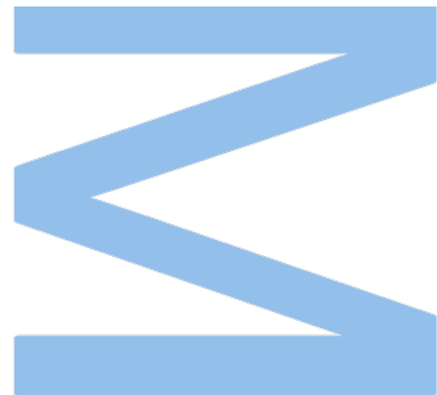
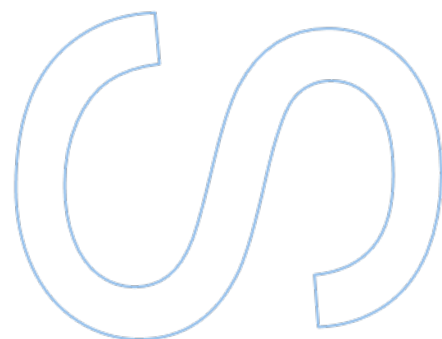
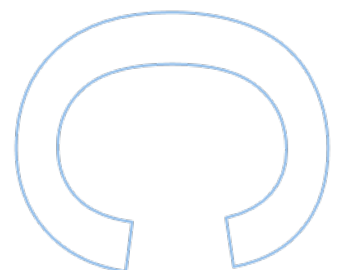
Genetic variation in wheat for fusarium foot rot and its biocontrol by *Clonostachys rosea*







Rosa Margarida Nogueira Ricardo
MSc in Applications in Biotechnology and Synthetic Biology
Department of Biology
Faculty of Sciences of the University of Porto
2024

Genetic variation in wheat for fusarium foot rot and its biocontrol by *Clonostachys rosea*

Rosa Margarida Nogueira Ricardo
MSc in Applications in Biotechnology and Synthetic Biology
Department of Biology
2024

Supervisor
Magnus Karlsson, Professor, Swedish University of Agricultural Sciences, Uppsala, Sweden
Co-supervisor
Susana Maria Pinto de Carvalho, Assistant Professor, Faculty of Sciences of the University of Porto

Dedicated to Sir David Attenborough, the voice I heard in my head every time I faced the most daunting research question: "Here we see a brave student attempting to navigate the treacherous waters of academia. Will she survive? Only time will tell."

Acknowledgments

I am going to start this section by thanking my supervisor Prof. Magnus Karlsson for all the help with writing this thesis, support and for allowing me to be part of the APP group and welcome me to SLU, Sweden. I also want to thank my co-supervisor Prof. Susana Carvalho for all the help with writing and critical reading of this thesis and for being in my corner since my bachelor's.

I want to give the biggest thank you and virtual hug to the PhD student Sidhant "Sid" Chaudhary for not only being my partner in crime in the lab but also for being my lighthouse during all the thesis process, without you I could not have performed this work the way I did, I hold the biggest gratitude to you.

To everyone in the APP group, I want to thank you for always being willing to help me, for accepting me as part of the team, and for always being so nice.

I also want to thank everyone who helped in the lab, especially Sofia, and for showing me what Bardcore is ;).

To all my "Sweden" friends, Alessandra, Berea, Eliverta, Eva, Francesca, Harkishan, Kat, Marvin, Mohamed, Sara, Sid, Susmita and Vilda thank you for all the laughs, for giving me support, for checking me into the reality of life (there are a few that this is for ahah). And mainly for making my life so colorful in dark Sweden. You guys will forever hold a little piece of my memories and my heart <3.

To my beautiful www.betos.com group, thank you for the visit in Sweden, both physically and virtually with lots of video calls. Thank you for the support, for all the times you made me feel validated with my feelings, and for the help when the anxiety of life was getting to me. To Ana Catarina, Carapato, Catarina Alexandra, Pedrosa and Surfista a big thank you, que deus vos pague <3.

Para os meus pais, António e Carla que tem a maior paciência comigo e que são a maior e melhor claque do mundo, o maior obrigado de todos por sempre me deixarem estudar não só o que eu queria, mas também por estarem lá para mim mesmo nas decisões mais difíceis <3. E também um beijinho e obrigado ao meu irmão Miguel por me atuar e apoiar hehe.

To anyone and everyone that is in my life who is always supporting me, thank you and see you later alligator :).

Resumo

A Revolução Verde, uma era crucial na agricultura, aumentou significativamente o rendimento das culturas através de vários meios, como o melhoramento das plantas, o aumento da utilização de fertilizantes e pesticidas e a mecanização. No entanto, apesar dos seus contributos para a segurança alimentar e o desenvolvimento económico, as práticas agrícolas modernas enfrentam desafios colocados pelas alterações ambientais e pelas doenças das culturas. Os pesticidas, embora eficazes, suscitam preocupações quanto à sustentabilidade a longo prazo e aos impactos ambientais. A Gestão Integrada das Doenças (IDM) e o controlo biológico oferecem alternativas promissoras, ao utilizarem abordagens ecológicas para combater as doenças das culturas. No entanto, pouco se sabe sobre a forma como a variação genética das plantas influencia a eficácia dos agentes de controlo biológico (BCAs). Este estudo tem como objetivo investigar a diversidade genética entre genótipos de trigo de inverno no que diz respeito à suscetibilidade à podridão de pé de fusarium (FFR) e à sua resposta ao agente de biocontrolo fúngico *Clonostachys rosea* durante a infeção por FFR. Hipotizou-se diferenças na resistência a FFR e compatibilidade com *C. rosea* entre genótipos de trigo de inverno num bioensaio, onde a severidade da doença e características associadas ao comprimento da raiz, comprimento do rebento e comprimento da planta sob dois tratamentos: *F. graminearum* (Fg) e Fg com *C. rosea* (FgCr) foram medidos. Através de análises estatísticas e estudos de correlação, diferenças significativas mostram a eficácia de *C. rosea* na redução da severidade da doença e na melhoria do crescimento das plantas, destacando o seu potencial para a gestão sustentável de doenças. Além disso, foram observadas correlações entre a severidade da doença, o crescimento das plantas e a eficácia do biocontrolo, fornecendo informações sobre as interações entre plantas e agentes patogénicos. Adicionalmente, uma matriz de marcadores de 20K, foi utilizada para efetuar um estudo de associação de todo o genoma. Os estudos de associação do genoma identificaram marcadores genéticos significativos associados à pontualidade da doença, comprimento do rebento e comprimento da raiz, mostrando diferentes localizações no genoma. Além disso, o bioensaio foi realizado utilizando *Trichoderma harzianum* como BCA, onde o efeito do tratamento não foi significativo para nenhuma das características estudadas.

Palavras-chave: Biocontrolo, *Clonostachys rosea*, *Fusarium graminearum*, fusarium foot root, Variação genética, Suscetibilidade das plantas, Estudo de associação do genoma (GWAS), trigo

Abstract

The Green Revolution, a pivotal era in agriculture, significantly enhanced crop yields through various means such as plant breeding, increased use of fertilizers and pesticides, and mechanization. However, despite its contributions to food security and economic development, modern agricultural practices face relevant challenges posed by climate changes and globalized markets that have exacerbated the incidence of crop diseases. Synthetic chemical pesticides, while effective, raise increasing concerns about long-term sustainability and environmental impacts. Integrated Disease Management (IDM) and biological control offer promising alternatives by employing eco-friendly approaches to combat crop diseases. Nevertheless, little is understood about how genetic variation in plants influences how effective biological control agents (BCAs) are. This study aims to investigate the genetic diversity among winter wheat genotypes concerning fusarium foot rot (FFR) susceptibility and their response to the fungal biocontrol agent *Clonostachys rosea* during FFR infection. Hypothesizing differences in resistance to FFR and compatibility with *C. rosea* among winter wheat genotypes in a bioassay, where disease severity, and traits associated with root length, shoot length, and plant length under two treatments: *F. graminearum* (Fg) and Fg with *C. rosea* (FgCr) were measured. Through statistical analyses and correlation studies, significant differences show *C. rosea* effectiveness in reducing disease severity and improving plant growth, highlighting its potential for sustainable disease management. Furthermore, correlations between disease severity, plant growth, and biocontrol efficacy were observed, providing insights into plant-pathogen interactions. Additionally, using a 20K marker array, a genome-wide association study was also performed. Genome-wide association studies identified significant genetic markers associated with disease score, shoot length, and root length, showing different locations in the genome. Additionally, the bioassay was performed using *Trichoderma harzianum* as a BCA, where the effect of treatment was not significant for any of the traits.

Keywords: Biocontrol, *Clonostachys rosea*, *Fusarium graminearum*, fusarium foot root, Genetic variation, Plant susceptibility, Genome-wide association study (GWAS), wheat

Table of Contents

List of Tables and Figures vi

List of abbreviations viii

Introduction 10

1. State of the art 15
   1.1. Biological control 15
   1.2. *Fusarium* species 21
2. Aim and objectives 25
3. Materials and methods 26
   3.1. Plant material 26
   3.2. Fungal strains preparation and application 26
   3.3. Bioassay setup 27
   3.4. Statistical analysis 28
   3.5. Genome-wide association studies (GWAS) 29
4. Results 30
   4.1. Analysis of variance (ANOVA) of traits 30
   4.2. Correlations between treatments within each trait 32
   4.3. Correlation between plant growth and disease severity 33
   4.4. Pairwise contrasts for assessment of *C. rosea* efficacy 34
   4.5. Correlation between *C. rosea* efficacy and *F. graminearum* 35
   4.6. Genome-wide associations studies 36
   4.7. Analysis of results of *T. afroharzianum* 38
5. Discussion 40
6. Conclusions and future perspectives 44

References 45

Supplementary Material 62

List of Tables and Figures

Table 1 Results of two-way ANOVA test showing F-values and level of significance for traits. Significance indicator denoting the significance level of the p-value (* indicates $p < 0,05$, and absence of asterisk indicates non-significance).

Table 2 Two-way ANOVA test showing F-values and level of significance for traits. Significance indicator denoting the level of significance of the p-value (* indicates $p < 0,05$, and absence of asterisk indicates non-significance).

Figure 1 Important mechanisms of action for fungal antagonists' biological control of plant fungal diseases (adapted from Thambugala et al., 2020).

Figure 2 Life cycle of *Fusarium* spp. and the interactions between the microbiota and diseases related to it in cereals. *Fusarium* inoculum is represented by the red dots (Karlsson et al., 2021).

Figure 3 Distribution of BLUEs in treatments Fg and FgCr for the traits (a) root length, (b) shoot length, (c) plant length and (d) disease score. Dashed horizontal lines represent the arithmetic means of BLUEs in each treatment for each trait.

Figure 4 Correlation analysis between treatments FgCr and Fg in 190 winter wheat genotypes for the traits (a) root length, (b) shoot length, (c) plant length and (d) disease score using Pearson correlation coefficient (R). Each data point represents the BLUEs of genotypes across treatments.

Figure 5 Pearson's correlation (R) between disease score (0-4) and plant length (cm) in treatments FgCr and Fg. Each data point represents the BLUEs for plant length and disease score.

Figure 6 Pairwise comparisons between treatments FgCr and Fg for 190 winter wheat genotypes for trait (a) disease score, (b) root length, (c) shoot length and (d) plant length. Each data point represents a contrast estimate for a specific winter wheat genotype. The vertical lines extending from the points denote the 95% confidence intervals around the contrast estimates. Data points annotated with a red asterisk (*) are statistically significant ($p < 0.05$) and show a non-zero contrast. Note: Genotypes presented are not in the same order across the four plots.

Figure 7 Pearson's correlation between *C. rosea* efficacy and treatment *F. graminearum* (Fg) for the traits (a) root length, (b) shoot length, (c) plant length and (d) disease score. Each data point represents the inter treatment contrast (Fg – FgCr or FgCr – Fg) and BLUEs of genotypes in treatment Fg.

Figure 8 Manhattan plot and corresponding QQ plots showing the association of SNP markers with the FFR disease score in treatment Fg (a), treatment FgCr (b) and disease score contrast or biocontrol efficacy (c). The solid horizontal line shows the significance threshold of −log10(P) = 3.85.

Suplementary Figure 1. Residuals vs fitted, Normal Q-Q and Density plots for Model of bioassay with treatment FgCr (*Clonostachys rosea*) in each trait. (a) disease score, (b) shoot length, (c) plant length, (d) root length.

Suplementary Figure 2. Manhattan plot and corresponding QQ plots showing the association of SNP markers with the FFR plant length in treatment Fg (a), treatment FgCr (b) and disease score contrast or biocontrol efficacy (c). The solid horizontal line shows the significance threshold of −log10(P) = 3.85.

Suplementary Figure 3. Manhattan plot and corresponding QQ plots showing the association of SNP markers with the FFR shoot length in treatment Fg (a), treatment FgCr (b) and disease score contrast or biocontrol efficacy (c). The solid horizontal line shows the significance threshold of −log10(P) = 3.85.

Suplementary Figure 4. Manhattan plot and corresponding QQ plots showing the association of SNP markers with the FFR root length in treatment Fg (a), treatment FgCr (b) and disease score contrast or biocontrol efficacy (c). The solid horizontal line shows the significance threshold of −log10(P) = 3.85.

Suplementary Figure 5. Residuals vs fitted, Normal Q-Q and Density plots for Model of bioassay with treatment FgTa. (a) disease score, (b) shoot length, (c) plant length, (d) root length.

Supplementary table 1 List of the 190 genotypes used in this study for the *Clonostachys rosea* bioassay. Genotypes marked in bolt were used in the bioassay with treatment *Fusarium graminearum* and *Trichoderma afroharzianum* (FgTa).

Supplementary table 2 Summary of the significant SNPs marker identified which are associated as resistance indicator in GWAS analysis with 181 winter wheat genotypes.

List of abbreviations

BCAs Biological Control Agents

BLUEs Best Linear Unbiased Estimators

CFUs Colony Forming Units

CWDEs Cell Wall Degrading Enzymes

DON Deoxynivalenol

EC European Commission

EMMs Estimated Marginal Means

EU European Union

FCR Fusarium Crown Root

FFR Fusarium Foot Root

Fg Treatment with *Fusarium graminearum*

FgCr Treatment with *Fusarium graminearum* and seed coated with *Clonostachys rosea*

FGSC *Fusarium graminearum* Species Complex

FgTa Treatment with *Fusarium graminearum* and seed coated with *Trichoderma afroharzianum*

FHB Fusarium Head Blight

GAPIT Genome Association and Prediction Integrated Tool

GLM General linear model

GWAS Genome-Wide Association Study

IDM Integrated Disease Management

IPM Integrated Pest Management

ISR Induced Systematic Resistance

PDA Potato dextrose Agar

QTL Quantitative Trait Locus

R	Pearson correlation coefficient
SAR	Systemic Acquired Resistance
SNP	Single-nucleotide polymorphism
ZEN	Zearalenone

Introduction

The green revolution was primarily centered on improving crop yields using plant breeding, increased fertilizer and pesticide inputs, and enhanced mechanization, which helped improve food supplies, and triggered economic development supporting the growth of the human population (Armanda et al., 2019; van de Wouw et al., 2010; von der Goltz et al., 2020). Cereal production tripled post-green revolution with only a 30% increase in cultivated land (John & Babu, 2021; Pingali, 2012). By 2050, the global population is estimated to be close to 10 billion, and the world food supply may expand between 35% to 56% relative to 2010 to meet populational demands (van Dijk et al., 2021). However, environmental changes strongly affect agricultural production compromised by abiotic (predominantly drought, cold, salinity, and heat) and by biotic factors (i.e. namely diseases caused by fungi, bacteria, viruses, etc.) leading to severe crop losses worldwide (Chaloner et al., 2021; Jeyasri et al., 2021).

Pesticides are crucial to agricultural production because they can reduce product losses caused by pathogens and increase food quality and yield at a reasonable price (Fenik et al., 2011; Sharma et al., 2019). "Pesticide" refers to a broad category of substances that are either naturally occurring or artificially manufactured, such as chemical fungicides, herbicides, rodenticides, molluscicides, nematicides, and insecticides (Aktar et al., 2009; Pathak et al., 2022; Tudi et al., 2021). While the use of pesticides revolutionized crop protection and, consequently, crop production in the 20th century, their widespread use has raised questions about the long-term viability of creating pathogen resistance, and accumulation in soil, water, air, plant parts, and biota-producing hazards for humans, (Lamichhane et al., 2018; Nicolopoulou-Stamati et al., 2016). In spite of the negative effects of pesticides, pesticide usage has increased by around 50% from 1.2 Kg/ha in 1990 to 1.8 Kg/ha in 2020 with a total amount of active ingredients at 2.7 million tonnes (FAO, 2022). Additionally, some diseases have shown resistance to certain active compounds, and it is urgent to develop more sustainable and effective strategies for controlling crop diseases (Ali et al., 2022; Santos et al., 2023).

Over the lengthy history of agriculture, several strategies have been developed to manage plant-pathogen interactions and create a system that is favorable for host plant growth and development and unfavorable for pathogen establishment, reproduction, and propagation (He et al., 2021). Integrated disease management (IDM) is a cost-effective and sustainable approach that employs various methods simultaneously to discourage crop diseases while minimizing the application of chemical pesticides (Karlsson Green et al., 2020; National Academies of Sciences et al., 2023). Directive 2009/128/EC within the European Union (EU) specifically mandates that any person who uses pesticides during their professional activities

within the EU must adhere to the integrated pest management (IPM) principles (Barzman et al., 2015).

Biological control, or biocontrol, has been pointed out as a sustainable alternative to synthetic pesticides with a high potential of preventing losses due to diseases and offers a good balance between implementation costs and benefits over an extended period (López-Núñez et al., 2021). Biocontrol is defined as the exploitation of living agents (incl. viruses) to combat pestilential organisms (incl. pathogens, pests, and weeds) or their biological activity (Eilenberg et al., 2001; Pandit et al., 2022; Stenberg et al., 2021) Typically, fungal and bacterial strains isolated from the phyllosphere, endosphere, or rhizosphere are used as biological control agents (BCAs) (Thambugala et al., 2020). Natural, conservative, classical, and augmentation are types of biocontrol that can be categorized as either resident biocontrol species (no deliberate involvement of humans) or involvement of direct application of extra organisms, respectively. By improving their environment, conservation biocontrol indirectly increases the populations of already-existing natural enemies. Augmentative biological control involves the periodic releases of natural enemies—either native or exotic—into the habitat where pests occur, to temporarily control pest populations. Unlike classical biocontrol, augmentative biocontrol requires continual releases to maintain effectiveness (Stenberg et al., 2021).

The fungi *Trichoderma* spp. and *Clonostachys rosea* have been shown in numerous studies to be efficient BCAs against *Fusarium graminearum* (Gimeno et al., 2019; Khairullina et al., 2023; Özkale et al., 2023; Tian et al., 2018). *Trichoderma harzianum* is an important soil fungus that is a member of the subphylum *Pezizomycotina* (Otieno et al., 2003). *Trichoderma harzianum* has been used as a BCA against more than 20 fungal plant diseases, including tomato fusarium wilt caused by *Fusarium oxysporum f. sp. lycopersici* and soybean stem rot caused by *Sclerotinia sclerotiorum* (El-Komy et al., 2015; Zhang et al., 2016). Additionally, *C. rosea* is a common fungal BCA found in soils (Meng et al., 2022). *Clonostachys rosea* demonstrates effective management of a range of plant diseases attributed to *Fusarium* spp., *Sclerotinia sclerotiorum*, and *Botrytis cinerea* (Cota et al., 2009; Nobre et al., 2005). Moreover, *T. afroharzianum* and *C. rosea* have been demonstrated to stimulate plant growth(Han et al., 2022). The efficacy of *Trichoderma spp*. and *C. rosea* against *Fusarium* spp. on winter wheat (*Triticum aestivum* L.) in growth chambers and the field was examined previously, demonstrating a partial reduction of 56–76% of wheat foot and root rot caused by *Fusarium culmorum* using biological seed treatments (Roberti et al., 2006). This demonstrates the great potential that BCAs have in fighting diseases like root rot and wheat foot. The interaction between plant genotypes, biocontrol agents, and pathogens is crucial for understanding how

these factors work and how effective they are regarding disease resistance and better BCA compatibility.

