, it should be taken into account that ploughing, as an often-recommended preventive measure, is not an all-round solution. Although it has been proven that incorporation of crop residues into the soil can reduce the frequency of *F. culmorum* and *F. graminearum*, it increases that of *F. verticillioides* (Pfordt et al. 2020).

The potential of *L. terrestris* to reduce mycotoxin concentrations seems to be independent of the crop and includes DON, 3-AcDON, and ZEN. Agricultural management that considers the needs and habitat requirements of earthworms can thus lead to a synergy in which the interaction of anthropogenic top-down effects (agricultural management) and natural bottom-up effects (bioregulation by earthworms) contributes to sustainable agricultural production on healthy and fertile soils. By accelerating toxin degradation in the mulch layer, *L. terrestris* has the potential to shift the ecosystem service/disservice balance towards services (toxin degradation) in reduced tillage systems and to keep soils healthy and productive in the long run.

**Supplementary Information** The online version contains supplementary material available at <https://doi.org/10.1007/s11104-021-04882-4>.

**Acknowledgements** We thank Christiane Münter and Dr. Deborah Linsler (University of Göttingen) for their support in planning and conducting the field experiments at ‘Garte Süd’, Dr. Siegfried Schittenhelm (Institute for Crop and Soil Science, JKI) for providing the opportunity to use the colour analyses software to determine changes in soil surface cover, Dr. Detlef Schenke for mycotoxin analysis, and PD Dr. Jan Thiele and Dr. Katrin Ronnenberg for their statistical advice. The excellent technical assistance of Sabine El Sayed, Marion Krause, Karin Zinn, Ina Stachewicz-Voigt, Sabine Peickert, Jürgen Liersch, Anke Führer, Peter Braunisch, Ole Siebeneicher and Lena Wöhl is gratefully acknowledged.

**Authors' contributions** All authors contributed to the study conception, experimental design, data collection, and analysis. All authors read and approved the final manuscript.

**Funding** Open Access funding enabled and organized by Projekt DEAL. The present study was conducted as part of the SoilMan project (grant number 01LC1620), which was funded through the 2015–16 BiodivERsA COFUND call for research proposals, with the national funders Federal Ministry of Education and Research (BMBF), The French National Research Agency (ANR), The Swedish Research Council for Environment, Agricultural Sciences and Spatial Planning (FORMAS), Ministry of Economy and Competitiveness (MINECO), Executive Agency for Higher Education, Research, Development and Innovation Funding (UEFISCDI) and Estonian Research Council (ETAG).

**Availability of data and material** The datasets generated and analysed during the current study are available from the corresponding author on reasonable request.

**Code availability** Not applicable.

#### Declarations

**Conflict of interest** The authors have no conflicts of interest to declare that are relevant to the content of this article.

**Consent to participate** Not applicable

**Consent for publication** Not applicable.

**Open Access** This article is licensed under a Creative Commons Attribution 4.0 International License, which permits use, sharing, adaptation, distribution and reproduction in any medium or format, as long as you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons licence, and indicate if changes were made. The images or other third party material in this article are included in the article's Creative

Commons licence, unless indicated otherwise in a credit line to the material. If material is not included in the article's Creative Commons licence and your intended use is not permitted by statutory regulation or exceeds the permitted use, you will need to obtain permission directly from the copyright holder. To view a copy of this licence, visit <http://creativecommons.org/licenses/by/4.0/>.

#### References

- Aguín O, Cao A, Pintos C, Santiago R, Mansilla P, Butrón A (2014) Occurrence of *Fusarium* species in maize kernels grown in northwestern Spain. *Plant Pathol* 63:946–951. <https://doi.org/10.1111/ppa.12151>
- Bacon CW, Hinton DM, Mitchell TR (2017) Is quorum signalling by mycotoxins a new risk-mitigating strategy for bacterial biocontrol of *Fusarium verticillioides* and other endophytic fungal species? *J Agric Food Chem* 65(33):7071–7080. <https://doi.org/10.1021/acs.jafc.6b03861>
- Bates D, Maechler M, Bolker B, Walker S (2015) Fitting linear mixed effects models using lme4. *J Stat Softw* 67(1):1–48. <https://doi.org/10.18637/jss.v067.i01>
- Belizán MME, de los Gomez AA, Terán Baptista ZP, Jimenez CM, del Sánchez Matías HM, CAN C, Sampietro DA (2019) Influence of water activity and temperature on growth and production of trichothecenes by *Fusarium graminearum sensu stricto* and related species in maize grains. *Int J Food Microbiol* 305:108242. <https://doi.org/10.1016/j.ijfoodmicro.2019.108242>
- Binet F, Fayolle L, Pussard M (1998) Significance of earthworms in stimulating soil microbial activity. *Biol Fertil Soils* 27(1): 79–84. <https://doi.org/10.1007/s003740050403>
- Bonkowski M, Griffiths BS, Ritz K (2000) Food preferences of earthworms for soil fungi. *Pedobiologia* 44:666–676. [https://doi.org/10.1078/S0031-4056\(04\)70080-3](https://doi.org/10.1078/S0031-4056(04)70080-3)
- Bottalico A (1998) *Fusarium* diseases of cereals: species complex and related mycotoxin profiles in Europe. *J Plant Pathol* 80(2):85–103. <https://doi.org/10.4454/jpp.v80i2.807>
- Bottalico A, Perrone G (2002) Toxicogenic *Fusarium* species and mycotoxins associated with head blight in small-grain cereals in Europe. *Eur J Plant Pathol* 108:611–624. <https://doi.org/10.1023/A:1020635214971>
- Brandfäss C, Karlovsky P (2008) Upscaled CTAB-based DNA extraction and real-time PCR assays for *Fusarium culmorum* and *F. graminearum* DNA in plant material with reduced sampling error. *Int J Mol Sci* 9:2306–2321. <https://doi.org/10.3390/ijms9112306>
- Brennan JM, Fagan B, van Maanen A, Cooke BM, Doohan FM (2003) Studies on *in vitro* growth and pathogenicity of European *Fusarium* fungi. *Eur J Plant Pathol* 109(6):577–587. <https://doi.org/10.1023/A:1024712415326>
- Briónes M, Schmidt O (2017) Conventional tillage decreases the abundance and biomass of earthworms and alters their community structure in a global meta-analysis. *Glob Chang Biol* 23(10):4396–4419. <https://doi.org/10.1111/gcb.13744>