Plants possess a robust immune system capable of combating most microbial invaders found in the environment, including bacteria, fungi, viruses, and oomycetes (Burketova et al., 2015). Various mechanisms are employed by BCAs to safeguard plants from the invasion of pathogens. These mechanisms include mycoparasitism, where the fungal host provides the mycoparasite with some or all of its nutrients, antibiosis, in which the BCA produces an antibiotic or inhibitory metabolite, induced resistance, which involves the initiation of defence responses in plants against pathogens (Ayaz et al., 2023). Additionally, BCAs can help promote plant growth (Ayaz et al., 2023). Even though is not classified as a biocontrol strategy, plant growth promotion is a significant property that can be attractive when choosing BCAs due to plant health benefits (Stenberg et al., 2021; Zamoum et al., 2015).

Resistance, or mutual incompatibility between the pathogen and the host, is a possible outcome of plant-pathogen interactions. This enables the host to stop or restrict the pathogen's growth (Heil & Bostock, 2002; Kause & Ødegård, 2012). When pathogens infect tissues of a resistant host plant, it causes a set of localized reactions in and around the infected cells in incompatible interactions (Lamb & Dixon, 1997). It has been previously shown that several BCAs can cause induced systemic resistance (ISR) in a range of infected crops. Through increased immunity, beneficial bacteria like *Pseudomonas* spp. and *Bacillus* spp. may help plants develop broad-spectrum disease resistance (Ayaz et al., 2021; Tahir et al., 2017). Induced systemic resistance primes plants for defence against pathogen attacks by activating genes linked to pathogenesis through phytohormone signalling pathways and defence regulatory proteins (Pieterse et al., 2014).

An increasing interest in commercializing BCAs, such as the most popular bacterial genera *Bacillus*, *Pseudomonas*, and *Agrobacterium*, as well as fungal genera *Trichoderma*, *Clonostachys*, *Aspergillus*, and *Penicillium*, has risen due to continuous research on using sustainable agricultural practices to enhance plant productivity (Ayaz et al., 2023). Variation among plant species and plant genotypes within a species can significantly affect how BCAs interact with plant-associated microbial communities (Hazen & Blum, 2016; Lacey et al., 2015; Smith & Goodman, 1999). Numerous investigations have been conducted to demonstrate how plants impact the biocontrol activities of BCAs (Arkhipov et al., 2023; Ryan et al., 2004; Smith & Goodman, 1999; Tucci et al., 2011). Specifically, different plant species exhibit variations in rhizosphere colonization, the generation of antibiotics, and the promotion of ISR (Umer et al., 2021).

Soilborne diseases, caused by various plant pathogens like bacteria, nematodes, and fungi, pose a threat to all crops, as evidenced by the significant yield boosts observed through

soil fumigation, even though other factors may contribute to this increase (Gohel et al., 2006; James Cook, 1992). By the end of the 21st century, under the moderate emission scenario 4,5, southern Europe, western Europe, and northern Scandinavia are anticipated to experience a significant rise in the frequency of drought events compared to recent times (Spinoni et al., 2018). The elevated occurrence of drought conditions, characterized by high temperatures and low moisture levels, may increase disease frequency and severity including fusarium foot root under climate change. This damage can be seen in cereals with symptoms including seed rot, damping-off before and after emergence, as well as reductions in straw production, grain yield, and grain quality. (Hollaway et al., 2013; Smiley et al., 2005).

Several *Fusarium* spp. are harmful plant pathogens that affect cereal plant species worldwide (Rauwane et al., 2020). A list of the top 10 fungal plant pathogens, ranked by scientific and agricultural significance, includes *Fusarium graminearum* at number four (Dean et al., 2012). The *F. graminearum* species complex (FGSC) can cause major yield and financial loss by causing fusarium head blight (FHB), fusarium foot rot (FFR), and fusarium crown rot (FCR) (Kazan & Gardiner, 2018; Rauwane et al., 2020). Many arid and semi-arid cropping regions including Australia, Canada, and South Africa experience FCR, causing devastating losses (Kazan & Gardiner, 2018). The pathogen manifests in infected plants as white, pink, orange, or reddish mycelial growth both inside and outside the infected area as well as in shriveled kernels (Hagerty et al., 2021a; McMullen et al., 2012). The *Fusarium* spp. is one of the most dangerous species regarding the production of secondary metabolites, namely mycotoxins that can affect both humans and animals (Mielniczuk & Skwaryło-Bednarz, 2020). Deoxynivalenol (DON), zearalenone (ZEN), and fumonisin B1 are among the most important mycotoxins on a world scale (Piacentini et al., 2019; Salgado et al., 2014). The lack of genetic resistance in wheat to relevant *Fusarium* spp. diseases highlight the complexity of the disease, and the risk of mycotoxin contamination of wheat grains requires the development of novel, efficient control strategies for diseases (Zhang et al., 2022). The symptoms of FFR are frequently associated with those of FCR since these pathogens can infect the roots and grow to the crown (Paulitz, 2006). The disease is known by several other names that are used interchangeably, such as dryland foot rot, dryland root rot, FFR, and fusarium root rot. Throughout this study, the designation used will be FFR. (Chakraborty et al., 2006; Hagerty et al., 2021b; Kazan & Gardiner, 2018; Winter et al., 2019).

The microbiome of the plant, which is important to control biological activity, is expected to be influenced by plant genotype (Collinge et al., 2022). Predicting the relationship between agronomically significant traits and plant function is becoming relevant in determining how genotype variation affects plant responses to BCAs (Collinge et al., 2022). To breed disease-resistant crops, certain genotypes can be selected for enhanced pathogen resistance, but they

can also be bred to vary in their ability to benefit from interactions with beneficial microorganisms, which can provide biocontrol of diseases or stimulate growth (Timper, 2014; Umer et al., 2021). A study evaluating the susceptibility of different oilseed rape cultivars against *Verticillium longisporum* infection as well as BCA compatibility showed that the BCA used could make the plant grow and provide protection against the pathogen, but both effects depend on the plant genotype (Abuamsha et al., 2011).

This study aims to investigate the genetic diversity among winter wheat genotypes concerning FFR susceptibility and their response to the fungal biocontrol agent *C. rosea* during FFR infection. The specific objectives of this dissertation are (i) to screen 190 winter wheat genotypes for resistance towards FFR disease caused by *F. graminearum* (ii) to evaluate genetic variation in plants for biocontrol compatibility with *C. rosea* in controlling *FFR* and (iii) to identify genetic markers segregating with FFR disease resistance in wheat and *C. rosea*-mediated biocontrol efficacy using a genome-wide association study.

1. State of the art

1.1. Biological control

1.1.1. *Clonostachys rosea*

Clonostachys rosea is one of the most successful examples of fungal BCAs. Previously known as *Gliocladium roseum*, it was re-classified as *C. rosea* due to differences in morphology, ecology, teleomorph, and DNA sequence that set it apart from other *Gliocladium* spp. (Schroers et al., 1999). *Clonostachys rosea* belongs to phylum Ascomycota, subphylum *Pezizomycotina*, class *Sordariomycetes*, subclass *Hypocreomycetidae*, order *Hypocreales*, family *Bionectriaceae* and genus *Clonostachys* (Sun et al., 2020; Tzelepis et al., 2015). Strains of *C. rosea* are present all over the world ranging in tropical, temperate, sub-arctic, and desert habitats, demonstrating a cosmopolitan distribution (Sun et al., 2020; Toledo et al., 2006; Zhai et al., 2016). Strains of *C. rosea* have been isolated from plant materials such as roots, leaves, flowers, and plant debris as well as from the soil and fungi (Costa et al., 2012; Mamarabadi et al., 2008; Muvea et al., 2014; Nobre et al., 2005; Bin Sun et al., 2017).

Clonostachys rosea is a soil-borne, saprophytic, filamentous, and mycoparasitic fungus that has been reported to act against several fungal diseases caused by different pathogens: *Alternaria* spp. (Jensen et al., 2007), *Bipolaris sorokiniana* (Jensen et al., 2002), *Botrytis cinerea* (Jensen et al., 2002, 2016), *Botrytis aclada* (Yohalem et al., 2004), *Fusarium culmorum* (Jensen et al., 2000), *Fusarium graminearum* (Kosawang et al., 2014; Schöneberg et al., 2015), *Rhizoctonia solani* (Tzelepis et al., 2015) and *Sclerotinia sclerotiorum* (Bin Sun et al., 2017). The principal mechanism *C. rosea* uses to combat pathogens is the secretion of cell-wall degrading enzymes (CWDEs), the production of secondary metabolites i.e., antibiotics and toxins, and the induction of plant resistance (Chatterton et al., 2009; Fatema et al., 2018). *Clonostachys rosea* is reported to secrete chitinases and glucanases to successfully degrade the cell wall of *Fusarium* sp. and *Pythium* sp. (Chatterton et al., 2009; Inglis & Kawchuk, 2002). *Clonostachys rosea* can also parasitize other fungi by attaching itself to the host fungi, creating infection structures, and penetrating the host (Hasan et al., 2022). *Clonostachys rosea* can also establish endophytic associations with plants that can affect the pathogen on the site of infection (Chatterton & Punja, 2010; Morandi et al., 2000). *Clonstachys rosea* is also reported to induce defence genes in plants, for example, the application of *C. rosea* increased the production of defence enzymes in the tomato leaves against grey mould (Mouekouba et al., 2014).

1.1.2. *Trichoderma harzianum*

Trichoderma spp. is a saprotrophic fungal species (teleomorph: Hypocrea) (Jaklitsch, 2009). They are found all over the world living in many different environments, such as soil, wood, bark, other fungi, and many more, proving their adaptability and potential for opportunism (Druzhinina et al., 2011; Jaklitsch, 2009; Oszust et al., 2020; Schmoll & Schuster, 2010). *Trichoderma* spp. has grown in significance recently due to its capacity to function as a fungicidal agent, promote plant growth, and provide plants with a variety of benefits in return for sucrose (Hermosa et al., 2012; Sood et al., 2020). A few of these include accelerating plant growth and yield, enhancing nutrient uptake, enhancing resistance to biotic and abiotic stresses, and altering the rhizosphere due to the chemical cues that the roots of plants release attracting *Trichoderma* spp. (Hermosa et al., 2012; López-Bucio et al., 2015).

Trichoderma spp. main mechanisms of biocontrol are mycoparasitism, antibiosis, competition for nutrients or space, and induction of plant defences. These mechanisms can work together or independently to suppress plant pathogens, making this fungus opportunistic, invasive, rapidly colonizing, and avirulent (Lorito et al., 2010; Mukhopadhyay & Kumar, 2020; Pacheco-Trejo et al., 2022; Wilson et al., 2008). It has been demonstrated that *Trichoderma* spp. has a high biocontrol potential for the management of post-harvest crown rot complex of bananas caused by a variety of fungal pathogens including *Colletotrichum musae*, *Fusarium verticillioides*, and *Lasiodiplodia theobromae* (Alvindia & Natsuaki, 2008; Sangeetha et al., 2009). In 2015, *Trichoderma spp.* isolates were present in more than 60% of all approved biopesticides, potentially for increased bioactivity (López-Bucio et al., 2015). Upon evaluating 27 different *Trichoderma* species, researchers found that *T. harzianum* demonstrated the highest production of recognized substances used to control pathogens and growth-promoting compounds. (Rush et al., 2021). *Trichoderma asperellum*, *T. atroviride*, *T. virens*, and *T. viride* are some of the other species most used in biocontrol and have been best studied in terms of their mechanisms of action (López-Bucio et al., 2015).

1.1.3. Biological control agents: mode of action

Although biocontrol as an effective strategy for managing plant diseases can be traced back centuries, it was not until the 19th century that biological control became the subject of advanced research (He et al., 2021). Natural enemies have long been employed in agriculture, and they have been used to treat almost every type of pest, for instance, herbivorous arthropods and entomopathogenic fungi (Gurr & You, 2016; Shields et al., 2019). The idea of employing BCAs to control plant disease was sparked by the finding of disease-suppressive soils, whereby antagonistic microorganisms present in the soil lessen the severity of soil-borne

diseases (Fira et al., 2018; Valan Arasu et al., 2023; Yao et al., 2023). Since most successful BCAs have evolved mechanisms for tolerating toxins from other organisms and are adapted to stressed conditions in these environments, they are typically effective competitors in the harsh biotic environment of soil, in the plant and its associated microbiome (Collinge et al., 2022).

To mitigate diseases in agriculture, biocontrol relies on the application of living organisms with an antagonistic role against the pathogen and/or with the capability to improve plant resistance against pathogens and can also be beneficial for plant health as it can reduce the bioavailability and plant uptake of dangerous chemicals (Pandit et al., 2022; Valan Arasu et al., 2023). The use of microorganisms as BCAs in IPM has become increasingly important in preventing disease emergence, either directly (by having an antagonistic effect on the pathogen) or indirectly (by increasing plant resistance mechanisms)(Köhl et al., 2019; Spadaro & Droby, 2016). Plants, pathogens, and the environment interact in complex ways that lead to plant diseases (Makiola et al., 2022; Singh et al., 2023). Through a variety of mechanisms, BCAs shields crops from disease-related harm (Köhl et al., 2019). Numerous biocontrol strategies such as induction of resistance in host tissue, antibiosis, parasitism, and competition for nutrients and space among others have been employed against pathogens (Jamalizadeh et al., 2011; Köhl et al., 2019) (Figure 1).

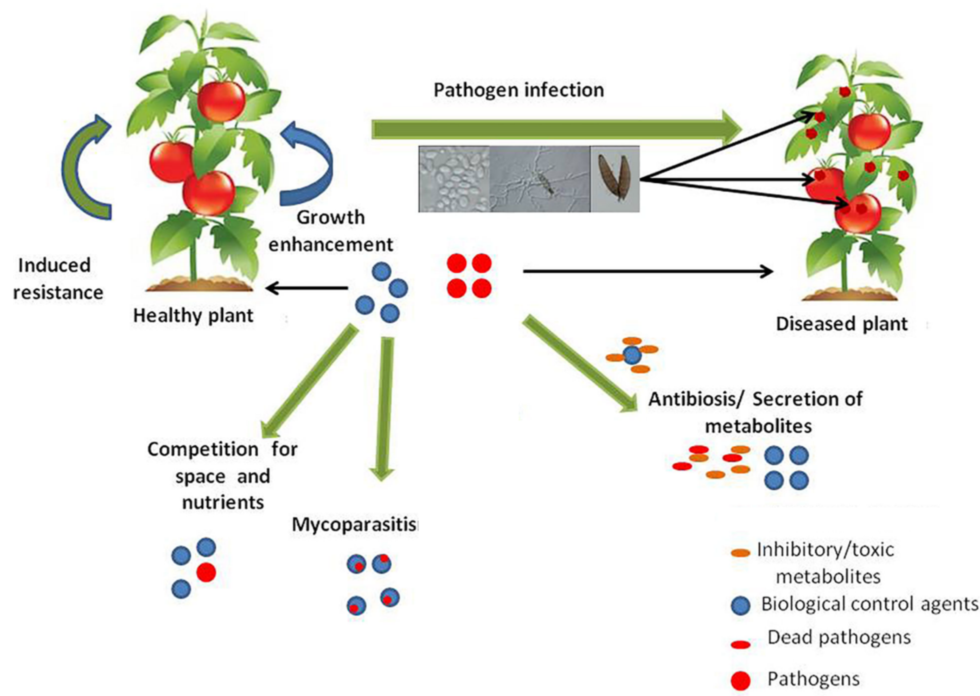


Figure 1 Important mechanisms of action for fungal antagonists' biological control of plant fungal diseases (adapted from Thambugala et al., 2020).

1.1.3.1. Competition

Competition is characterized by niche overlap, which arises when two or more microbial populations simultaneously demand the same resource (Jamalizadeh et al., 2011; Köhl et al., 2019; Roca-Couso et al., 2021). In biocontrol systems, competition for nutrients such as carbohydrates, nitrogen, and oxygen, and available space is frequently proposed as a possible mechanism of action (Thambugala et al., 2020). For this to occur, a particular nutrient or resource must be required by both the pathogen and the antagonist. When the antagonist is present in sufficient numbers at the right time and place and uses nutrients or resources more effectively than the pathogen, this mode can be effective (Sánchez-Montesinos et al., 2021).

1.1.3.2. Antibiosis

Antibiosis is the inhibition or destruction of a microorganism by substances like a specific toxin, antibiotics, or enzymes that are produced by another microorganism, such as fungi and bacteria (Dudeja et al., 2021; Palmieri et al., 2022). Several species like *Trichoderma* spp., *Clonostachys* spp., and *Aspergillus* spp. are known to produce antibiotic compounds, like 6-PAP, Clonorosein, and viridin that are effective against a variety of microorganisms (Dudeja et al., 2021; Ghorbanpour et al., 2018; Han et al., 2020; Sun et al., 2020; Verma et al., 2007).

1.1.3.2. Mycoparasitism

A direct competitive relationship in which one organism feeds on the other to obtain nutrients is called parasitism (Köhl et al., 2019). Mycoparasitism is used when one fungus (mycoparasite) parasites another fungus (host) (Ghorbanpour et al., 2018). Mycoparasites are known to secrete enzymes that break down the cell walls of fungi, including chitinases, glucanases, and proteases (Pachenari & Dix, 1980). The physical penetration of the mycoparasite into the host hyphae, as well as the secretion of different enzymes or secondary metabolites that cause the degradation of fungal structures and subsequent uptake of nutrients and metabolites from the host fungus, are the mechanisms by which mycoparasitism is mediated (Daguerre et al., 2014).

1.1.3.3. Plant growth promotion

Microorganisms can have an indirect impact on crop health and phenotypic plasticity by influencing plant growth and defence responses on a large scale (Goh et al., 2013). Plant root

colonization with plant growth-promoting microorganisms can protect plants from pathogens, promote plant growth, and incentivize the movement of microorganisms in response to chemical signals to root exudates (El-Saadony et al., 2022). Plant growth-promoting rhizobacteria employ a variety of strategies, both direct and indirect, to stimulate plant growth. Examples of direct mechanisms include synthesizing plant hormones, solubilizing phosphate, and improving iron absorption. Indirect effects are manifested through the production of antibiotics, competition for nutrients, parasitism, inhibition of pathogen toxins, and inducing resistance (Elnahal et al., 2022). Predominantly found in genera like *Trichoderma*, *Penicillium*, and *Aspergillus*, these organisms enhance nutrient availability through organic acid and siderophore production, synthesize plant growth regulators, secrete hydrolytic enzymes, reduce ethylene levels, improve water uptake, induce plant defense, and alleviate abiotic stresses via gene expression modulation (Argumedo-Delira et al., 2022; Javed et al., 2020; Jin et al., 2022; Lombardi et al., 2020). A few *Trichoderma* spp. can directly oppose plant pathogens and promote plant growth, but they can also disrupt signalling networks in their host plants to increase disease resistance and stress tolerance (Alfiky & Weisskopf, 2021).