- Brown GG (1995) How do earthworms affect microfloral and faunal community diversity? *Plant Soil* 170:209–231. [https://doi.org/10.1007/978-94-011-0479-1\\_22](https://doi.org/10.1007/978-94-011-0479-1_22)
- Brown GG, Barois I, Lavelle P (2000) Regulation of soil organic matter dynamics and microbial activity in the drilosphere and the role of interactions with other edaphic functional domains. *Eur J Soil Biol* 36(3-4):177–198. [https://doi.org/10.1016/S1164-5563\(00\)01062-1](https://doi.org/10.1016/S1164-5563(00)01062-1)
- Champeil A, Doré T, Fourbet J-F (2004) *Fusarium* head blight: epidemiological origin of the effects of cultural practices on head blight attacks and the production of mycotoxins by *Fusarium* in wheat grains. *Plant Sci* 166(6):1389–1415. <https://doi.org/10.1016/j.plantsci.2004.02.004>
- Claassen R, Bowman M, McFadden J, Smith D, Wallander S (2018) Tillage intensity and conservation cropping in the United States. *Econ inform bull* 197, U.S. Department of Agriculture, Economic Research Service. <https://www.ers.usda.gov/webdocs/publications/90201/eib-197.pdf?v=5955.8>. Accessed 19 Feb 2021
- Cook RJ, Christensen AA (1976) Growth of cereal root-rot fungi as affected by temperature-water potential interactions. *Phytopathology* 66:193–197. <https://doi.org/10.1094/Phyto-66-193>
- Crawley MJ (2007) *The R book*. Wiley, Chichester. <https://doi.org/10.1002/9780470515075>
- Czembor E, Stepień Ł, Waśkiewicz A (2015) Effect of environmental factors on *Fusarium* species and associated mycotoxins in maize grain grown in Poland. *PLoS One* 10(7):e0133644. <https://doi.org/10.1371/journal.pone.0133644>
- Dill-Macky R, Jones RK (2000) The effect of previous crop residues and tillage on *Fusarium* head blight of wheat. *Plant Dis* 84(1):71–76. <https://doi.org/10.1094/PDIS.2000.84.1.71>
- EC (European Commission) (2006a) Commission Regulation (EC) No 1881/2006 of 19 December 2006 setting maximum levels for certain contaminants in foodstuffs. *Off J Eur Union L* 364(20.12.2006):5–24
- EC (European Commission) (2006b) Commission Recommendation of 17 August 2006 on the presence of deoxynivalenol, zearalenone, ochratoxin A, T-2 and HT-2 and fumonisins in products intended for animal feeding (2006/576/EC). *Off J Eur Union L* 229(23.08.2006):7–9
- EC (European Commission) (2006c) Commission Recommendation of 17 August 2006 on the prevention and reduction of *Fusarium* toxins in cereals and cereal products (2006/583/EC). *Off J Eur Union L* 234(29.08.2006):35–40
- EC (European Commission) (2007) Commission Regulation (EC) No 1126/2007 of 28 September 2007 amending Regulation (EC) No 1881/2006 setting maximum levels for certain contaminants in foodstuffs as regards *Fusarium* toxins in maize and maize products. *Off J Eur Union L* 255(29.09.2007):14–17
- EC (European Commission) (2018) EU agricultural outlook for markets and income, 2018–2030. *Eur. Comm. DG Agriculture and Rural Development*, Brussels
- Elmholt S (2008) Mycotoxins in the soil environment. In: Karlovsky P. (ed.): secondary metabolites in soil ecology. *Soil Biol.* 14:167–203, Springer, Berlin, Heidelberg. [https://doi.org/10.1007/978-3-540-74543-3\\_9](https://doi.org/10.1007/978-3-540-74543-3_9)
- Fargione JE, Cooper TR, Flaspohler DJ, Hill J, Lehman C, McCoy T, McLeod S, Nelson EJ, Oberhauser KS, Tilman D (2009) Bioenergy and wildlife: threats and opportunities for grassland conservation. *Bioscience* 59:767–777. <https://doi.org/10.1525/bio.2009.59.9.8>
- Ferrigo D, Raiola A, Causin R (2016) *Fusarium* toxins in cereals: occurrence, legislation, factors promoting the appearance and their management. *Molecules* 21(5):627. <https://doi.org/10.3390/molecules21050627>
- Fox J, Weisberg S (2019) *An R companion to applied regression*, 3rd edn. Sage, Thousand Oaks, CA
- Fründ H-C, Butt K, Capowicz Y, Eisenhauer N, Emmerling C, Ernst G, Potthoff M, Schädler M, Schrader S (2010) Using earthworms as model organisms in the laboratory: recommendations for experimental implementations. *Pedobiologia* 53:119–125. <https://doi.org/10.1016/j.pedobi.2009.07.002>
- Galeana-Sánchez E, Sánchez-Rangel D, de la Torre-Hernández ME, Nájera-Martínez M, Ramos-Villegas P, Plasencia J (2017) Fumonisin B1 produced in planta by *Fusarium verticillioides* is associated with inhibition of maize  $\beta$ -1,3-glucanase activity and increased aggressiveness. *Physiol Mol Plant Pathol* 100:75–83. <https://doi.org/10.1016/j.pmp.2017.07.003>
- Gautam P, Dill-Macky R (2012) Free water can leach mycotoxins from *Fusarium*-infected wheat heads. *J Phytopathol* 160(9):484–490. <https://doi.org/10.1111/j.1439-0434.2012.01928.x>
- Goncharov AA, Glebova AA, Tiunov AV (2020) Trophic interactions between *Fusarium* species and soil fauna: a meta-analysis of experimental studies. *Appl Soil Ecol* 145:103302. <https://doi.org/10.1016/j.apsoil.2019.06.005>
- Goswami RS, Kistler HC (2004) Heading for disaster: *Fusarium graminearum* on cereal crops. *Mol Plant Pathol* 5:515–525. <https://doi.org/10.1111/j.1364-3703.2004.00252.x>
- Hogg AC, Johnston RH, Dyer AT (2007) Applying real-time quantitative PCR to *Fusarium* crown rot of wheat. *Plant Dis* 91:1021–1028. <https://doi.org/10.1094/PDIS-91-8-1021>
- Hudec K, Muchova D (2010) Influence of temperature and species origin on *Fusarium spp.* and *Microdochium nivale* pathogenicity to wheat seedlings. *Plant Prot Sci* 46(2):59–65. <https://doi.org/10.17221/12/2009-PPS>
- Jacobs A, Rauber R, Ludwig B (2009) Impacts of reduced tillage on carbon and nitrogen storage of two Haplic Luvisols after 40 years. *Soil Tillage Res* 102:158–164. <https://doi.org/10.1016/j.still.2008.08.012>
- Joint FAO/WHO Codex Alimentarius Commission (2017) Codex Alimentarius: code of practice for the prevention and reduction of mycotoxin contamination in cereals (CXC 51–2003). Joint FAO/WHO Food Standards Programme. [http://www.fao.org/fao-who-codexalimentarius/sh-proxy/en/?lnk=1&url=https%253A%252F%252Fworkspace.fao.org%252Fsites%252Fcodex%252Fstandards%252FCXC%252B51-2003%252FCXC\\_051e.pdf](http://www.fao.org/fao-who-codexalimentarius/sh-proxy/en/?lnk=1&url=https%253A%252F%252Fworkspace.fao.org%252Fsites%252Fcodex%252Fstandards%252FCXC%252B51-2003%252FCXC_051e.pdf). Accessed 19 Feb 2021
- Kassam A, Friedrich T, Shaxson F, Pretty J (2009) The spread of conservation agriculture: justification, sustainability and uptake. *Int J Agric Sustain* 7(4):292–320. <https://doi.org/10.3763/ijas.2009.0477>
- Kolpin DW, Schenzel J, Meyer MT, Phillips PJ, Hubbard LE, Scott T-M, Bucheli TD (2014) Mycotoxins: diffuse and point source contributions of natural contaminants of emerging concern to streams. *Sci Total Environ* 470-471:669–676. <https://doi.org/10.1016/j.scitotenv.2013.09.062>