1.1.3.4. Induction of plant resistance

Plant-induced resistance is primarily caused by effective pathogen recognition and fast defensive response (Syed Ab Rahman et al., 2018). Enhancement of plant resistance takes advantage of mechanisms of the plant immune system, where two phenotypically similar forms of systemic immunity have been identified in plants: the ability of beneficial non-pathogenic microorganisms to prime plants for ISR and the local infection with plant pathogens that can lead to systemic acquired resistance (SAR) (Pieterse et al., 2009). Systemic acquired resistance in plants is a defense response triggered by pathogen infection, leading to hypersensitivity and localized necrosis. This occurs when plants activate their defense mechanisms in response to the pathogen's initial invasion (Van Loon et al., 1998). While ISR is effective against a variety of pathogens, it differs from SAR in that the induction does not cause the host plant to exhibit outward symptoms (Van Loon et al., 1998). *Bacillus amyloliquefaciens*, *B. atrophaeus*, *B. cereus,* and others have been shown to effectively combat fungal attacks through ISR (Yu et al., 2022). Recent research indicates that these beneficial microbes trigger early ISR responses in plants, including increased expression of pathogenesis-related genes and also the production of defense-related enzymes like phenylalanine ammonia-lyase, polyphenol oxidase, peroxidase, β-1,3 glucanase, and chitinase, as well as the accumulation of reactive oxygen species (Guo et al., 2019; M. Wang et al., 2020).

1.1.3.5. Plant genetic variation

Multiple studies in different crops like lentils, tomatoes, sugar beet, and maize have shown that plant genetic background is important for biocompatibility between plants and BCAs (Bazghaleh et al., 2020; Harman et al., 2004; Schmidt et al., 2020; Tucci et al., 2011). Prashar and Vandenberg (2017) did a study where they evaluated two commercially available *Trichoderma* formulations for control of aphanomyces root rot and promotion of plant growth in 23 lentil genotypes. They found that the two *Trichoderma* strains had a genotype-specific influence on the agronomic performance of lentil genotypes under *Aphanomyces euteiches* infection and control conditions. Despite root colonization in all treated plants, the effect of *Trichoderma* treatments varied significantly, ranging from positive to negative, differentially affecting plant growth of lentil genotypes (Prashar & Vandenberg, 2017). In another study, the aim was to determine which genotypes among 50 genotypes of sorghum were compatible with biocontrol agent *Fusarium oxysporum* f. sp. *strigae* and whether this specific fungus could be used to control the weeds in soils that were infested. Striga plant emergence was significantly reduced due to the strong rhizosphere establishment and effective attachment of the biocontrol agent to the sorghum root, particularly when paired with Striga-resistant sorghum varieties, resulting in a decrease in the number of Striga by up to 92% (Rebeka et al., 2013).

1.1.4. Microbial biocontrol formulations for commercial applications

Due in large part to their host specificity, inconsistent performance in varying field environments, commercial applications, and the limitations of the legislation in UE, usage of biological control of plant diseases has been slow to take off. Using cutting-edge biotechnological techniques, it is imperative to create novel BCA formulations with improved levels of stability, effectiveness, and survival to solve this issue (Fravel, 2005; Thambugala et al., 2020). Many products with a biological basis are marketed globally to manage fungal plant diseases. For example, a few commercial products based on *C. rosea* have been made available globally, including Kamoi® in Brazil, Vectorite® and Endofine® in Canada, and Prestop® (Lahlali & Peng, 2014; Mascarin et al., 2022). There are also several Bioprotection products based on *T. harzianum* that are sold commercially (such as Shibeijian, Trichodex, and Topshield) that can be used to control a variety of plant diseases (Li et al., 2023; Stenberg et al., 2021).

1.2. *Fusarium* species

The filamentous fungi from the genus *Fusarium*, order Hypocreales, phylum Ascomycota are an economically important group, containing a plethora of agronomically important plant pathogens, mycotoxin producers, and opportunistic human pathogens, infecting via both soil and air (Ekwomadu & Mwanza, 2023; Ma et al., 2013). The *Fusarium* genus has at least 300 phylogenetically distinct species or species complexes with varying host specificity (Ma et al., 2013; O'Donnell et al., 2015). *Fusarium* spp. can adapt to multiple habitats and are commonly found in areas with moderate climates, contaminating crops around the world (Ekwomadu & Mwanza, 2023). *Fusarium* spp. cause multiple diseases like rots, blights, cankers, and wilts in field crops, forest crops, ornamental plants, and horticulture plants but also in animals and humans through the production of mycotoxins affecting especially the immunosuppressed individuals (Anaissie & Nucci, 2002; Dean et al., 2012; Evans et al., 2004; Ma et al., 2013). Multiple economically relevant plant species are susceptible to at least one or more *Fusarium* spp. (Aoki et al., 2014). *Fusarium* spp. have developed sophisticated penetration, infection, and colonization strategies to weaken the plant defences of vulnerable hosts and spread disease (Rauwane et al., 2020).

1.2.1. *Fusarium graminearum*

Fusarium graminearum, *F. pseudograminearum*, and *F. culmorum* are a group of closely related species, that are part of the complex of *Fusarium* species associated with diseases of cereals (Backhouse & Burgess, 2002; Del Ponte et al., 2022). The fungus *F. graminearum* (teleomorph: *Gibberella zeae*) is a highly destructive pathogen that is a pervasive species in the fusarium complex that causes head blight (FHB), root rot, and foot rot (FFR) in barley (*Hordeum vulgare* L.) and wheat crops (Osborne & Stein, 2007; Xu et al., 2018).

The main species associated with FFR and FHB differ among countries and regions within countries, making the distribution of diseases caused by each species often related to climate (Backhouse & Burgess, 2002). Fusarium head blight is a significant factor in limiting wheat production leading to severe economic losses in Europe, North America, and Asia (Cowger et al., 2018; Pasquali et al., 2016; Xu et al., 2018). In the United States and Canada, FHB losses in wheat during the 1990s are estimated to have exceeded 3 billion dollars (Chin et al., 2023). Fusarium foot rot poses a threat to barley and wheat production in arid and semi-arid globally, including major wheat-producing countries like the United States, Canada, New Zealand, China, and many others (Kazan & Gardiner, 2018). For example, in the Pacific Northwest of the United States, natural inoculum levels led to a 35% decrease in wheat grain

yield, while in Australian cereals, FFR is estimated to cause a 10% yield loss (Murray & Brennan, 2009; Smiley et al., 2005). Sweden over the last ten years has experienced high levels of *Fusarium* spp. produced mycotoxin contamination, which has resulted in significant economic losses and issues for farmers (Karlsson et al., 2023).

An integrated control system for managing FFR should include measures to be applied at every stage of production, from the field to the final consumer, to control mycotoxin in the grain chain (Kadkol et al., 2021; Khudhair et al., 2014; Malosetti et al., 2021; Nada et al., 2022). Pre-harvest tactics are mostly dependent on the environment and the use of various agricultural techniques, including crop rotation, crop selection, irrigation, soil tillage, chemical or biological control of plant diseases, insect control among others (Chen et al., 2019; Ekwomadu & Mwanza, 2023). It should be noted that pre-harvest techniques used cannot ensure that food will not contain mycotoxins (Karlovsky et al., 2016). The most crucial post-harvest tactics for successfully preventing the growth of fungi and the production of mycotoxin in harvested commodities include temperature control during storage and transportation, relative humidity, and the management of the grains' moisture content and water activity (Kabak et al., 2006; Magan et al., 2010).

1.2.2. Mycotoxins of the *Fusarium graminearum* species complex

The *F. graminearum* species complex (FGSC), comprising a minimum of 16 distinct species, is accountable for substantial mycotoxin contamination, resulting in marked declines in grain yield and quality (Hao et al., 2017). This includes issues such as aborted or shriveled seeds, diminished seed size, and the rejection of grain in small-grain cereals like oats (*Avena sativa* L.), wheat, and barley (Nelson et al., 1994; Osborne & Stein, 2007; Rocher et al., 2022; van der Lee et al., 2015; Vogelgsang et al., 2019). Multiple mycotoxins are produced by *Fusarium* spp. when they infect crops by producing aggressive secondary metabolites like T2 toxin, nivalenol, DON, fumonisin, and ZEN (Karlsson et al., 2023; Morimura et al., 2020). *Fusarium graminearum* uses DON in conjunction with CWDEs to get past host defences in small-grain cereals allowing the fungus to spread and grow into the inner tissues to feed on host cell debris (Rauwane et al., 2020). To regulate the use of food and feed contaminated with DON and ZEN mycotoxins, the European Commission (EC) has established maximum limits of 1.25 mg/kg DON and 0.1 mg/kg ZEN for raw wheat and barley grains, and 1.75 mg/kg DON for oats (Abbas & Yli-Mattila, 2022).

1.2.3. *Fusarium graminearum* disease cycle and symptoms

During the winter period, *F. graminearum* persists on crop residues such as maize stalks, mainly as mycelium. In the form of perithecia and saprophytic mycelia, which release ascospores and macroconidia, respectively (the primary inoculum) when environmental conditions are favourable (Gao et al., 2020; Keller et al., 2014; Trail, 2009). The fungal structures disperse through wind or rain splashes, landing on vulnerable plant surfaces where they colonize the plant tissue (Parry et al., 1995; Trail, 2009). Independently of where *F. graminearum* attacks, necrosis develops as the fungus grows quickly and colonizes the tissue behind the infection front, spreading radially (Jansen et al., 2005; Markell & Francl, 2003). As *F. graminearum* infects the coleoptile, it advances through the subcrown internode and seedling leaf sheaths before infiltrating the stem epidermal tissues, often entering via stomatal openings (Knight & Sutherland, 2013, 2016). For FFR disease, *F. graminearum* affects the stem base, crown, and roots of wheat plants (Mudge et al., 2006). In severe cases, FFR results in the death of young seedlings either before or shortly after emergence. Eventually, brown lesions at the base of the stem may result from infections of this disease, and later in the season, these lesions may spread to most of the stem (Zhang et al., 2022). Favourable conditions like warm and moist spring weather, are conducive for *F. graminearum* growth and conidial maturation (Markell & Francl, 2003; Sutton, 1982). During this time, conidia and ascospores are released at the same time as the flowering of cereal crops (Markell & Francl, 2003; Sutton, 1982). Airborne spores cause FHB in wheat fields by landing on cereal crop florets, germinating, and entering the plant through degenerating anther tissues or natural openings like the base of the bracts (Trail, 2009). The fungus almost immediately expresses genes for DON biosynthesis after infecting wheat, allowing it to spread in the plant (Jansen et al., 2005). At the necrotrophic stage, the fungus begins to colonize the host substrate more vigorously, and eventually plant death triggers complete colonization of the substrate (Figure 2) (Goswami & Kistler, 2004; Inch & Gilbert, 2003; Karlsson et al., 2021). The pathogen manifests in infected plants as white, pink, orange, or reddish mycelial growth both inside the stem and outside the infected area as well as in shriveled kernels (Hagerty et al., 2021a; McMullen et al., 2012).

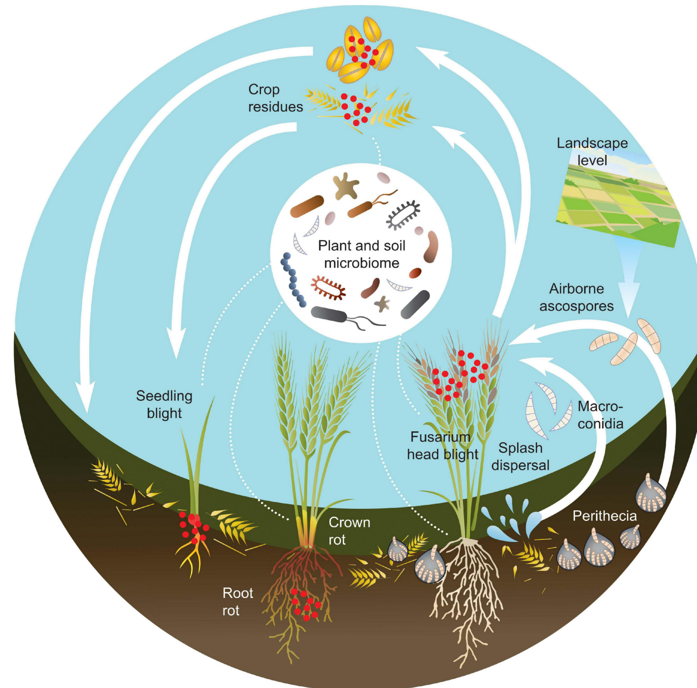


Figure 2 Life cycle of *Fusarium* spp. and the interactions between the microbiota and diseases related to it in cereals. *Fusarium* inoculum is represented by the red dots (Karlsson et al., 2021).

1.2.4. Plant response to fusarium foot rot

Plants employ an array of chemical and physical defence mechanisms to combat infections. These include the expression of various pathogenesis-related proteins, production of phytoalexins, lignification of plant cell walls, synthesis of lytic enzymes, and activation of the hypersensitive response (Jamalizadeh et al., 2011; Köhl et al., 2019). Key signalling molecules in plant defence, including ethylene and jasmonate, are vital for combating necrotrophic pathogens and facilitating crucial plant-microbe interactions. Furthermore, salicylic acid plays a pivotal role in defending against biotrophic pathogens and in inducing SAR (Ghozlan et al., 2020; Li et al., 2019). In response to treatment with beneficial microorganisms, salicylic acid triggers a hypersensitive reaction leading to localized programmed cell death, restricting pathogen spread, and inducing the expression of pathogenesis-related genes, enhancing defence against stress (Syed Ab Rahman et al., 2018). Plant defence mechanisms are often activated by stimuli recognized by receptors or chemical pathways. Pathogen-associated molecular patterns are among such stimuli, triggering defence responses including ISR and SAR, which bolster host resistance against the identified pathogen (Ghorbanpour et al., 2018; Köhl et al., 2019).

2. Aim and objectives

The main aim of this study is to explore the genetic variation among winter wheat genotypes for fusarium foot rot and responsiveness to the fungal biological control agent *Clonostachys rosea* during Fusarium root infection.

We hypothesize that:

(i) winter wheat genotypes differ in their resistance towards FFR disease;

(ii) winter wheat genotypes differ in their compatibility with the fungal biological control agent *Clonostachys rosea*;

(iii) disease resistance and biocontrol compatibility are inherited independently from each other.

Therefore, the specific objectives of this study are:

(i) to screen 190 winter wheat genotypes for resistance towards FFR disease caused by *F. graminearum*;

(ii) to evaluate genetic variation in plants for biocontrol compatibility with *C. rosea* in controlling FFR disease;

(iii) to identify genetic markers segregating with FFR disease resistance in wheat and *C. rosea*-mediated biocontrol efficacy using a genome-wide association study.

As a side question, this study aims to explore the genetic variation among winter wheat genotypes for fusarium foot rot and responsiveness to the fungal biological control agent Trich*oderma afroharzianum* during Fusarium root infection.

Therefore, the specific objectives of this study are:

(i) to select 20 winter wheat genotypes based on different biocontrol efficacy of *C. rosea* for resistance towards FFR disease caused by *F. graminearum*;

(ii) to evaluate plant genetic variation for biocontrol compatibility with *T. afroharzianum* in controlling FFR disease.

3. Materials and methods

3.1. Plant material

This study assessed 190 winter wheat genotypes, includes landraces and cultivars, acquired from the Nordic Genetic Resources Centre, Alnarp, Sweden, representing a diverse array of germplasm sourced from seven distinct countries spanning a broad geographical spectrum including Sweden, Afghanistan, Germany, Finland, Norway, Netherlands, and Germany (see Supplementary Table 1 for details)(Odilbekov et al., 2019). Uniform seeds, characterized by consistent size and color, stored at 4°C were surface sterilized before use. The sterilization process involved immersion in milliQ water (1 min), followed by detergent treatment (1 min), three cycles of rinsing with milliQ water (1 min each), exposure to a 2% NaOCl solution (10 min), and a final rinse with milliQ water (3 min). The seeds were air-dried before use.

3.2. Fungal strains preparation and application

Fusarium graminearum strain PH-1 was revived from a glycerol stock maintained at -80°C by inoculating agar plugs onto potato dextrose agar (PDA) (VWR Chemicals) petri plates. These plates were then incubated at 20°C in darkness for 7 days. Agar plugs of 5 mm diameter were excised from the actively proliferating regions and were used for inoculation in pots, facilitating disease development in the bioassay setup.

Both *Clonostachys rosea* strain IK726 and *Trichoderma afroharzianum* strain T22 were revived from glycerol stocks preserved at -80°C. *Clonostachys rosea* was cultured on PDA petri plates at 20°C under dark conditions for approximately 20 days. Spores were harvested from the PDA plates by flooding the plates with sterile water, followed by filtration using Miracloth to segregate spores from mycelium. Seeds were then coated with a suspension containing 1×10^6 colony-forming units per ml (CFU/ml) and adjusted with sterile water using a hemacytometer chamber (Bright-Line Hemacytometer).

To assess the efficacy of the seed coating with the biocontrol agents (BCAs), a re-isolation of CFU was done using a dilution series on three check winter wheat genotypes (Kranich, Stava, and Festival). 10 seeds from each genotype were added to 10 ml sterile deionized water in a falcon tube to release the CFU in water, the falcon tubes were vortexed for 20 to 30 seconds and serial dilution to 10^{-1}, 10^{-2}, and 10^{-3}. PDA plates (100 µl) were inoculated with 10^{-2} and 10^{-3} dilutions, with three duplicates prepared for each genotype. After four days of room temperature incubation in darkness, the CFUs were counted. At a 10^{-2}

dilutions Stava, Festival and Kranich had 6.67 x 10^3 CFU/ml, 7.33 x 10^3 CFU/ml, and 8 x 10^3 CFU/ml, respectively.

3.3. Bioassay setup

To explore the genetic variability of 190 winter wheat genotypes concerning fusarium foot rot (FFR) susceptibility and response to the biocontrol agent *C. rosea* for FFR disease, an *in vivo* bioassay was conducted using an adapted protocol done previously (Knudsen et al., 1995). Each tray with 40 plastic pots (5 × 5 × 5 cm) was filled with moistened sand (Rådasand, 0.5 to 1 mm), and in each pot, 3 seeds were sown evenly spaced in a 2 cm deep and 1.5 cm wide hole. Additionally, a 5-mm plug of *F. graminearum* mycelium was placed between the seeds, positioned facing upward, and then covered with sand. For seed coating, seeds were shaken on a rotary shaker at 110 rpm for 30 minutes. The experimental setup involved two treatments:

i) seeds coated with *C. rosea* IK726 at 1 × 10^6 cfu/ml (FgCr);

ii) control treatment without *C. rosea*, soaked in sterile water (Fg).

The experiment used a randomized complete block design (RCBD) where trays were treated as blocks, and genotypes assigned were randomized in pots within each block or tray. 5 trays were randomly selected for each treatment, making 5 biological replicates for each genotype in each treatment. The experiment was also divided into 6 batches to accommodate the number of genotypes and replicates involved in the study. Three check genotypes i.e. Kranich, Stava, and Festival were included in each tray of each batch to control for batch variation. Trays were housed in a phytotron chamber with a light photoperiod of 16 hours at 200 μmol/m^2 s at 20°C and a dark photoperiod of 8 hours at 16°C.