- Lavelle P (1988) Earthworm activities and the soil system. *Biol Fertil Soils* 6(3):237–251. <https://doi.org/10.1007/BF00260820>
- Leplat J, Friberg H, Abid M, Steinberg C (2013) Survival of *Fusarium graminearum*, the causal agent of *Fusarium* head blight. A review. *Agron Sustain Dev* 33:97–111. <https://doi.org/10.1007/s13593-012-0098-5>
- Li YG, Jiang D, Xu LK, Zhang SQ, Ji PS, Pan HY, Jiang BW, Shen ZB (2019) Evaluation of diversity and resistance of maize varieties to *Fusarium* spp. causing ear rot in maize under conditions of natural infection. *Czech J. genet. Plant Breed* 55:131–137. <https://doi.org/10.17221/81/2018-CJGPB>
- Manstretta V, Rossi V (2016) Effects of temperature and moisture on development of *Fusarium graminearum* perithecia in maize stalk residues. *Appl Environ Microbiol* 82:184–191. <https://doi.org/10.1128/AEM.02436-15>
- Marín S, Sanchís V, Teixido A, Saenz R, Ramos AJ, Vinas I, Magan N (1996) Water and temperature relations and microconidial germination of *Fusarium moniliforme* and *Fusarium proliferatum* from maize. *Can J Microbiol* 42(10):1045–1050. <https://doi.org/10.1139/m96-134>
- Masiello M, Somma S, Ghionna V, Logrieco AF, Moretti A (2019) In vitro and in field response of different fungicides against *Aspergillus flavus* and *Fusarium* species causing ear rot disease of maize. *Toxins* 11(1):11. <https://doi.org/10.3390/toxins11010011>
- Meier U (2018) Growth stages of mono- and dicotyledonous plants. BBCH Monograph. Julius Kühn-Institute (JKI), Quedlinburg. <https://doi.org/10.5073/20180906-074619>
- Meyer-Wolfarth F, Schrader S, Oldenburg E, Weinert J, Brunotte J (2017) Biocontrol of the toxigenic plant pathogen *Fusarium culmorum* by soil fauna in an agroecosystem. *Mycotoxin Res* 33:237–244. <https://doi.org/10.1007/s12550-017-0282-1>
- Morel R (1996) Cultivated soils; Les sols cultivés – Technique et documentation, 2nd edn. Lavoisier, Paris
- Müller MEH, Urban K, Köppen R, Siegel D, Korn U, Koch M (2014) Mycotoxins as antagonistic or supporting agents in the interaction between phytopathogenic *Fusarium* and *Alternaria* fungi. *World Mycotoxin J* 8(3):311–321. <https://doi.org/10.3920/WMJ2014.1747>
- Munkvold GP (2017) *Fusarium* species and their associated mycotoxins. In: Moretti A, Susca A (eds) *Mycotoxigenic Fungi: methods and protocols, methods in molecular biology* 1542. Springer, New York, pp 51–106. [https://doi.org/10.1007/978-1-4939-6707-0\\_4](https://doi.org/10.1007/978-1-4939-6707-0_4)
- Oksanen J, Blanchet FG, Friendly M, Kindt R, Legendre P, McGlenn D, Minchin PR, O'Hara RB, Simpson GL, Solymos P, Stevens MHH, Szoecs E, Wagner H (2019) *Vegan: community ecology package*. R package version 2: 5–5
- Oldenburg E, Ellner F (2015) Distribution of disease symptoms and mycotoxins in maize ears infected by *Fusarium culmorum* and *Fusarium graminearum*. *Mycotoxin Res* 31: 117–126. <https://doi.org/10.1007/s12550-015-0222-x>
- Oldenburg E, Kramer S, Schrader S, Weinert J (2008) Impact of the earthworm *Lumbricus terrestris* on the degradation of *Fusarium*-infected and deoxynivalenol-contaminated wheat straw. *Soil Biol Biochem* 40(12):3049–3053. <https://doi.org/10.1016/j.soilbio.2008.09.004>