Plants were harvested and washed 19 days after inoculation. Root length, shoot length, and plant length were measured, and disease severity was assessed on a scale ranging from 0 to 4, with intervals of 0.5, where 0= healthy plant, 1 = slightly brown roots and coleoptiles, 2 = moderately brown roots and coleoptiles, 3 = severely brown roots and coleoptiles, and 4 = dead plants (Knudsen et al., 1995).

Additionally, as a supplementary investigation, the same bioassay was conducted to assess the compatibility and efficacy of *Trichoderma afroharzianum* in controlling FFR.To understand whether winter wheat genotypes had comparable responses to *T. afroharzianum* as they did to *C. rosea* for FFR, 20 genotypes were selected based on their respective efficacy levels with *C. rosea* (classified as low, mid, or high efficacy; see Supplementary Table 1 for details). The experiment was conducted following the same protocol as *C. rosea*.

3.4. Statistical analysis

Statistical analysis was executed through a linear model framework to examine the effects of each factor on traits. This approach accounted for potential variations stemming from experimental design factors and the interaction between biological factors treatment and genotype.

The linear model used in the analysis is as follows:

$$Y = \mu + \text{Batch} + \text{Batch/Block} + \text{Treatment} + \text{Genotype} + \text{Treatment*Genotype} + \varepsilon$$

where:

- Y represents the response variables (disease score, plant length, shoot length, or root length)
- μ denotes the overall mean or intercept
- Batch accounts for variation attributable to different experimental batches
- Batch/Block accounts for variation attributable to different blocks nested within batches
- Treatment indicates the effect of the applied treatments. (two levels: Fg and FgCr)
- Genotype indicates the effect of the plant genotype. nordID denotes genotype ID
- Treatment * Genotype accounts for interaction among plant genotype and treatment effects
- ε represents the residual effect

Analysis of variance (ANOVA) was performed on the linear model to evaluate the significance ($\alpha=0.05$) of the above-described experimental design factors and biological factors. To confirm if the model assumptions were satisfied, normality and homoscedasticity of variance were checked graphically on the model residuals.

Subsequently, post-hoc analyses were conducted, to estimate the best linear unbiased estimators (BLUEs) of genotypes using the marginal means of genotypes in each treatment. Following the computation of BLUEs for the genotypes within each treatment, pairwise comparisons were conducted for each genotype across treatments. Tukey's honestly significant difference method was used for these comparisons to adjust for multiple comparisons and control the error rate. These pairwise comparisons for each genotype were used as an estimator for biocontrol efficacy. Moreover, Pearson’s correlation between treatments and traits was performed using BLUE estimates.

Data analysis was performed using the R statistical language, version 4.3.1 "Beagle Scouts" (R Core Team, 2023). Linear regression models were fitted using the lm() function (Wilkinson & Rogers, 1973). ANOVA was performed using the anova() function (Everitt, 1992). Post-hoc analysis was conducted employing the emmeans() function (Searle et al., 2024), followed by pairs() function (Becker et al., 1988) for pairwise comparisons. Correlations were estimated using the stat_cor() function. Particularly, the tidyverse package (Wickham et al., 2019) was utilized for majority of data processing and visualization.

3.5. Genome-wide association studies (GWAS)

188 wheat genotypes from the current panel were previously genotyped using a 20K single nucleotide polymorphism (SNP) marker array at TraitGenetics GmbH, Germany (Odilbekov et al., 2019). A total of 7360 SNP markers were used for genome-wide association study (GWAS) after filtering out the markers with > 20 % missing alleles and < 5 % minor allele frequency. To perform GWAS, general linear model (GLM) was used, which is a single locus GWAS model (Price et al., 2006). To correct for relatedness and population structure, the kinship matrix and the first 17 principal components (explaining 50 % variation) were used as covariates in the analyses. For significant marker-trait association, a threshold of negative log (1/number of SNP markers) was used to overcome the overstringency of Bonferroni test threshold (0.05/number of SNP markers) and low sample size (M. Wang et al., 2012; Yang et al., 2011). To perform GWAS the genome association and prediction integrated tool (GAPIT) version 3 was used (Wang & Zhang, 2021).

4. Results

A bioassay was conducted to assess potential variances among winter wheat genotypes in their resistance to FFR disease and their compatibility with the fungal biological control agent *C. rosea*. Additionally, the bioassay tested if resistance to the disease and compatibility with biocontrol agents are inherited independently.

4.1. Analysis of variance (ANOVA) of traits

An analysis of variance (ANOVA) was conducted to assess the influence of various factors on traits root length, shoot length, plant length, and disease score across 190 winter wheat genotypes (Table 1). Visual examination of diagnostic plots, including fitted vs. residual plots for homoscedasticity and QQ plots for normality, showed that the model assumptions were met for every trait. The data points in QQ plots closely matched the diagonal line, indicating normality. Furthermore, homoscedasticity was marked by the fitted vs. residual plots, which showed a random scatter (Supplementary Figure 1). Significant differences between treatments ($p < 0.05$) were observed across all assessed traits (Table 1, Figure 3). For root length, the mean value was 5.24 cm for Fg and 9.30 cm for FgCr (Figure 3a). Similarly, in shoot length, FgCr plants exhibited a mean value of 13.05 cm, contrasting with Fg mean length of 4.85 cm (Figure 3b).

Moreover, FgCr plants showed a mean plant length of 22.34 cm, whereas Fg had a mean length of 10.08 cm (Figure 3c). Examining disease scores, Fg displayed a higher disease score, with a mean score of 3.40, whereas FgCr exhibited a mean disease score of 1.42 (Figure 3d). Overall, in root length, shoot length, and plant length, values tend to be higher under the FgCr treatment compared to the Fg treatment. On the other hand, disease scores tend to be lower under the FgCr treatment compared to the Fg treatment.

Moreover, a significant plant genotype (nordID) effect was observed for shoot length ($F = 2.28$, $p < 0.05$), plant length ($F = 1.77$, $p < 0.05$), and disease score ($F = 1.75$, $p < 0.05$), but not for root length ($F = 1.15$, $p > 0.05$). The interaction between treatment and genotype (GxT) was also significant in all traits (table 1). This interaction highlights that different genotypes perform differently across the treatments.

Table 1 Results of two-way ANOVA test showing F-values and level of significance for traits. Significance indicator denoting the significance level of the p-value (* indicates $p < 0.05$, and absence of asterisk indicates non-significance).

Trait	Source of Variation	DF	Sum Of Squares	Mean Squares	F- value	*p*-value	*p* < 0,05
Root length (cm)	B	5	2384.69	476.94	91.63	2.1E-84	*
	T	1	8761.25	8761.25	1683.15	4E-248	*
	G	189	1134.03	6.00	1.15	0.09	
	BxBL	24	1244.88	51.87	9.95	2.6E-34	*
	GxT	187	1670.08	8.93	1.72	5.6E-08	*
	Residuals	1516	7891.17	5.21			
Shoot length(cm)	B	5	2532.12	506.42	42.20	8.6E-41	*
	T	1	33836.99	33836.99	2819.63	0	*
	G	189	5174.90	27.38	2.28	1.8E-17	*
	BxBL	24	1500.30	62.51	5.21	6.4E-15	*
	GxT	187	4365.91	23.35	1.95	1.6E-11	*
	Residuals	1516	18192.75	12.00			
Plant length (cm)	B	5	9391.94	1878.39	68.202	2E-64	*
	T	1	76957,71	76957,71	2794,24	0	*
	G	189	9248.25	48.93	1.78	6E-09	*
	BxBL	24	5187.30	216.14	7.85	1.2E-25	*
	GxT	187	9783.69	52.319	1.90	8.6E-11	*
	Residuals	1517	41780.56	27.54			
Disease score (0-4)	B	5	63.77	12.75	23.57	7.1E-23	*
	T	1	1978.48	1978.48	3656.05	0	*
	G	189	179.87	0.95	1.76	1.1E-08	*
	BxBL	24	98.28	4.10	7.57	1.7E-24	*
	GxT	187	143.58	0.77	1.42	0.0003	*
	Residuals	1517	820.93	0.54			

B: Batch, BL: Block, G: Genotype, T: Treatment, BxBL: Batch X Block interaction, GxT: Genotype X Treatment interaction

DF: Degrees of freedom

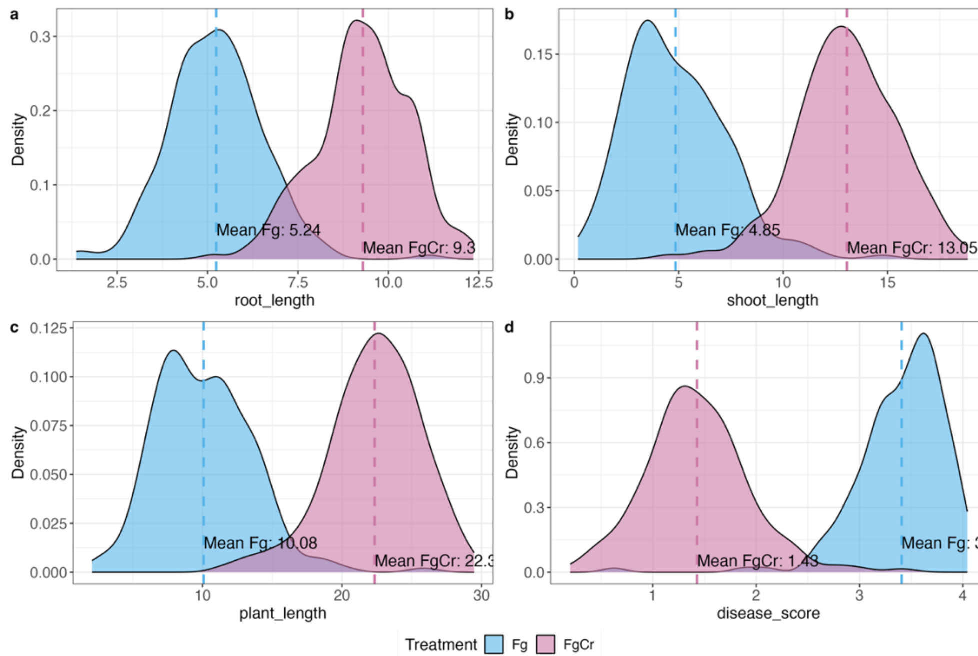


Figure 3 Distribution of BLUEs in treatments Fg and FgCr for the traits (a) root length, (b) shoot length, (c) plant length and (d) disease score. Dashed horizontal lines represent the arithmetic means of BLUEs in each treatment for each trait.

4.2. Correlations between treatments within each trait

Correlations between treatments FgCr (*F. graminearum* + *C. rosea*) and Fg (*F. graminearum* only) for each trait, disease score, shoot length, plant length, and root length were analysed with a Pearson's correlation coefficient (R). Both root length and disease score showed a significant weak correlation R = -0.18 ($p < 0.05$) and R = 0.17 ($p < 0.05$), respectively (Figure 4a, d). Alternatively, no significant ($p > 0.05$) correlation was observed for shoot length (Figure 4b) and plant length (Figure 4c). This variation in statistical significance suggests that among the 190 genotypes, there is a variability in genotypes performance across treatments treatments FgCr (*F. graminearum* + *C. rosea*) and Fg (*F. graminearum* only).

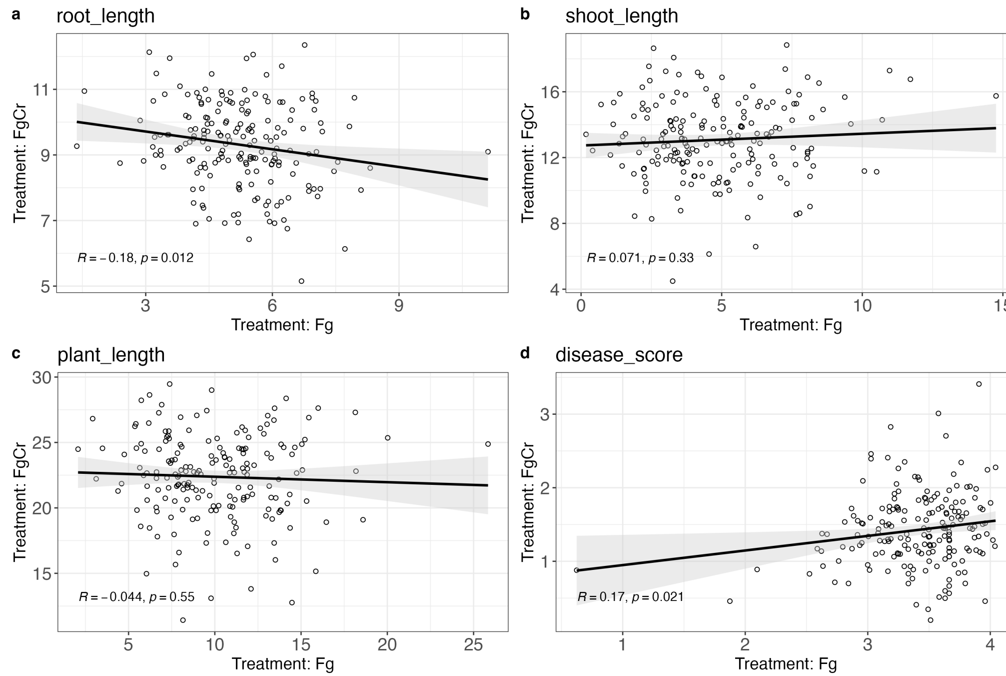


Figure 4 Correlation analysis between treatments FgCr and Fg in 190 winter wheat genotypes for the traits (a) root length, (b) shoot length, (c) plant length and (d) disease score using Pearson correlation coefficient (R). Each data point represents the BLUEs of genotypes across treatments.

4.3. Correlation between plant growth and disease severity

Pearson's correlation analysis was conducted to investigate the relationship between plant length and disease score in two treatments: FgCr (*C. rosea*) and Fg (*Fusarium graminearum*). In FgCr treatment, a moderate negative correlation was observed between plant length and disease score (R = -0.54, $p < 0.05$), indicating that as disease score increases, plant length tends to decrease. For the Fg treatment, a stronger negative correlation was observed between plant length and disease score (R = -0.82, $p < 0.05$). This indicates a more pronounced inverse relationship between disease severity and plant length in the Fg treatment compared to FgCr (Figure 5). The fact that the Fg treatment's negative correlation is stronger indicates that plants in this treatment are more severely impacted by disease, which leads to a larger reduction in plant length. The treatment containing *C. rosea* provides some mitigation

against the disease's impact on plant length, as indicated by the moderate correlation observed in the FgCr treatment.

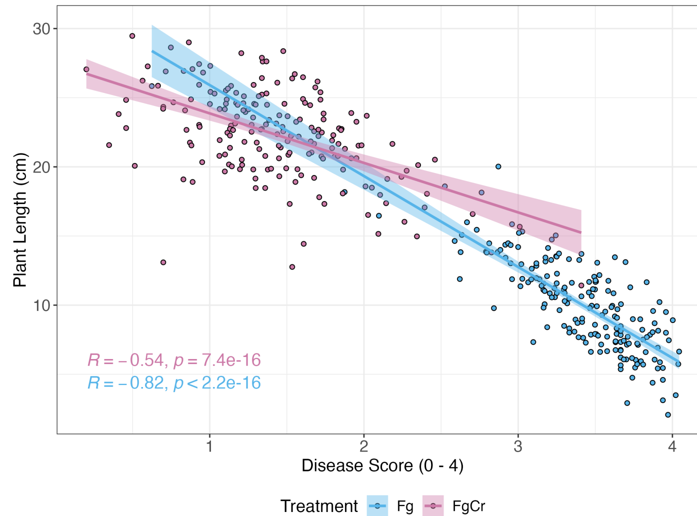


Figure 5 Pearson’s correlation (R) between disease score (0-4) and plant length (cm) in treatments FgCr and Fg. Each data point represents the BLUEs for plant length and disease score.

4.4. Pairwise contrasts for assessment of *C. rosea* efficacy

To identify the genotypes for which significant (p < 0.05) *C. rosea* was observed, the comparison between *C. rosea* treatment (FgCr) and control treatment (Fg) was performed using the pairwise contrast estimates (Fg - FgCr or FgCr - Fg) for each of the 190 genotypes for traits disease score, plant length, shoot length, and root length.

For disease score, 180 genotypes showed a significant (p < 0.05) reduction of disease in the FgCr treatment (Figure 6a), showing that *C. rosea* has a high biocontrol efficacy against the *F. graminearum* across a wide range of genotypes. For root length, pairwise contrasts between the treatments showed that 135 genotypes had a significant (p < 0.05) improvement in root length in the FgCr treatment (Figure 6b). For shoot length, 166 genotypes showed significant (p < 0.05) improvement in length in treatment FgCr (Figure 6c). Lastly, for plant length, 163 genotypes had a significant (p < 0.05) increase in plant length in the FgCr treatment (Figure 6d). Overall, the contrast between the treatments showed that *C. rosea* is highly effective in reducing disease severity and improving plant growth across a diverse range of wheat genotypes, but not all genotypes had the same response for *C. rosea*. *Clonostachys rosea* efficiently reduces *F. graminearum* symptoms, but not with the same efficacy in all genotypes showing variation in biocontrol compatibility of *C. rosea* in controlling *F. graminearum*.

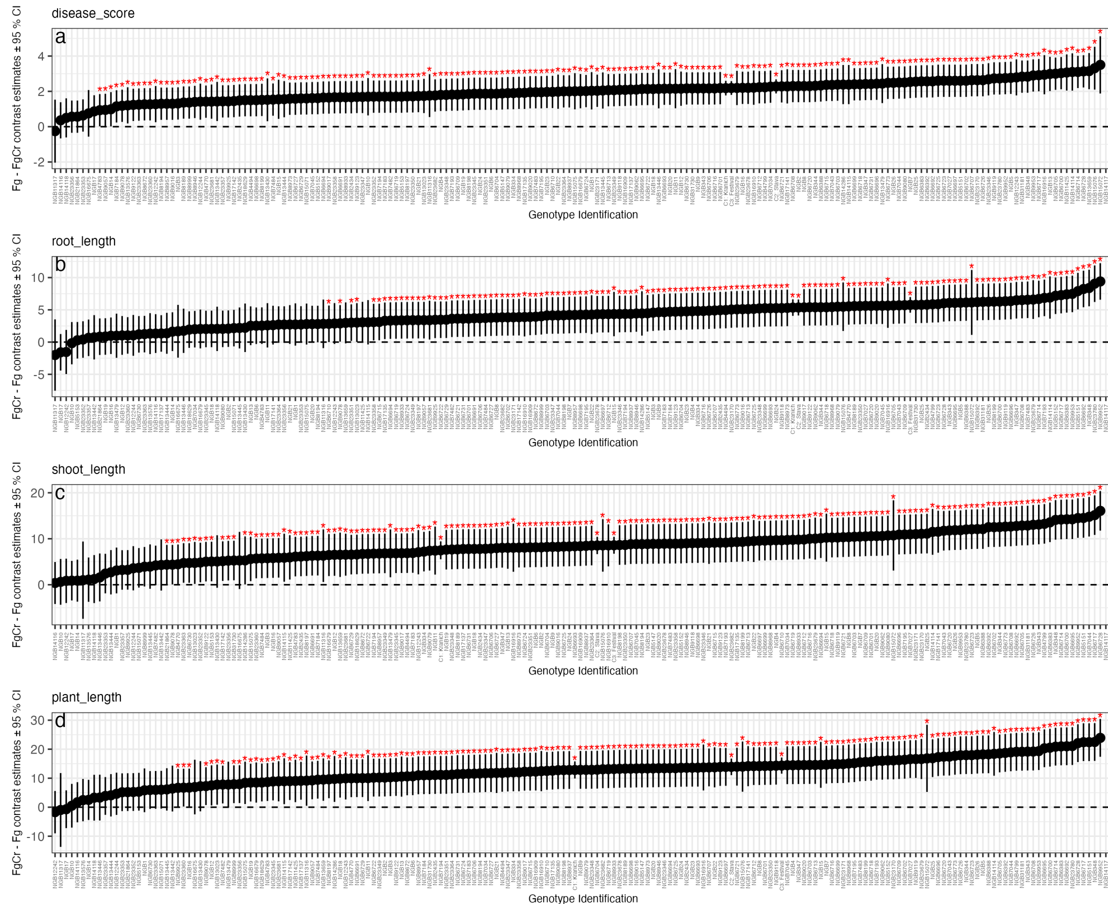


Figure 6 Pairwise comparisons between treatments FgCr and Fg for 190 winter wheat genotypes for trait (a) disease score, (b) root length, (c) shoot length and (d) plant length. Each data point represents a contrast estimate for a specific winter wheat genotype. The vertical lines extending from the points denote the 95% confidence intervals around the contrast estimates. Data points annotated with a red asterisk (*) are statistically significant ($p < 0.05$) and show a non-zero contrast. Note: Genotypes presented are not in the same order across the four plots.

4.5. Correlation between *C. rosea* efficacy and *F. graminearum*

A correlation analysis was conducted between *C. rosea* efficacy, as previously estimated through the pairwise contrast comparison of the treatments, and the treatment *F. graminearum*. This analysis revealed significant correlations ($p < 0.05$) for all traits (Figure 7). Negative correlations were found between the treatment Fg and the *C. rosea* efficacy for root length (R = -0.77, $p < 0.05$), shoot length (R = -0.68, $p < 0.05$), and plant length (R = -0.74, $p < 0.05$) (Figure 7a – 7c). The correlations suggest that plants with lower growth in the Fg treatment benefited more from the treatment with *C. rosea*. In disease score, a moderate positive correlation (R = 0.57, $p < 0.05$), showing that as values in disease score increased, biocontrol efficacy also increased (Figure 7d). This suggests that biocontrol efficacy increased in susceptible genotypes.

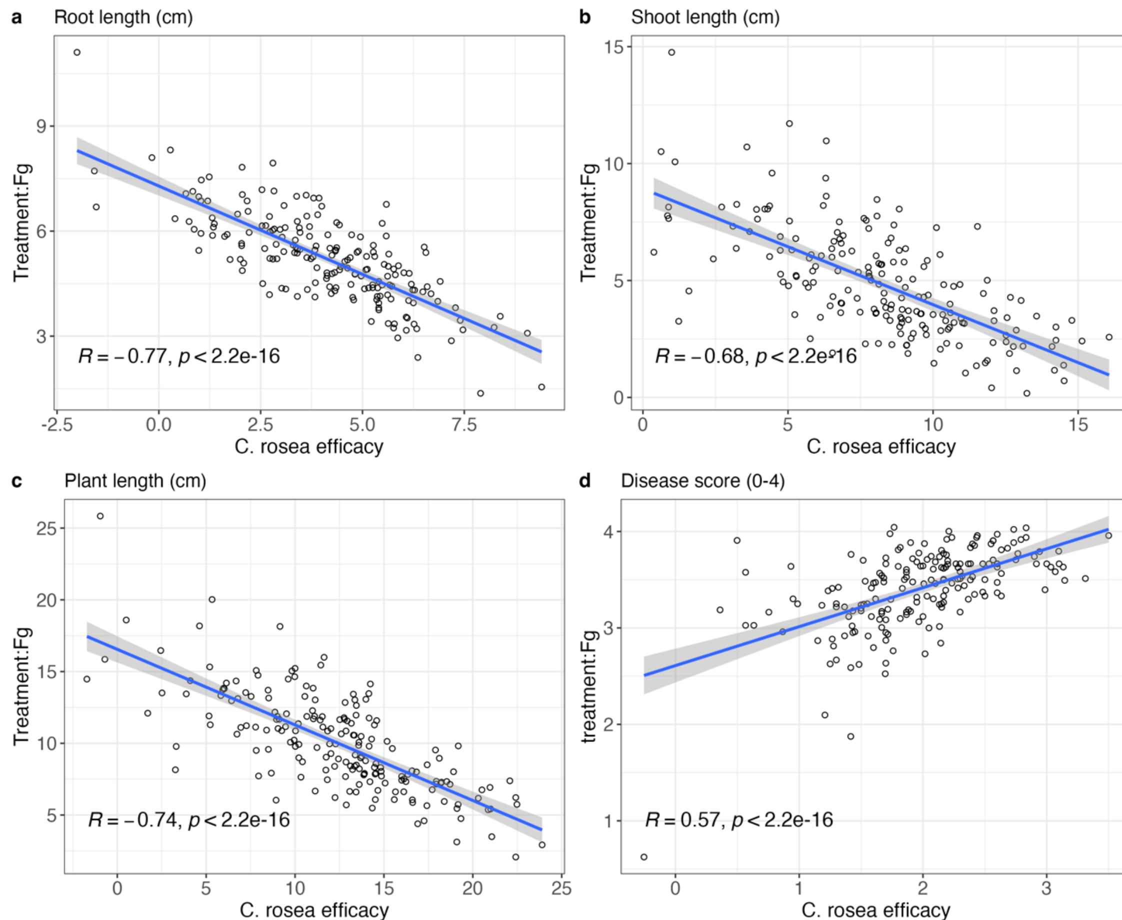


Figure 7 Pearson's correlation between *C. rosea* efficacy and treatment *F. graminearum* (Fg) for the traits (a) root length, (b) shoot length, (c) plant length and (d) disease score. Each data point represents the inter treatment contrast (Fg – FgCr or FgCr – Fg) and BLUEs of genotypes in treatment Fg.

4.6. Genome-wide associations studies

Phenotypic evaluations of genotypes subjected to both Fg and FgCr treatments were conducted, along with pairwise comparisons to determine the efficacy of *C. rosea* across each trait: plant length, shoot length, root length, and disease score were subjected to genome-wide association studies (GWAS). Significance for genome-wide associations between markers and traits was determined at a threshold of $P \leq 0.00014$, adjusted from $P \leq 1/n$, where n = 7360 representing the number of SNP markers. Analysis was performed for 181 genotypes for treatment-level associations and for 180 genotypes were for *C. rosea* efficacy. For trait disease score in the treatment Fg, five significant marker-trait associations were observed i.e. Excalibur_c7026_2635 on chromosome 1A at 53 cM, BS00089497_51 on chromosome 2A at 115 cM and Kukri_c40121_373 at 116 cM, BS00096604_51 and RFL_Contig2459_2314 at chromosome 4B at 71 cM and 73 cM, respectively (Figure 8a, Supplementary table 2). No significant marker-trait association was found for disease score in treatment FgCr (Figure 8b). For disease score contrast or *C. rosea* biocontrol efficacy, seven significant marker-trait

associations were observed, of which four markers i.e. Excalibur_rep_c111629_239, wsnp_Ex_rep_c109138_92064554, BobWhite_c3564_81 and BS00021972_51 on chromosome 7B at 77 cM and three markers i.e. BS00010557_51, wsnp_Ku_rep_c68953_68153061and Kukri_c20611_293 at 78 cM (Figure 8c, Supplementary table 2).

For trait plant length contrast, only one significant marker trait association was observed i.e. Ra_c956_2318 in chromosome 7A at 228 cM (Supplementary table 2, Supplementary Figure 2c). For trait shoot length at treatment FgCr, one significant marker trait association was observed i.e. Tdurum_contig10036_474 at chromosome 1B at 112 cM (Supplementary table 2, Supplementary Figure 3b). For trait root length at treatment Fg, one significant marker trait association was observed i.e. Tdurum_contig15235_951 at chromosome 6B at 65 cM (Supplementary table 2, Supplementary Figure 4a).

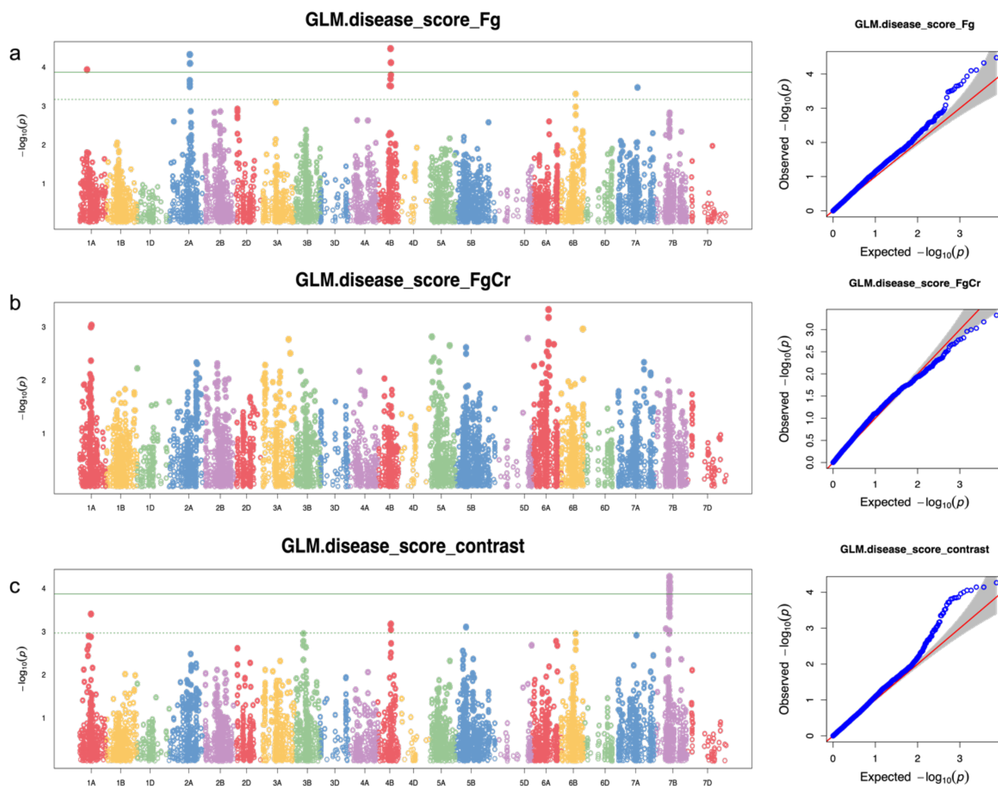


Figure 8 Manhattan plot and corresponding QQ plots showing the association of SNP markers with the FFR disease score in treatment Fg (a), treatment FgCr (b) and disease score contrast or biocontrol efficacy (c). The solid horizontal line shows the significance threshold of −log10(P) = 3.85.

4.7. Analysis of results of *T. afroharzianum*

Similar to *C. rosea* large bioassay, an ANOVA was conducted to assess the influence of the effects of treatment, genotype (nordID), and their interaction on traits root length, shoot length, plant length, and disease score across 20 winter wheat genotypes (Supplementary table 1). Across the traits, normality, and homoscedasticity assumptions were satisfied following a visual inspection, where data points in QQ plots closely matched the diagonal line, indicating normality and also, homoscedasticity was marked by the fitted vs. residual plots, which showed a random scatter (Supplementary Figure 5)

Plant genotpyes (nordID) exhibited a significant effect across multiple traits, including disease score ($F = 2.03$, $p < 0.05$), shoot length ($F = 1.68$, $p < 0.05$), and plant length ($F = 1.85$, $p < 0.05$). However, no statistically significant difference was found among plant genotypes for root length ($F = 1.56$, $p > 0.05$). The effect of treatment was not significant for the traits, disease score ($F = 1.20$, $p > 0.05$), root length ($F = 0.07$, $p > 0.05$), shoot length ($F = 4.35$, $p > 0.05$), and plant length ($F = 0.02$, $p > 0.05$). Overall, the mean values in each treatment were similar within all traits (Table 2). Additionally, the interaction between treatment and genotype (nordID) was not significant ($p > 0.05$) for any of the traits, indicating no genotype-specific effects of *T. afroharzianum* against *F. graminearum* (Table 2).

Table 2 Two-way ANOVA test showing F-values and level of significance for traits. Significance indicator denoting the level of significance of the p-value (* indicates $p < 0.05$, and absence of asterisk indicates non-significance).

Trait	**Source of variation**	**DF**	**SumsOfSqs**	**MeanSqs**	**F - value**	***P* - value**	***p* <0,05**
Disease score (0-4)	BL	7	9.95	1.42	3.69	0.0007	*
	T	1	0.46	0.46	1.20	0.27	
	G	19	14.86	0.78	2.03	0.01	*
	GxT	19	8.19	0.43	1.12	0.33	
	Residuals	272	104.66	0.38			
Root length (cm)	BL	7	187.36	26.77	3.97	0.0003	*
	T	1	0.47	0.49	0.07	0.79	
	G	19	205.13	10.80	1.60	0.06	
	GxT	19	85.85	4.52	0.67	0.85	
	Residuals	273	1842.53	6.75			
Shoot length (cm)	BL	7	102.55	14.65	1.31	0.25	
	T	1	48.63	48.63	4.35	0.04	*
	G	19	356.55	18.77	1.68	0.04	*

	GxT	19	231.68	12.19	1.09	0.36	
	Residuals	264	2951.90	11.18			
Plant length (cm)	BL	7	617.18	88.17	3.27	0.002	*
	T	1	0.52	0.52	0.02	0.89	
	G	19	947.40	49.86	1.85	0.02	*
	GxT	19	597.77	31.46	1.17	0.29	
	Residuals	273	7358.95	26.96			

BL: Block, G: Genotype, T: Treatment, GxT: Genotype X Treatment interaction

DF: Degrees of freedom

5. Discussion

Root and stem base rot diseases are one of the major limiting factors for wheat production, particularly under high pathogen pressure (Poole et al., 2015). Fusarium foot rot (FFR), caused by *Fusarium graminearum*, is one of the most common and serious wheat diseases worldwide (Özer et al., 2023). *Clonostachys rosea* is a mycoparasitic fungus capable of attacking many important plant pathogens, such as *F. graminearum* in the rhizosphere and phyllosphere (Gimeno et al., 2019). In the present study, an *in vivo* bioassay was performed on 190 winter wheat genotypes to investigate the variation in resistance towards FFR and variation in responsiveness of *C. rosea* IK726 biocontrol during fusarium foot rot infection.

The process of plant-pathogen interaction is well known and includes signal activation that can trigger a quick defensive reaction against a variety of plant pathogens. This reaction aids in preventing additional disease infection in the host plant (Gururani et al., 2012). Resistance traits are broadly defined as host characteristics that reduce the extent of pathogen infection (Andersen et al., 2018). Therefore, resistance traits are those that lower pathogen-host contact as well as the rate at which pathogens proliferate after infection (Kover & Schaal, 2002). Disease resistance is a crucial tool for managing diseases (Collinge et al., 2022).

Some studies comparing the dynamics of diseases in monocultures and multiline mixtures showed that low genetic variation can hasten the spread of outbreaks in the field (Garrett & Mundt, 1999; Jordan et al., 1998; Zhu et al., 2000). We observed that the panel of genotypes used had a variation in statistical significance in treatment correlation in the measured traits suggesting that among the 190 genotypes, there is variability in genotypes' response across the treatments to *C. rosea* (FgCr) and *F. graminearum* (Fg) (Figure 4). Highlighting the importance of considering genotype-specific responses when evaluating biocontrol agents.

Several studies have shown that the effectiveness of BCAs in combating pathogens varies depending on the specific genotype of the plant involved (Bazghaleh et al., 2020; Harman et al., 2004; Prashar & Vandenberg, 2017; Tucci et al., 2011). The effectiveness of biological control agents depends on factors such as their host specificity, the genetic structure of the target population, and the overall choice of biocontrol agents (Burdon & Marshall, 1981; Müller-Schärer & Schaffner, 2008). Wheat crop improvement depends on genetic diversity and the use of natural variation for selection using local varieties and wild relatives (Mourad et al., 2019). Diversity in germplasm for disease resistance variation is consistently used in breeding programs. The traditional method of resistance breeding involves screening plant germplasm collections for resistance resources using various pathogen isolates (Li et al., 2021).

It has been widely reported that biocontrol agents can actively induce the defence mechanism in plants (Zehra et al., 2021). The principal biocontrol mechanisms employed by *C. rosea* are induction of plant defence, direct parasitism of pathogenic fungi, antibiosis, production of fungal cell wall degrading enzymes, and plant growth promotion (Chatterton et al., 2009; Hasan et al., 2022; Roberti et al., 2008; Rodriguez et al., 2011). Transcriptomic and exometabolomic profiling of antagonistic and defensive modes of *C. rosea* action against *F. graminearum* has previously shown that *C. rosea* shifts from secondary metabolite-mediated inhibition to cell wall degradation enzymes (CWDEs) upon closer contact with *F. graminearum*. This transition is reflected in the differential expression of genes encoding CWDEs and proteolysis-related enzymes (Demissie et al., 2020). Direct interaction between *C. rosea* and *F. graminearum* is essential for optimal efficacy, potentially explaining the increased suppression of *F. graminearum* before it initiates sexual reproduction (Schöneberg et al., 2015). Different plant genotypes can alter the frequency in the rate, location (e.g., roots or leaves), and timing (young seedlings, flowering, or nearing maturity) of the activation of their defence mechanism (De Vleesschauwer & Höfte, 2009). The exchange of several chemical signals enables the BCA and the plant to specifically recognize each other, leading to alterations in their respective transcriptomes, proteomes, and metabolomes (Kaur & Suseela, 2020). By comparing the differences caused by the interaction between plant and BCA before and after induction with *F. graminearum*, there is a possibility of identifying which genes are involved in the defense response and how they vary between different genotypes. Further investigation into the molecular mechanisms underlying genotype-specific responses in biocontrol agents such as *C. rosea* is important to optimise and increase the effectiveness of these biocontrol approaches against diverse plant pathogens.

A successful strategy for developing *C. rosea* as a biocontrol agent depends significantly on the diversity of plant-associated microbes. This microbial diversity is essential for beneficial fungi that could be harnessed for biocontrol purposes (Johnston-Monje et al., 2021). The rhizosphere is the roots' first defence against pathogenic threats. The soil microbiota within the rhizosphere suppresses pathogens, creating a favourable environment for the establishment and efficacy of biocontrol agents like *C. rosea* (Syed Ab Rahman et al., 2018). The structure of the microbial community in the rhizosphere is influenced by several factors, including plant species and genotypes, as well as external factors like climate and soil type (Landa et al., 2006). These factors must be considered when exploring the potential of *C. rosea* in various agricultural contexts to ensure its effectiveness and adaptability as a BCA.