- Oldenburg E, Höppner F, Ellner F (2018) *Fusarium verticillioides*-Infektionen und Fumonisin-Kontaminationen beim Mais. *J Kult* 70(5):166–167
- Ortiz CS, Richards C, Terry A, Parra J, Shim WB (2015) Genetic variability and geographical distribution of mycotoxigenic *Fusarium verticillioides* strains isolated from maize fields in Texas. *The plant Pathol. J.* 31(3):203–211. <https://doi.org/10.5423/PPJ.OA.02.2015.0020>
- Pavlik P, Vlckova V, Machar I (2019) Changes to land area used for grain maize production in Central Europe due to predicted climate change. *Int J Agron* 2019(3–4):1–9. <https://doi.org/10.1155/2019/9168285>
- Pelosi C, Pey B, Hedde M, Caro G, Capowicz Y, Guernion M, Peigné J, Piron D, Bertrand M, Cluzeau D (2014) Reducing tillage in cultivated fields increases earthworm functional diversity. *Appl Soil Ecol* 83:79–87. <https://doi.org/10.1016/j.apsoil.2013.10.005>
- Pereyra SA, Dill-Macky R (2008) Colonization of the residues of diverse plant species by *Gibberella zeae* and their contribution to *Fusarium* head blight inoculum. *Plant Dis* 92(5):800–807. <https://doi.org/10.1094/PDIS-92-5-0800>
- Pereyra SA, Sims AL, Dill-Macky R (2004) Survival and inoculum production of *Gibberella zeae* in wheat residue. *Plant Dis* 88(7):724–730. <https://doi.org/10.1094/PDIS.2004.88.7.724>
- Perincherry L, Lalak-Kańczugowska J, Stepien Ł (2019) *Fusarium*-produced mycotoxins in plant-pathogen interactions. *Toxins* 11:664. <https://doi.org/10.3390/toxins11110664>
- Pettitt TR, Parry DW, Pollea RW (1996) Effect of temperature on the incidence of nodal foot rot symptoms in winter wheat crops in England and Wales caused by *Fusarium culmorum* and *Microdochium nivale*. *Agric For Meteorol* 79:233–242. [https://doi.org/10.1016/0168-1923\(95\)02281-3](https://doi.org/10.1016/0168-1923(95)02281-3)
- Pfordt A, Ramos Romero L, Schiwiek S, Karlovsky P, von Tiedemann A (2020) Impact of environmental conditions and agronomic practices on the prevalence of *Fusarium* species associated with ear- and stalk rot in maize. *Pathogens* 9:236. <https://doi.org/10.3390/pathogens9030236>
- Proctor RH, Hohn TM, McCormick SP (1995) Reduced virulence of *Gibberella zeae* caused by disruption of a trichothecene toxin biosynthetic gene. *Mol Plant-Microbe Interact* 8(4): 593–601. <https://doi.org/10.1094/mpmi-8-0593>
- R Core Team (2018) R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna
- Ryu D, Bulleman LB (1999) Effect of cycling temperatures on the production of deoxynivalenol and zearalenone by *Fusarium graminearum* NRRL 5883. *J Food Prot* 62(12): 1451–1455. <https://doi.org/10.4315/0362-028x-62.12.1451>
- Schrader S, Kramer S, Oldenburg E, Weinert J (2009) Uptake of deoxynivalenol by earthworms from *Fusarium*-infected wheat straw. *Mycotoxin Res* 25:53–58. <https://doi.org/10.1007/s12550-009-0007-1>
- Schrader S, Wolfarth F, Oldenburg E (2013) Biological control of soil-borne phytopathogenic fungi and their mycotoxins by soil fauna. *Bull UASMV Agric* 70(2):291–298
- Schrader S, van Capelle C, Meyer-Wolfarth F (2020) Regenwürmer als Partner bei der Bodennutzung. *Biol unserer Zeit* 3(50):192–198. <https://doi.org/10.1002/biuz.202010706>
- Snijders CHA (1995) Breeding for resistance to *Fusarium* in wheat and maize. In: Miller JD, Trenholm HL (eds)