To evaluate genetic variation in plants for biocontrol compatibility with *C. rosea* in controlling *F. graminearum* we observed a significant reduction of disease severity with *C. rosea* in 180 winter wheat genotypes, showing that *C. rosea* is effective in mitigating FFR infection across multiple winter wheat genotypes. However, *C. rosea* efficacy was not the same in all genotypes across the studied traits, showing that some genotypes are more susceptible to *F. graminearum* than others. This variation emphasises the genetic diversity in winter wheat genotypes regarding their compatibility with *C. rosea* for FFR biocontrol, and also the role of genetic factors in disease susceptibility making certain genotypes have characteristics that make them less vulnerable to FFR disease. We also found a significant positive correlation between disease susceptibility and *C. rosea* biocontrol efficacy, highlighting the performance of *C. rosea* as a BCA in more susceptible genotypes. In line with our research, a study with *Pseudomonas chlororaphis* CP07 strain had a better disease suppression effect on a more susceptible genotype than the more resistant genotypes of *Theobroma cacao* towards *Phytophthora palmivora* (Miguelez-Sierra et al., 2019). This suggests that in genotypes with higher susceptibility, the presence of a BCA serves as a defense mechanism, likely due to the reduced resistance in these genotypes and thus benefit more from the additional protection provided by the biocontrol agent.

Furthermore, when the genotypes were only exposed to *F. graminearum* (Fg) a correlation showed genotype susceptibility to the pathogen leading to a reduction in plant length ($R = -0,82$, $p < 0,05$) compared with the plants treatment with *C. rosea* (FgCr) (Figure 5). Two independent studies showed that the application of *C. rosea* could stimulate both root and above-ground foliage growth in tomato seedlings and root growth and a slight increase in the thickness basal portions of wheat seedlings (Han et al., 2022; Roberti et al., 2008).

In this study, genome-wide association was performed in all traits revealing significant marker-trait associations for disease score, shoot length, root length, and plant length. The SNP marker association was identified at chromosome 7B for disease score, 6B for root length in the presence of the pathogen, 1B for shoot length in the presence of the BCA, and 7A for plant length in contrast. These SNP markers are associated with specific genetic regions in the chromosomes that can influence the phenotypic variation of the trait. GWAS can identify the quantitative trait locus (QTLs) of functional alleles and find resistance through *F. graminearum* associated with FFR. In a previous study, SNP markers associated with resistance to FCR have been found in the same chromosomes as in this study (John & Babu, 2021). Some studies report QTLs for stem browning on chromosomes 1B, and 7A in wheat regarding *F. pseudograminearum* (Bovill et al., 2010; Martin et al., 2015; Rahman et al., 2020). The identification of new QTL for FFR resistance and incorporation of resistance into winter

wheat cultivars is crucial to enhanced disease resistance and produced genetic diversity (Paterson, 2002).

Our analysis of the *Trichoderma afroharzianum* bioassay results revealed that *T. afroharzianum* did not reduce FFR symptoms across all tested winter wheat genotypes. Even though NordID and treatment had significant effects on specific traits, there were no significant interaction effects between treatment and NordID across all assessed traits. This indicates that winter wheat genotypes differ in their susceptibility to FFR disease and compatibility with *C. rosea* and *T. afroharzianum*. In line with your findings, a study was conducted on wheat straws to investigate the potential of *C. rosea* and 10 strains of the genus *Trichoderma* (including T-22) in suppressing perithecia production of three strains of two *Fusarium* spp., *F. graminearum,* and *F. crookwellense*, revealed that simultaneous application of the pathogen and antagonist resulted in a 74% reduction in perithecia by *C. rosea* and a 29% decrease in perithecia formation by *T. afroharzianum* (Schöneberg et al., 2015). Similarly, in our work, we applied both the pathogen and biocontrol agents at the same time. This simultaneous application may explain why we observed similar results, indicating that the timing and method of application could be critical factors in achieving effective disease suppression.

These results have important implications for breeding programmes that try to improve winter wheat varieties' compatibility with biocontrol agents and disease resistance. They stress the significance of combining biocontrol techniques with genetic diversity as effective and dependable ways to manage plant diseases. Effective resistance can be developed by plant breeding or genetic engineering, including the use of novel genomic technologies, even though it is frequently unavailable (Collinge et al., 2022). Through the use of closely linked DNA markers, marker-assisted selection enables the indirect selection of traits and can help further enhance biocompatibility and disease resistance (Benko-Iseppon et al., 2003). This work emphasizes how important it is to take genotype-specific responses into account when assessing biocontrol agents such as *C. rosea*. Additionally, it draws attention to the possibility of utilising genetic variability to maximise the application of biocontrol agents, thereby promoting more resilient and sustainable farming methods.

6. Conclusions and future perspectives

In summary, this thesis underscores the significant challenges posed by root and stem base rot diseases, particularly fusarium foot rot (FFR), to wheat production, while also highlighting the promising potential of *Clonostachys rosea* as a biocontrol agent against FFR. Our study reveals not only variations in winter wheat genotypes' resistance to FFR but also disparities in their compatibility with *C. rosea*, indicating independence between disease resistance and biocontrol compatibility.

Enhancing genetic diversity and employing marker-assisted selection is pivotal in refining wheat breeding approaches to improve disease resistance and biocontrol effectiveness. Further exploration into the molecular mechanisms controlling *C. rosea* biocontrol capabilities is necessary to enhance its efficacy in disease management.

Understanding the reasons behind contrasting genotypic responses to *C. rosea* in FFR-infected plants could provide valuable insights for optimizing biocontrol strategies. Breeding programs should integrate marker-assisted selection to develop improved varieties with higher resistance to FFR and enhanced compatibility with *C. rosea*.

References

Abbas, A., & Yli-Mattila, T. (2022). Biocontrol of *Fusarium graminearum*, a Causal Agent of Fusarium Head Blight of Wheat, and Deoxynivalenol Accumulation: From *In Vitro* to *In Planta*. *Toxins 2022, Vol. 14, Page 299*, *14*(5), 299. https://doi.org/10.3390/TOXINS14050299

Abuamsha, R., Salman, M., & Ehlers, R. U. (2011). Differential Resistance of Oilseed Rape Cultivars (*Brassica napus* ssp. *oleifera*) to *Verticillium longisporum* Infection is Affected by Rhizosphere Colonisation with Antagonistic Bacteria, *Serratia plymuthica* and *Pseudomonas chlororaphis*. *BioControl*, *56*(1), 101–112. https://doi.org/10.1007/S10526-010-9308-8

Aktar, W., Sengupta, D., & Chowdhury, A. (2009). Impact of Pesticides Use in Agriculture: Their Benefits and Hazards. *Interdisciplinary Toxicology*, *2*(1), 1. https://doi.org/10.2478/V10102-009-0001-7

Alfiky, A., & Weisskopf, L. (2021). Deciphering *Trichoderma*-Plant-Pathogen Interactions for Better Development of Biocontrol Applications. *Journal of Fungi (Basel, Switzerland)*, *7*(1), 1–18. https://doi.org/10.3390/JOF7010061

Ali, A., Kumar, R., Mazákova, J., Maňasová, M., Zouhar, M., & Pánek, M. (2022). Evaluation of the Ability of Seven Active Ingredients of Fungicides to Suppress *Phytophthora cactorum* at Diverse Life Stages, and Variability in Resistance Found Among Isolates. *Journal of Fungi*, *8*(10), 1039. https://doi.org/10.3390/JOF8101039/S1

Alvindia, D. G., & Natsuaki, K. T. (2008). Evaluation of Fungal Epiphytes Isolated from Banana Fruit Surfaces for Biocontrol of Banana crown rot disease. *Crop Protection*, *27*(8), 1200–1207. https://doi.org/10.1016/J.CROPRO.2008.02.007

Anaissie, E., & Nucci, M. (2002). Cutaneous Infection by *Fusarium* species in Healthy and Immunocompromised Hosts: Implications for Diagnosis and Management. *Clinical Infectious Diseases*, *35*(8), 909–920. https://doi.org/10.1086/342328

Andersen, E. J., Ali, S., Byamukama, E., Yen, Y., & Nepal, M. P. (2018). Disease Resistance Mechanisms in Plants. *Genes*, *9*(7). https://doi.org/10.3390/GENES9070339

Aoki, T., O'Donnell, K., & Geiser, D. M. (2014). Systematics of Key Phytopathogenic *Fusarium* species: Current Status and Future Challenges. *Journal of General Plant Pathology*, *80*(3), 189–201. https://doi.org/10.1007/S10327-014-0509-3/FIGURES/1

Argumedo-Delira, R., Gómez-Martínez, M. J., & Mora-Delgado, J. (2022). Plant Growth Promoting Filamentous Fungi and Their Application in the Fertilization of Pastures for Animal Consumption. *Agronomy 2022, Vol. 12, Page 3033*, *12*(12), 3033. https://doi.org/10.3390/AGRONOMY12123033

Arkhipov, A., Carvalhais, L. C., & Schenk, P. M. (2023). PGPR Control *Phytophthora capsici* in Tomato Through Induced Systemic Resistance, Early Hypersensitive Response and Direct Antagonism in a Cultivar-Specific Manner. *European Journal of Plant Pathology 2023 167:4*, *167*(4), 811–832. https://doi.org/10.1007/S10658-023-02734-8

Armanda, D. T., Guinée, J. B., & Tukker, A. (2019). The Second Green Revolution: Innovative Urban Agriculture's Contribution to Food Security and Sustainability – A Review. *Global Food Security*, *22*, 13–24. https://doi.org/10.1016/J.GFS.2019.08.002

Ayaz, M., Ali, Q., Farzand, A., Khan, A. R., Ling, H., & Gao, X. (2021). Nematicidal Volatiles from *Bacillus atrophaeus* GBSC56 Promote Growth and Stimulate Induced Systemic Resistance in Tomato against *Meloidogyne incognita*. *International Journal of Molecular Sciences*, *22*(9). https://doi.org/10.3390/IJMS22095049

Ayaz, M., Li, C. H., Ali, Q., Zhao, W., Chi, Y. K., Shafiq, M., Ali, F., Yu, X. Y., Yu, Q., Zhao, J. T., Yu, J. W., Qi, R. De, & Huang, W. K. (2023). Bacterial and Fungal Biocontrol Agents for Plant Disease Protection: Journey from Lab to Field, Current Status, Challenges, and Global Perspectives. *Molecules*, *28*(18). https://doi.org/10.3390/MOLECULES28186735

Backhouse, D., & Burgess, L. W. (2002). Climatic Analysis of the Distribution of *Fusarium graminearum*, *F. pseudograminearum* and *F. culmorum* on Cereals in Australia. *Australasian Plant Pathology*, *31*(4), 321–327. https://doi.org/10.1071/AP02026

Barzman, M., Bàrberi, P., Birch, A. N. E., Boonekamp, P., Dachbrodt-Saaydeh, S., Graf, B., Hommel, B., Jensen, J. E., Kiss, J., Kudsk, P., Lamichhane, J. R., Messéan, A., Moonen, A. C., Ratnadass, A., Ricci, P., Sarah, J. L., & Sattin, M. (2015). Eight Principles of Integrated Pest Management. *Agronomy for Sustainable Development*, *35*(4), 1199–1215. https://doi.org/10.1007/S13593-015-0327-9

Bazghaleh, N., Prashar, P., Woo, S., & Vandenberg, A. (2020). Effects of Lentil Genotype on the Colonization of Beneficial *Trichoderma* Species and Biocontrol of Aphanomyces Root Rot. *Microorganisms 2020, Vol. 8, Page 1290*, *8*(9), 1290. https://doi.org/10.3390/MICROORGANISMS8091290

Becker, R. A., Chambers, J. M., & Wilks, A. Reeve. (1988). *The new S language : A Programming Environment for Data Analysis and Graphics*. 702.

Benko-Iseppon, A. M., Winter, P., Huettel, B., Staginnus, C., Muehlbauer, F. J., & Kahl, G. (2003). Molecular Markers Closely Linked to *Fusarium* Resistance Genes in Chickpea Show Significant Alignments to Pathogenesis-Related Genes Located on Arabidopsis Chromosomes 1 and 5. *TAG. Theoretical and Applied Genetics. Theoretische Und Angewandte Genetik*, *107*(2), 379–386. https://doi.org/10.1007/S00122-003-1260-X

Bovill, W. D., Horne, M., Herde, D., Davis, M., Wildermuth, G. B., & Sutherland, M. W. (2010). Pyramiding QTL Increases Seedling Resistance to crown rot (*Fusarium pseudograminearum*) of Wheat (*Triticum aestivum*). *Theoretical and Applied Genetics*, *121*(1), 127–136. https://doi.org/10.1007/S00122-010-1296-7

Burdon, J. J., & Marshall, D. R. (1981). Biological Control and the Reproductive Mode of Weeds. *The Journal of Applied Ecology*, *18*(2), 649. https://doi.org/10.2307/2402423

Burketova, L., Trda, L., Ott, P. G., & Valentova, O. (2015). Bio-Based Resistance Inducers for Sustainable Plant Protection Against Pathogens. *Biotechnology Advances*, *33*(6), 994–1004. https://doi.org/10.1016/J.BIOTECHADV.2015.01.004

Chakraborty, S., Liu, C. J., Mitter, V., Scott, J. B., Akinsanmi, O. A., Ali, S., Dill-Macky, R., Nicol, J., Backhouse, D., & Simpfendorfer, S. (2006). Pathogen Population Structure and Epidemiology are Keys to Wheat crown rot and Fusarium Head Blight Management. *Australasian Plant Pathology*, *35*(6), 643–655. https://doi.org/10.1071/AP06068

Chaloner, T. M., Gurr, S. J., & Bebber, D. P. (2021). Plant Pathogen Infection Risk Tracks Global Crop Yields Under Climate Change. *Nature Climate Change 2021 11:8*, *11*(8), 710–715. https://doi.org/10.1038/s41558-021-01104-8

Chatterton, S., & Punja, Z. K. (2010). Factors Influencing Colonization of Cucumber Roots by *Clonostachys rosea f. catenulata*, a Biological Disease Control Agent. *Biocontrol Science and Technology*, *20*(1), 37–55. https://doi.org/10.1080/09583150903350253

Chatterton, S., Punja, Z. K., & Chatterton, S. (2009). Chitinase and beta-1,3-glucanase Enzyme Production by the Mycoparasite *Clonostachys rosea f. catenulata* Against Fungal Plant Pathogens. *Canadian Journal of Microbiology*, *55*(4), 356–367. https://doi.org/10.1139/W08-156

Chen, Y., Kistler, H. C., & Ma, Z. (2019). *Fusarium graminearum* Trichothecene Mycotoxins: Biosynthesis, Regulation, and Management. *Annual Review of Phytopathology*, *57*, 15–39. https://doi.org/10.1146/ANNUREV-PHYTO-082718-100318

Chin, T., Pleskach, K., Tittlemier, S. A., Henriquez, M. A., Bamforth, J., Withana Gamage, N., Ashfaq, T., Lee, S. J., Kurera, M. S., Patel, B., & Walkowiak, S. (2023). A Status Update on fusarium head blight on Western Canadian Wheat. *Canadian Journal of Plant Pathology*, *45*(3), 277–289. https://doi.org/10.1080/07060661.2023.2177352

Collinge, D. B., Jensen, D. F., Rabiey, M., Sarrocco, S., Shaw, M. W., & Shaw, R. H. (2022). Biological Control of Plant Diseases – What has been Achieved and What is the Direction? *Plant Pathology*, *71*(5), 1024–1047. https://doi.org/10.1111/PPA.13555

Costa, L. B., Rangel, D. E. N., Morandi, M. A. B., & Bettiol, W. (2012). Impact of UV-B Radiation on *Clonostachys rosea* Germination and Growth. *World Journal of Microbiology & Biotechnology*, *28*(7), 2497–2504. https://doi.org/10.1007/S11274-012-1057-7

Cota, L. V., Maffia, L. A., Mizubuti, E. S. G., & Macedo, P. E. F. (2009). Biological Control by *Clonostachys rosea* as a Key Component in the Integrated Management of Strawberry Gray Mold. *Biological Control*, *50*(3), 222–230. https://doi.org/10.1016/J.BIOCONTROL.2009.04.017

Cowger, C., Professor, A., & Sutton, A. L. (2018). The Southeastern U.S. Fusarium Head Blight Epidemic of 2003. *Https://Doi.Org/10.1094/PHP-2005-1026-01-RS*, *6*(1). https://doi.org/10.1094/PHP-2005-1026-01-RS

Daguerre, Y., Siegel, K., Edel-Hermann, V., & Steinberg, C. (2014). Fungal Proteins and Genes Associated with Biocontrol Mechanisms of Soil-Borne Pathogens: A Review. *Fungal Biology Reviews*, *28*(4), 97–125. https://doi.org/10.1016/j.fbr.2014.11.001

De Vleesschauwer, D., & Höfte, M. (2009). Chapter 6 Rhizobacteria-Induced Systemic Resistance. *Advances in Botanical Research*, *51*(C), 223–281. https://doi.org/10.1016/S0065-2296(09)51006-3

Dean, R., Van Kan, J. A. L., Pretorius, Z. A., Hammond-Kosack, K. E., Di Pietro, A., Spanu, P. D., Rudd, J. J., Dickman, M., Kahmann, R., Ellis, J., & Foster, G. D. (2012). The Top 10 Fungal Pathogens in Molecular Plant Pathology. *Molecular Plant Pathology*, *13*(4), 414–430. https://doi.org/10.1111/J.1364-3703.2011.00783.X

Del Ponte, E. M., Moreira, G. M., Ward, T. J., O'Donnell, K., Nicolli, C. P., Machado, F. J., Duffeck, M. R., Alves, K. S., Tessmann, D. J., Waalwijk, C., van der Lee, T., Zhang, H., Chulze, S. N., Stenglein, S. A., Pan, D., Vero, S., Vaillancourt, L. J., Schmale, D. G., Esker, P. D., … Lee, T. (2022). *Fusarium graminearum* Species Complex: A Bibliographic Analysis and Web-Accessible Database for Global Mapping of Species and Trichothecene Toxin Chemotypes. *Phytopathology*, *112*(4), 741–751. https://doi.org/10.1094/PHYTO-06-21-0277-RVW

Demissie, Z. A., Witte, T., Robinson, K. A., Sproule, A., Foote, S. J., Johnston, A., Harris, L. J., Overy, D. P., & Loewen, M. C. (2020). Transcriptomic and Exometabolomic Profiling Reveals Antagonistic and Defensive Modes of *Clonostachys rosea* Action against Fusarium graminearum. *Molecular Plant-Microbe Interactions*, *33*(6), 842–858. https://doi.org/10.1094/MPMI-11-19-0310-R

Druzhinina, I. S., Seidl-Seiboth, V., Herrera-Estrella, A., Horwitz, B. A., Kenerley, C. M., Monte, E., Mukherjee, P. K., Zeilinger, S., Grigoriev, I. V., & Kubicek, C. P. (2011). *Trichoderma*: The Genomics of Opportunistic Success. *Nature Reviews. Microbiology*, *9*(10), 749–759. https://doi.org/10.1038/NRMICRO2637

Dudeja, S., Chhokar, V., Beniwal, V., Badgujjar, H., Chauhan, R., Soni, S., & Kumar, A. (2021). Optimization and Production of Antimicrobial Compounds by *Aspergillus flavus* MTCC 13062 and its Synergistic Studies. *Biocatalysis and Agricultural Biotechnology*, *35*, 102065. https://doi.org/10.1016/J.BCAB.2021.102065

Eilenberg, J., Hajek, A., & Lomer, C. (2001). Suggestions for Unifying the Terminology in Biological Control. *BioControl*, *46*(4), 387–400. https://doi.org/10.1023/A:1014193329979

Ekwomadu, T. I., & Mwanza, M. (2023). *Fusarium* Fungi Pathogens, Identification, Adverse Effects, Disease Management, and Global Food Security: A Review of the Latest Research. *Agriculture 2023, Vol. 13, Page 1810*, *13*(9), 1810. https://doi.org/10.3390/AGRICULTURE13091810

El-Komy, M. H., Saleh, A. A., Eranthodi, A., & Molan, Y. Y. (2015). Characterization of Novel *Trichoderma asperellum* Isolates to Select Effective Biocontrol Agents Against Tomato Fusarium Wilt. *The Plant Pathology Journal*, *31*(1), 50–60. https://doi.org/10.5423/PPJ.OA.09.2014.0087

Elnahal, A. S. M., El-Saadony, M. T., Saad, A. M., Desoky, E. S. M., El-Tahan, A. M., Rady, M. M., AbuQamar, S. F., & El-Tarabily, K. A. (2022). The Use of Microbial Inoculants for Biological Control, Plant Growth Promotion, and Sustainable Agriculture: A Review.

European Journal of Plant Pathology, *162*(4), 759–792. https://doi.org/10.1007/S10658-021-02393-7

El-Saadony, M. T., Saad, A. M., Soliman, S. M., Salem, H. M., Ahmed, A. I., Mahmood, M., El-Tahan, A. M., Ebrahim, A. A. M., Abd El-Mageed, T. A., Negm, S. H., Selim, S., Babalghith, A. O., Elrys, A. S., El-Tarabily, K. A., & AbuQamar, S. F. (2022). Plant Growth-Promoting Microorganisms as Biocontrol Agents of Plant Diseases: Mechanisms, Challenges and Future Perspectives. *Frontiers in Plant Science*, *13*. https://doi.org/10.3389/FPLS.2022.923880

Evans, J., Levesque, D., De Lahunta, A., & Jensen, H. E. (2004). Intracranial Fusariosis: A Novel Cause of Fungal Meningoencephalitis in a Dog. *Veterinary Pathology*, *41*(5), 510–514. https://doi.org/10.1354/VP.41-5-510

Everitt, B. (1992). Book reviews : Chambers JM, Hastie TJ eds 1992: Statistical Models in S. California: Wadsworth and Brooks/Cole. ISBN 0 534 16765-9. *1*(2), 220–221. https://doi.org/10.1177/096228029200100208

FAO. (2022). Pesticides Use, Pesticides Trade and Pesticides Indicators. In *Pesticides use, pesticides trade and pesticides indicators* (Vol. 46). FAO. https://doi.org/10.4060/CC0918EN

Fatema, U., Broberg, A., Jensen, D. F., Karlsson, M., & Dubey, M. (2018). Functional Analysis of Polyketide Synthase Genes in the Biocontrol Fungus *Clonostachys rosea*. *Scientific Reports*, *8*(1). https://doi.org/10.1038/S41598-018-33391-1

Fenik, J., Tankiewicz, M., & Biziuk, M. (2011). Properties and Determination of Pesticides in Fruits and Vegetables. *TrAC Trends in Analytical Chemistry*, *30*(6), 814–826. https://doi.org/10.1016/J.TRAC.2011.02.008

Fira, D., Dimkić, I., Berić, T., Lozo, J., & Stanković, S. (2018). Biological Control of Plant Pathogens by *Bacillus* Species. *Journal of Biotechnology*, *285*, 44–55. https://doi.org/10.1016/J.JBIOTEC.2018.07.044

Fravel, D. R. (2005). Commercialization and Implementation of Biocontrol. *Annual Review of Phytopathology*, *43*, 337–359. https://doi.org/10.1146/ANNUREV.PHYTO.43.032904.092924

Gao, S., Zheng, Z., Hu, H., Jiang, Y., Liu, M., Stiller, J., Zhou, M., & Liu, C. (2020). Delineating a Locus Conferring Fusarium crown rot Resistance on Chromosome arm 1HL in Barley by Developing and Analysing a Large Population Derived from Near Isogenic Lines. *The Crop Journal*, *8*(6), 1002–1010. https://doi.org/10.1016/J.CJ.2020.03.008

Garrett, K. A., & Mundt, C. C. (1999). Epidemiology in Mixed Host Populations. *Phytopathology*, *89*(11), 984–990. https://doi.org/10.1094/PHYTO.1999.89.11.984

Ghorbanpour, M., Omidvari, M., Abbaszadeh-Dahaji, P., Omidvar, R., & Kariman, K. (2018). Mechanisms Underlying the Protective Effects of Beneficial Fungi Against Plant Diseases. *Biological Control*, *117*, 147–157. https://doi.org/10.1016/J.BIOCONTROL.2017.11.006

Ghozlan, M. H., EL-Argawy, E., Tokgöz, S., Lakshman, D. K., Mitra, A., Ghozlan, M. H., EL-Argawy, E., Tokgöz, S., Lakshman, D. K., & Mitra, A. (2020). Plant Defense against Necrotrophic Pathogens. *American Journal of Plant Sciences*, *11*(12), 2122–2138. https://doi.org/10.4236/AJPS.2020.1112149

Gimeno, A., Sohlberg, E., Pakula, T., Limnell, J., Keller, B., Laitila, A., & Vogelgsang, S. (2019). TaqMan qPCR for Quantification of *Clonostachys rosea* Used as a Biological Control Agent Against *Fusarium graminearum*. *Frontiers in Microbiology*, *10*(JULY), 464010. https://doi.org/10.3389/FMICB.2019.01627

Goh, C. H., Veliz Vallejos, D. F., Nicotra, A. B., & Mathesius, U. (2013). The Impact of Beneficial Plant-Associated Microbes on Plant Phenotypic Plasticity. *Journal of Chemical Ecology*, *39*(7), 826–839. https://doi.org/10.1007/S10886-013-0326-8

Gohel, V., Singh, A., Vimal, M., Ashwini, P., & Chhatpar, H. (2006). Bioprospecting and Antifungal Potential of Chitinolytic Microorganisms. *African Journal of Biotechnology*, *5*(2), 54–72. https://www.ajol.info/index.php/ajb/article/view/137726

Goswami, R. S., & Kistler, H. C. (2004). Heading for Disaster: *Fusarium graminearum* on Cereal Crops. *Molecular Plant Pathology*, *5*(6), 515–525. https://doi.org/10.1111/J.1364-3703.2004.00252.X

Guo, Q., Li, Y., Lou, Y., Shi, M., Jiang, Y., Zhou, J., Sun, Y., Xue, Q., & Lai, H. (2019). *Bacillus amyloliquefaciens* Ba13 Induces Plant Systemic Resistance and Improves Rhizosphere Microecology Against Tomato Yellow Leaf Curl Virus Disease. *Applied Soil Ecology*, *137*, 154–166. https://doi.org/10.1016/J.APSOIL.2019.01.015

Gurr, G. M., & You, M. (2016). Conservation Biological Control of Pests in the Molecular Era: New Opportunities to Address Old Constraints. *Frontiers in Plant Science*, *6*(JAN2016), 171328. https://doi.org/10.3389/FPLS.2015.01255

Gururani, M. A., Venkatesh, J., Upadhyaya, C. P., Nookaraju, A., Pandey, S. K., & Park, S. W. (2012). Plant Disease Resistance Genes: Current Status and Future Directions. *Physiological and Molecular Plant Pathology*, *78*, 51–65. https://doi.org/10.1016/J.PMPP.2012.01.002

Hagerty, C. H., Irvine, T., Rivedal, H. M., Yin, C., & Kroese, D. R. (2021a). Diagnostic Guide: Fusarium Crown Rot of Winter Wheat. *Plant Health Progress*, *22*(2), 176–181. https://doi.org/10.1094/PHP-10-20-0091-DG

Hagerty, C. H., Irvine, T., Rivedal, H. M., Yin, C., & Kroese, D. R. (2021b). Diagnostic Guide: Fusarium Crown Rot of Winter Wheat. *Plant Health Progress*, *22*(2), 176–181. https://doi.org/10.1094/PHP-10-20-0091-DG

Han, P., Zhang, X., Xu, D., Zhang, B., Lai, D., & Zhou, L. (2020). Metabolites from *Clonostachys* Fungi and Their Biological Activities. *Journal of Fungi*, *6*(4), 1–30. https://doi.org/10.3390/JOF6040229

Han, Z., Ghanizadeh, H., Zhang, H., Li, X., Li, T., Wang, Q., Liu, J., & Wang, A. (2022). *Clonostachys rosea* Promotes Root Growth in Tomato by Secreting Auxin Produced through the Tryptamine Pathway. *Journal of Fungi*, *8*(11), 1166. https://doi.org/10.3390/JOF8111166

Hao, J. J., Xie, S. N., Sun, J., Yang, G. Q., Liu, J. Z., Xu, F., Ru, Y. Y., & Song, Y. L. (2017). Analysis of *Fusarium graminearum* Species Complex From Wheat–Maize Rotation Regions in Henan (China). *Plant Disease*, *101*(5), 720–725. https://doi.org/10.1094/PDIS-06-16-0912-RE

Harman, G. E., Petzoldt, R., Comis, A., & Chen, J. (2004). Interactions Between *Trichoderma harzianum* Strain T22 and Maize Inbred Line Mo17 and Effects of These Interactions on Diseases Caused by *Pythiuin ultimum* and *Colletotrichum graminicola*. *Phytopathology*, *94*(2), 147–153. https://doi.org/10.1094/PHYTO.2004.94.2.147

Hasan, R., Lv, B., Uddin, M. J., Chen, Y., Fan, L., Sun, Z., Sun, M., & Li, S. (2022). Monitoring Mycoparasitism of *Clonostachys rosea* against *Botrytis cinerea* Using GFP. *Journal of Fungi*, *8*(6). https://doi.org/10.3390/JOF8060567

Hazen, R. F., & Blum, M. J. (2016). Host Genetic Variation and Microenvironment Shape an Emergent Plant–Antagonist Interaction. *Evolutionary Ecology*, *30*(6), 1043–1060. https://doi.org/10.1007/S10682-016-9868-X

He, D. C., He, M. H., Amalin, D. M., Liu, W., Alvindia, D. G., & Zhan, J. (2021). Biological Control of Plant Diseases: An Evolutionary and Eco-Economic Consideration. *Pathogens*, *10*(10). https://doi.org/10.3390/PATHOGENS10101311

Heil, M., & Bostock, R. M. (2002). Induced Systemic Resistance (ISR) Against Pathogens in the Context of Induced Plant Defences. *Annals of Botany*, *89*(5), 503. https://doi.org/10.1093/AOB/MCF076

Hermosa, R., Viterbo, A., Chet, I., & Monte, E. (2012). Plant-Beneficial Effects of *Trichoderma* and of its Genes. *Microbiology (Reading, England)*, *158*(Pt 1), 17–25. https://doi.org/10.1099/MIC.0.052274-0

Hollaway, G. J., Evans, M. L., Wallwork, H., Dyson, C. B., & Mckay, A. C. (2013). Yield Loss in Cereals, Caused by *Fusarium culmorum* and *F. pseudograminearum*, Is Related to Fungal DNA in Soil Prior to Planting, Rainfall, and Cereal Type. *Plant Disease*, *97*(7), 977–982. https://doi.org/10.1094/PDIS-09-12-0867-RE

Inch, S., & Gilbert, J. (2003). The Incidence of *Fusarium* Species Recovered from Inflorescences of Wild Grasses in Southern Manitoba. *Canadian Journal of Plant Pathology*, *25*(4), 379–383. https://doi.org/10.1080/07060660309507093

Inglis, G. D., & Kawchuk, L. M. (2002). Comparative degradation of Oomycete, Ascomycete, and Basidiomycete Cell Walls by Mycoparasitic and Biocontrol Fungi. *Canadian Journal of Microbiology*, *48*(1), 60–70. https://doi.org/10.1139/W01-130

Jaklitsch, W. M. (2009). European species of Hypocrea Part I. The green-spored species. *Studies in Mycology*, *63*, 1–91. https://doi.org/10.3114/SIM.2009.63.01

Jamalizadeh, M., Etebarian, H. R., Aminian, H., & Alizadeh, A. (2011). A Review of Mechanisms of Action of Biological Control Organisms Against Post-Harvest Fruit Spoilage. *EPPO Bulletin*, *41*(1), 65–71. https://doi.org/10.1111/J.1365-2338.2011.02438.X

James Cook, R. (1992). Wheat Root Health Management and Environmental Concern. *Canadian Journal of Plant Pathology*, *14*(1), 76–85. https://doi.org/10.1080/07060669209500908

Jansen, C., Von Wettstein, D., Schäfer, W., Kogel, K. H., Felk, A., & Maier, F. J. (2005). Infection Pattern in Barley and Wheat Spikes Inoculated with Wild-Type and Trichodiene Synthase Gene Disrupted Fusarium graminearum. *Proceedings of the National Academy of Sciences of the United States of America*, *102*(46), 16892–16897. https://doi.org/10.1073/pnas.0508467102

Javed, A., Hussain Shah, A., Hussain, A., Shinwari, Z. K., Ali Khan, S., Khan, W., Sohail, A., & Jan, A. (2020). Potential of Endophytic Fungus Aspergillus terreus as Pontent Plant Growth Promoter. *Pak. J. Bot*, *52*(3), 1083–1086. https://doi.org/10.30848/PJB2020-3(44)

Jensen, B., Knudsen, I. M. B., & Jensen, D. F. (2000). Biological Seed Treatment of Cereals with Fresh and Long-Term Stored Formulations of *Clonostachys rosea*: Biocontrol Efficacy Against *Fusarium culmorum*. *European Journal of Plant Pathology*, *106*(3), 233–242. https://doi.org/10.1023/A:1008794626600

Jensen, B., Knudsen, I. M. B., & Jensen, D. F. (2002). Survival of Conidia of *Clonostachys rosea* on Stored Barley Seeds and their Biocontrol Efficacy Against Seed-Borne *Bipolaris sorokiniana*. *Biocontrol Science and Technology*, *12*(4), 427–441. https://doi.org/10.1080/09583150220146013

Jensen, B., Knudsen, I. M. B., Madsen, M., & Jensen, D. F. (2007). Biopriming of Infected Carrot Seed with an Antagonist, *Clonostachys rosea*, Selected for Control of Seedborne *Alternaria* spp. *Https://Doi.Org/10.1094/PHYTO.2004.94.6.551*, *94*(6), 551–560. https://doi.org/10.1094/PHYTO.2004.94.6.551

Jensen, B., Lübeck, P. S., & Jørgensen, H. J. L. (2016). *Clonostachys rosea* Reduces Spot Blotch in Barley by Inhibiting Prepenetration Growth and Sporulation of *Bipolaris sorokiniana* without Inducing Resistance. *Pest Management Science*, *72*(12), 2231–2239. https://doi.org/10.1002/PS.4260

Jeyasri, R., Muthuramalingam, P., Satish, L., Pandian, S. K., Chen, J. T., Ahmar, S., Wang, X., Mora-Poblete, F., & Ramesh, M. (2021). An Overview of Abiotic Stress in Cereal Crops: Negative Impacts, Regulation, Biotechnology and Integrated Omics. *Plants 2021, Vol. 10, Page 1472*, *10*(7), 1472. https://doi.org/10.3390/PLANTS10071472

Jin, F., Hu, Q., Zhao, Y., Lin, X., Zhang, J., & Zhang, J. (2022). Enhancing Quinoa Growth Under Severe Saline-Alkali Stress by Phosphate Solubilizing Microorganism *Penicillium funicuiosum* P1. *PLOS ONE*, *17*(9), e0273459. https://doi.org/10.1371/JOURNAL.PONE.0273459

John, D. A., & Babu, G. R. (2021). Lessons From the Aftermaths of Green Revolution on Food System and Health. *Frontiers in Sustainable Food Systems*, *5*, 644559. https://doi.org/10.3389/FSUFS.2021.644559

Johnston-Monje, D., Gutiérrez, J. P., & Lopez-Lavalle, L. A. B. (2021). Seed-Transmitted Bacteria and Fungi Dominate Juvenile Plant Microbiomes. *Frontiers in Microbiology*, *12*, 737616. https://doi.org/10.3389/FMICB.2021.737616

Jordan, D. R., Tao, Y. Z., Godwin, I. D., Henzell, R. G., Cooper, M., & McIntyre, C. L. (1998). Loss of Genetic Diversity Associated with Selection for Resistance to Sorghum Midge in Australian Sorghum. *Euphytica*, *102*(1), 1–7. https://doi.org/10.1023/A:1018311908636

Kabak, B., Dobson, A. D. W., & Var, I. (2006). Strategies to Prevent Mycotoxin Contamination of Food and Animal Feed: A Review. *Critical Reviews in Food Science and Nutrition*, *46*(8), 593–619. https://doi.org/10.1080/10408390500436185

Kadkol, G. P., Meza, J., Simpfendorfer, S., Harden, S., & Cullis, B. (2021). Genetic Variation for Fusarium Crown Rot Tolerance in Durum Wheat. *PLoS ONE*, *16*(2). https://doi.org/10.1371/JOURNAL.PONE.0240766

Karlovsky, P., Suman, M., Berthiller, F., De Meester, J., Eisenbrand, G., Perrin, I., Oswald, I. P., Speijers, G., Chiodini, A., Recker, T., & Dussort, P. (2016). Impact of Food Processing and Detoxification Treatments on Mycotoxin Contamination. *Mycotoxin Research 2016 32:4*, *32*(4), 179–205. https://doi.org/10.1007/S12550-016-0257-7

Karlsson Green, K., Stenberg, J. A., & Lankinen, Å. (2020). Making Sense of Integrated Pest Management (IPM) in the Light of Evolution. *Evolutionary Applications*, *13*(8), 1791. https://doi.org/10.1111/EVA.13067

Karlsson, I., Mellqvist, E., & Persson, P. (2023). Temporal and Spatial Dynamics of *Fusarium* spp. and Mycotoxins in Swedish Cereals During 16 years. *Mycotoxin Research*, *39*(1), 3–18. https://doi.org/10.1007/S12550-022-00469-9/FIGURES/6

Karlsson, I., Persson, P., & Friberg, H. (2021). Fusarium Head Blight From a Microbiome Perspective. *Frontiers in Microbiology*, *12*, 628373. https://doi.org/10.3389/FMICB.2021.628373

Kaur, S., & Suseela, V. (2020). Unraveling Arbuscular Mycorrhiza-Induced Changes in Plant Primary and Secondary Metabolome. *Metabolites*, *10*(8), 1–30. https://doi.org/10.3390/METABO10080335

Kause, A., & Ødegård, J. (2012). The Genetic Analysis of Tolerance to Infections: A Review. *Frontiers in Genetics*, *3*(DEC). https://doi.org/10.3389/FGENE.2012.00262

Kazan, K., & Gardiner, D. M. (2018). Fusarium Crown Rot Caused by *Fusarium pseudograminearum* in Cereal Crops: Recent Progress and Future Prospects. *Molecular Plant Pathology*, *19*(7), 1547–1562. https://doi.org/10.1111/MPP.12639

Keller, M. D., Bergstrom, G. C., & Shields, E. J. (2014). The Aerobiology of *Fusarium graminearum*. *Aerobiologia*, *30*(2), 123–136. https://doi.org/10.1007/S10453-013-9321-3

Khairullina, A., Micic, N., Jørgensen, H. J. L., Bjarnholt, N., Bülow, L., Collinge, D. B., & Jensen, B. (2023). Biocontrol Effect of *Clonostachys rosea* on *Fusarium graminearum* Infection and Mycotoxin Detoxification in Oat (*Avena sativa*). *Plants*, *12*(3), 500. https://doi.org/10.3390/PLANTS12030500