- Mycotoxins in grain: compounds other than Aflatoxin. Eagan Press, St. Paul, MN, p 3758
- Swift MJ, Heal OW, Anderson JM (1979) Decomposition in terrestrial ecosystems, studies in ecology Vol. 5. Blackwell, Oxford
- Tissier ML, Handrich Y, Dallongeville O, Robin J-P, Habold C (2016) Diets derived from maize monoculture cause maternal infanticides in the endangered European hamster due to a vitamin B3 deficiency. *Proc R Soc B* 284(1847):20162168. <https://doi.org/10.1098/rspb.2016.2168>
- Torres MR, Ramos AJ, Soler J, Sanchis V, Marín S (2003) SEM study of water activity and temperature effects on the initial growth of *Aspergillus ochraceus*, *Alternaria alternata* and *Fusarium verticillioides* on maize grain. *Int J Food Microbiol* 81(3):185–193. [https://doi.org/10.1016/S0168-1605\(02\)00226-X](https://doi.org/10.1016/S0168-1605(02)00226-X)
- van Capelle C, Schrader S, Brunotte J (2012) Tillage-induced changes in the functional diversity of soil biota – a review with a focus on German data. *Eur J Soil Biol* 50:165–181. <https://doi.org/10.1016/j.ejsobi.2012.02.005>
- van der Lee T, Zhang H, van Diepeningen A, Waalwijk C (2015) Biogeography of *Fusarium graminearum* species complex and chemotypes: a review. *Food Addit Contam Part A Chem Anal Control Expo Risk Assess* 32(4):453–460. <https://doi.org/10.1080/19440049.2014.984244>
- Vanhoutte I, Audenaert K, de Gelder L (2016) Biodegradation of mycotoxins: tales from known and unexplored worlds. *Front Microbiol* 7:561. <https://doi.org/10.3389/fmicb.2016.00561>
- Velluti A, Marín S, Bettucci L, Ramos AJ, Sanchis V (2000) The effect of fungal competition on colonization of maize grain by *Fusarium moniliforme*, *F. proliferatum* and *F. graminearum* and on fumonisin B1 and zearalenone formation. *Int. J. Food Microbiol* 59(1–2):59–66. [https://doi.org/10.1016/s0168-1605\(00\)00289-0](https://doi.org/10.1016/s0168-1605(00)00289-0)
- Venables WN, Ripley BD (2002) *Modern applied statistics with S*, 4th edn. Springer, New York
- Venkatesh N, Keller NP (2019) Mycotoxins in conversation with bacteria and fungi. *Front Microbiol* 10:403. <https://doi.org/10.3389/fmicb.2019.00403>
- Vogelgsang S, Hecker A, Musa T, Dom B, Forrer HR (2011) On-farm experiments over 5 years in a grain maize/winter wheat rotation: effect of maize residue treatments on *Fusarium graminearum* infection and deoxynivalenol contamination in wheat. *Mycotoxin Res* 27:81–96. <https://doi.org/10.1007/s12550-010-0079-y>
- Waalwijk C, Kastelein P, de Vries I, Kerényi Z, van der Lee T, Hesselink T, Köhl J, Kema G (2003) Major changes in *Fusarium spp.* in wheat in the Netherlands. *Eur J Plant Pathol* 109:743–754. <https://doi.org/10.1023/A:1026086510156>
- Wachowska U, Packa D, Wiwart M (2017) Microbial inhibition of *Fusarium* pathogens and biological modification of trichothecenes in cereal grains. *Toxins* 9:408. <https://doi.org/10.3390/toxins9120408>
- Wang H, Guo Q, Li X, Li X, Yu Z, Li X, Yang T, Su Z, Zhang H, Zhang C (2020) Effects of long-term no-tillage with different straw mulching frequencies on soil microbial community and the abundances of two soil-borne pathogens. *Appl Soil Ecol* 148:103488. <https://doi.org/10.1016/j.apsoil.2019.103488>
- Wegulo SN, Baenziger PS, Hernandez Nopsa J, Bockus WW, Hallen-Adams H (2015) Management of *Fusarium* head blight of wheat and barley. *Crop Prot* 73:100–107. <https://doi.org/10.1016/j.cropro.2015.02.025>
- Wei T, Simko V (2017) R package “corrplot”: Visualization of a correlation matrix (Version 0.84). Available from <https://github.com/taiyun/corrplot>
- Wenda-Piesik A, Lemańczyk G, Twarużek M, Błajet-Kosicka A, Kazek M, Grajewski J (2017) *Fusarium* head blight incidence and detection of *Fusarium* toxins in wheat in relation to agronomic factors. *Eur J Plant Pathol* 149:515–531. <https://doi.org/10.1007/s10658-017-1200-2>
- Wickham H (2016) *ggplot2: elegant graphics for data analysis*. Springer, New York Available from <https://ggplot2.tidyverse.org>
- Wolfarth F, Schrader S, Oldenburg E, Weinert J, Brunotte J (2011) Earthworms promote the reduction of *Fusarium* biomass and deoxynivalenol content in wheat straw under field conditions. *Soil Biol Biochem* 43:1858–1865. <https://doi.org/10.1016/j.soilbio.2011.05.002>
- Wolfarth F, Schrader S, Oldenburg E, Brunotte J (2016) Mycotoxin contamination and its regulation by the earthworm species *Lumbricus terrestris* in presence of other soil fauna in an agroecosystem. *Plant Soil* 402:331–342. <https://doi.org/10.1007/s11104-015-2772-2>
- Xu X-M, Monger W, Ritieni A, Nicholson P (2007) Effect of temperature and duration of wetness during initial infection periods on disease development, fungal biomass and mycotoxin concentrations on wheat inoculated with single, or combinations of, *Fusarium* species. *Plant Pathol* 56:943–956. <https://doi.org/10.1111/j.1365-3059.2007.01650.x>
- Zhang XF, Zou CJ, Cui LN, Li X, Yang XR, Luo HH (2012) Identification of pathogen causing maize ear rot and inoculation technique in Southwest China. *Southwest China J Agric Sci* 25(6):2078–2082
- Zhang Y, He J, Jia L-J, Yuan T-L, Zhang D, Guo Y, Wang Y, Tang W-H (2016) Cellular tracking and gene profiling of *Fusarium graminearum* during maize stalk rot disease development elucidates its strategies in confronting phosphorus limitation in the host apoplast. *PLoS Pathog* 12(3):e1005485. <https://doi.org/10.1371/journal.ppat.1005485>

**Publisher's note** Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.