Khudhair, M., Melloy, P., Lorenz, D. J., Obanor, F., Aitken, E., Datta, S., Luck, J., Fitzgerald, G., & Chakraborty, S. (2014). Fusarium Crown Rot Under Continuous Cropping of Susceptible and Partially Resistant Wheat in Microcosms at Elevated CO2. *Plant Pathology*, *63*(5), 1033–1043. https://doi.org/10.1111/PPA.12182

Knight, N. L., & Sutherland, M. W. (2013). Spread of *Fusarium pseudograminearum* in Wheat Seedling Tissues from a Single Inoculation Point. *Australasian Plant Pathology*, *42*(5), 609–615. https://doi.org/10.1007/S13313-013-0225-Z

Knight, N. L., & Sutherland, M. W. (2016). Histopathological Assessment of *Fusarium pseudograminearum* Colonization of Cereal Culms During Crown Rot Infections. *Plant Disease*, *100*(2), 252–259. https://doi.org/10.1094/PDIS-04-15-0476-RE

Knudsen, I. M. B., Hockenhull, J., & Jensen, D. F. (1995). Biocontrol of Seedling Diseases of Barley and Wheat Caused by *Fusarium culmorum* and *Bipolaris sorokiniana*: Effects of Selected Fungal Antagonists on Growth and Yield Components. *Plant Pathology*, *44*(3), 467–477. https://doi.org/10.1111/J.1365-3059.1995.TB01669.X

Köhl, J., Kolnaar, R., & Ravensberg, W. J. (2019). Mode of Action of Microbial Biological Control Agents Against Plant Diseases: Relevance Beyond Efficacy. *Frontiers in Plant Science*, *10*, 454982. https://doi.org/10.3389/FPLS.2019.00845

Kosawang, C., Karlsson, M., Vélëz, H., Rasmussen, P. H., Collinge, D. B., Jensen, B., & Jensen, D. F. (2014). Zearalenone Detoxification by Zearalenone Hydrolase is Important for the Antagonistic Ability of *Clonostachys rosea* Against Mycotoxigenic *Fusarium graminearum*. *Fungal Biology*, *118*(4), 364–373. https://doi.org/10.1016/J.FUNBIO.2014.01.005

Kover, P. X., & Schaal, B. A. (2002). Genetic Variation for Disease Resistance and Tolerance Among *Arabidopsis thaliana* Accessions. *Proceedings of the National Academy of Sciences of the United States of America*, *99*(17), 11270. https://doi.org/10.1073/PNAS.102288999

Lacey, L. A., Grzywacz, D., Shapiro-Ilan, D. I., Frutos, R., Brownbridge, M., & Goettel, M. S. (2015). Insect Pathogens as Biological Control Agents: Back to the Future. *Journal of Invertebrate Pathology*, *132*, 1–41. https://doi.org/10.1016/j.jip.2015.07.009

Lahlali, R., & Peng, G. (2014). Suppression of clubroot by *Clonostachys rosea* via antibiosis and induced host resistance. *Plant Pathology*, *63*(2), 447–455. https://doi.org/10.1111/PPA.12112

Lamb, C., & Dixon, R. A. (1997). The Oxidative Burst in Plant Disease Resistance. *Annual Review of Plant Physiology and Plant Molecular Biology*, *48*, 251–275. https://doi.org/10.1146/ANNUREV.ARPLANT.48.1.251

Lamichhane, J. R., Osdaghi, E., Behlau, F., Köhl, J., Jones, J. B., & Aubertot, J. N. (2018). Thirteen Decades of Antimicrobial Copper Compounds Applied in Agriculture. A Review. *Agronomy for Sustainable Development 2018 38:3*, *38*(3), 1–18. https://doi.org/10.1007/S13593-018-0503-9

Landa, B. B., Mavrodi, O. V., Schroeder, K. L., Allende-Molar, R., & Weller, D. M. (2006). Enrichment and Genotypic diversity of phlD-containing Fluorescent *Pseudomonas* spp. in Two Soils After a Century of Wheat and Flax Monoculture. *FEMS Microbiology Ecology*, *55*(3), 351–368. https://doi.org/10.1111/J.1574-6941.2005.00038.X

Li, F., Ghanizadeh, H., Song, W., Miao, S., Wang, H., Chen, X., Liu, J., & Wang, A. (2023). Combined Use of *Trichoderma harzianum* and *Clonostachys rosea* to Manage *Botrytis cinerea* Infection in Tomato Plants. *European Journal of Plant Pathology*, *167*(4), 637–650. https://doi.org/10.1007/S10658-023-02732-W

Li, N., Han, X., Feng, D., Yuan, D., & Huang, L. J. (2019). Signaling Crosstalk between Salicylic Acid and Ethylene/Jasmonate in Plant Defense: Do We Understand What They Are Whispering? *International Journal of Molecular Sciences*, *20*(3). https://doi.org/10.3390/IJMS20030671

Li, Q., Wang, B., Yu, J., & Dou, D. (2021). Pathogen-Informed Breeding for Crop Disease Resistance. *Journal of Integrative Plant Biology*, *63*(2), 305–311. https://doi.org/10.1111/JIPB.13029

Lombardi, N., Caira, S., Troise, A. D., Scaloni, A., Vitaglione, P., Vinale, F., Marra, R., Salzano, A. M., Lorito, M., & Woo, S. L. (2020). *Trichoderma* Applications on Strawberry Plants Modulate the Physiological Processes Positively Affecting Fruit Production and Quality. *Frontiers in Microbiology*, *11*, 522584. https://doi.org/10.3389/FMICB.2020.01364

López-Bucio, J., Pelagio-Flores, R., & Herrera-Estrella, A. (2015a). *Trichoderma* as Biostimulant: Exploiting the Multilevel Properties of a Plant Beneficial Fungus. *Scientia Horticulturae*, *196*, 109–123. https://doi.org/10.1016/J.SCIENTA.2015.08.043

López-Bucio, J., Pelagio-Flores, R., & Herrera-Estrella, A. (2015b). *Trichoderma* as biostimulant: exploiting the multilevel properties of a plant beneficial fungus. *Scientia Horticulturae*, *196*, 109–123. https://doi.org/10.1016/J.SCIENTA.2015.08.043

López-Núñez, F. A., Marchante, E., Heleno, R., Duarte, L. N., Palhas, J., Impson, F., Freitas, H., & Marchante, H. (2021). Establishment, Spread and Early Impacts of the First Biocontrol Agent Against an Invasive Plant in Continental Europe. *Journal of Environmental Management*, *290*, 112545. https://doi.org/10.1016/J.JENVMAN.2021.112545

Lorito, M., Woo, S. L., Harman, G. E., & Monte, E. (2010). Translational Research on *Trichoderma*: From 'Omics to the Field. *Annual Review of Phytopathology*, *48*, 395–417. https://doi.org/10.1146/ANNUREV-PHYTO-073009-114314

Ma, L. J., Geiser, D. M., Proctor, R. H., Rooney, A. P., O'Donnell, K., Trail, F., Gardiner, D. M., Manners, J. M., & Kazan, K. (2013). *Fusarium* Pathogenomics. *67*, 399–416. https://doi.org/10.1146/ANNUREV-MICRO-092412-155650

Magan, N., Aldred, D., Mylona, K., & Lambert, R. J. W. (2010). Limiting Mycotoxins in Stored Wheat. *Food Additives and Contaminants*, *27*(5), 644–650. https://doi.org/10.1080/19440040903514523

Makiola, A., Holdaway, R. J., Wood, J. R., Orwin, K. H., Glare, T. R., & Dickie, I. A. (2022). Environmental and Plant Community Drivers of Plant-Pathogen Composition and Richness. *New Phytologist*, *233*(1), 496–504. https://doi.org/10.1111/NPH.17797

Malosetti, M., Zwep, L. B., Forrest, K., van Eeuwijk, F. A., & Dieters, M. (2021). Lessons from a GWAS Study of a Wheat Pre-Breeding Program: Pyramiding Resistance Alleles to Fusarium Crown Rot. *Theoretical and Applied Genetics*, *134*(3), 897–908. https://doi.org/10.1007/S00122-020-03740-8

Mamarabadi, M., Jensen, B., Jensen, D. F., & Lübeck, M. (2008). Real-time RT-PCR expression Analysis of Chitinase and Endoglucanase Genes in the Three-Way Interaction Between the Biocontrol Strain *Clonostachys rosea* IK726, *Botrytis cinerea* and Strawberry. *FEMS Microbiology Letters*, *285*(1), 101–110. https://doi.org/10.1111/J.1574-6968.2008.01228.X

Markell, S. G., & Francl, L. J. (2003). Fusarium Head Blight Inoculum: Species Prevalence and *Gibberella zeae* Spore Type. *Plant Disease*, *87*(7), 814–820. https://doi.org/10.1094/PDIS.2003.87.7.814

Martin, A., Bovill, W. D., Percy, C. D., Herde, D., Fletcher, S., Kelly, A., Neate, S. M., & Sutherland, M. W. (2015). Markers for Seedling and Adult Plant Crown Rot Resistance in Four Partially Resistant Bread Wheat Sources. *Theoretical and Applied Genetics*, *128*(3), 377–385. https://doi.org/10.1007/S00122-014-2437-1

Mascarin, G. M., da Silva, A. V. R., da Silva, T. P., Kobori, N. N., Morandi, M. A. B., & Bettiol, W. (2022). *Clonostachys rosea*: Production by Submerged Culture and Bioactivity Against *Sclerotinia sclerotiorum* and *Bemisia tabaci*. *Frontiers in Microbiology*, *13*, 851000. https://doi.org/10.3389/FMICB.2022.851000

McMullen, M., Bergstrom, G., De Wolf, E., Dill-Macky, R., Hershman, D., Shaner, G., & Van Sanford, D. (2012). A Unified Effort to Fight an Enemy of Wheat and Barley: Fusarium Head Blight. *Plant Disease*, *96*(12), 1712–1728. https://doi.org/10.1094/PDIS-03-12-0291-FE

Meng, F., Lv, R., Cheng, M., Mo, F., Zhang, N., Qi, H., Liu, J., Chen, X., Liu, Y., Ghanizadeh, H., & Wang, A. (2022). Insights Into the Molecular Basis of Biocontrol of *Botrytis cinerea* by *Clonostachys rosea* in Tomato. *Scientia Horticulturae*, *291*, 110547. https://doi.org/10.1016/J.SCIENTA.2021.110547

Mielniczuk, E., & Skwaryło-Bednarz, B. (2020). Fusarium Head Blight, Mycotoxins and Strategies for Their Reduction. *Agronomy 2020, Vol. 10, Page 509*, *10*(4), 509. https://doi.org/10.3390/AGRONOMY10040509

Miguelez-Sierra, Y., Acebo-Guerrero, Y., El Jaziri, M., Bertin, P., & Hernández-Rodríguez, A. (2019). *Pseudomonas chlororaphis* CP07 Strain Reduces Disease Severity Caused by Phytophthora palmivora in Genotypes of *Theobroma cacao*. *European Journal of Plant Pathology*, *155*(4), 1133–1143. https://doi.org/10.1007/S10658-019-01842-8

Morandi, M. A. B., Sutton, J. C., & Maffia, L. A. (2000). Effects of Host and Microbial Factors on Development of *Clonostachys rosea* and Control of *Botrytis cinerea* in Rose. *European Journal of Plant Pathology*, *106*(5), 439–448. https://doi.org/10.1023/A:1008738513748

Morimura, H., Ito, M., Yoshida, S., Koitabashi, M., Tsushima, S., Camagna, M., Chiba, S., Takemoto, D., Kawakita, K., & Sato, I. (2020). *In Vitro* Assessment of Biocontrol Effects on Fusarium Head Blight and Deoxynivalenol (DON) Accumulation by DON-Degrading

Bacteria. *Toxins 2020, Vol. 12, Page 399*, *12*(6), 399. https://doi.org/10.3390/TOXINS12060399

Mouekouba, L. D. O., Zhang, L., Guan, X., Chen, X., Chen, H., Zhang, J., Zhang, J., Li, J., Yang, Y., & Wang, A. (2014). Analysis of *Clonostachys rosea*-Induced Resistance to Tomato Gray Mold Disease in Tomato Leaves. *PLOS ONE*, *9*(7), e102690. https://doi.org/10.1371/JOURNAL.PONE.0102690

Mourad, A. M. I., Alomari, D. Z., Alqudah, A. M., Sallam, A., & Salem, K. F. M. (2019). Recent Advances in Wheat (*Triticum spp.)* Breeding. *Advances in Plant Breeding Strategies: Cereals*, *5*, 559–593. https://doi.org/10.1007/978-3-030-23108-8_15

Mudge, A. M., Dill-Macky, R., Dong, Y., Gardiner, D. M., White, R. G., & Manners, J. M. (2006). A Role for the Mycotoxin deoxynivalenol in Stem Colonisation During Crown Rot Disease of Wheat Caused by *Fusarium graminearum* and *Fusarium pseudograminearum*. *Physiological and Molecular Plant Pathology*, *69*(1–3), 73–85. https://doi.org/10.1016/J.PMPP.2007.01.003

Mukhopadhyay, R., & Kumar, D. (2020). *Trichoderma*: A Beneficial Antifungal Agent and Insights into its Mechanism of Biocontrol Potential. *Egyptian Journal of Biological Pest Control*, *30*(1), 1–8. https://doi.org/10.1186/S41938-020-00333-X/FIGURES/3

Müller-Schärer, H., & Schaffner, U. (2008). Classical Biological Control: Exploiting Enemy Escape to Manage Plant Invasions. *Biological Invasions*, *10*(6), 859–874. https://doi.org/10.1007/S10530-008-9238-X

Murray, G. M., & Brennan, J. P. (2009). Estimating Disease Losses to the Australian Wheat Industry. *Australasian Plant Pathology*, *38*(6), 558–570. https://doi.org/10.1071/AP09053

Muvea, A. M., Meyhöfer, R., Subramanian, S., Poehling, H. M., Ekesi, S., & Maniania, N. K. (2014). Colonization of Onions by Endophytic Fungi and Their Impacts on the Biology of Thrips tabaci. *PLOS ONE*, *9*(9), e108242. https://doi.org/10.1371/JOURNAL.PONE.0108242

Nada, S., Nikola, T., Bozidar, U., Ilija, D., & Andreja, R. (2022). Prevention and Practical Strategies to Control Mycotoxins in the Wheat and Maize Chain. *Food Control*, *136*, 108855. https://doi.org/10.1016/J.FOODCONT.2022.108855

National Academies of Sciences, E. and M. H. and M. D. B. on G. H. F. on M. T., Haag, T., Nicholson, A., & Ashby, E. (2023). Integrated Plant Disease Management. *The Role of Plant Agricultural Practices on Development of Antimicrobial Resistant Fungi Affecting Human Health*. https://doi.org/10.17226/26833

Nelson, P. E., Dignani, M. C., & Anaissie, E. J. (1994). Taxonomy, Biology, and Clinical Aspects of *Fusarium* Species. *Clinical Microbiology Reviews*, *7*(4), 479–504. https://doi.org/10.1128/CMR.7.4.479

Nicolopoulou-Stamati, P., Maipas, S., Kotampasi, C., Stamatis, P., & Hens, L. (2016). Chemical Pesticides and Human Health: The Urgent Need for a New Concept in Agriculture. *Frontiers in Public Health*, *4*, 178764. https://doi.org/10.3389/FPUBH.2016.00148

Nobre, S. A. M., Maffia, L. A., Mizubuti, E. S. G., Cota, L. V., & Dias, A. P. S. (2005). Selection of *Clonostachys rosea* Isolates from Brazilian Ecosystems Effective in Controlling *Botrytis cinerea*. *Biological Control*, *34*(2), 132–143. https://doi.org/10.1016/J.BIOCONTROL.2005.04.011

Odilbekov, F., Armoniené, R., Koc, A., Svensson, J., & Chawade, A. (2019). GWAS-Assisted Genomic Prediction to Predict Resistance to Septoria Tritici Blotch in Nordic Winter Wheat at Seedling Stage. *Frontiers in Genetics*, *10*, 476082. https://doi.org/10.3389/FGENE.2019.01224

O'Donnell, K., Ward, T. J., Robert, V. A. R. G., Crous, P. W., Geiser, D. M., & Kang, S. (2015). DNA Sequence-Based Identification of *Fusarium*: Current Status and Future Directions. *Phytoparasitica*, *43*(5), 583–595. https://doi.org/10.1007/s12600-015-0484-z

Osborne, L. E., & Stein, J. M. (2007). Epidemiology of Fusarium Head Blight on Small-Grain Cereals. *International Journal of Food Microbiology*, *119*(1–2), 103–108. https://doi.org/10.1016/J.IJFOODMICRO.2007.07.032

Oszust, K., Cybulska, J., & Frąc, M. (2020). How Do *Trichoderma* Genus Fungi Win a Nutritional Competition Battle against Soft Fruit Pathogens? A Report on Niche Overlap Nutritional Potentiates. *International Journal of Molecular Sciences*, *21*(12), 1–19. https://doi.org/10.3390/IJMS21124235

Otieno, W., Jeger, M., & Termorshuizen, A. (2003). Effect of Infesting Soil with *Trichoderma harzianum* and Amendment with Coffee Pulp on Survival of Armillaria. *Biological Control*, *26*(3), 293–301. https://doi.org/10.1016/S1049-9644(02)00168-8

Özer, G., Erper, İ., Yıldız, Ş., Bozoğlu, T., Zholdoshbekova, S., Alkan, M., Tekin, F., Uulu, T. E., İmren, M., Dababat, A. A., & Derviş, S. (2023). Fungal Pathogens Associated with Crown and Root Rot in Wheat-Growing Areas of Northern Kyrgyzstan. *Journal of Fungi*, *9*(1), 124. https://doi.org/10.3390/jof9010124

Özkale, E., Yörük, E., Budak, M., & Korkmaz, E. M. (2023). *Trichoderma atroviride* Suppresses *Fusarium graminearum* by Altering Primary and Secondary Metabolite Biosynthesis Profiling. *Plant Pathology*, *72*(8), 1428–1441. https://doi.org/10.1111/PPA.13768

Pacheco-Trejo, J., Aquino-Torres, E., Reyes-Santamaría, M. I., Islas-Pelcastre, M., Pérez-Ríos, S. R., Madariaga-Navarrete, A., & Saucedo-García, M. (2022). Plant Defensive Responses Triggered by *Trichoderma* spp. as Tools to Face Stressful Conditions. *Horticulturae 2022, Vol. 8, Page 1181*, *8*(12), 1181. https://doi.org/10.3390/HORTICULTURAE8121181

Pachenari, A., & Dix, N. J. (1980). Production of Toxins and Wall Degrading Enzymes by *Gliocladium roseum*. *Transactions of the British Mycological Society*, *74*(3), 561–566. https://doi.org/10.1016/S0007-1536(80)80057-X

Palmieri, D., Ianiri, G., Del Grosso, C., Barone, G., De Curtis, F., Castoria, R., & Lima, G. (2022). Advances and Perspectives in the Use of Biocontrol Agents against Fungal Plant Diseases. *Horticulturae 2022, Vol. 8, Page 577*, *8*(7), 577. https://doi.org/10.3390/HORTICULTURAE8070577

Pandit, M. A., Kumar, J., Gulati, S., Bhandari, N., Mehta, P., Katyal, R., Rawat, C. D., Mishra, V., & Kaur, J. (2022). Major Biological Control Strategies for Plant Pathogens. *Pathogens*, *11*(2). https://doi.org/10.3390/PATHOGENS11020273

Parry, D. W., Jenkinson, P., & McLeod, L. (1995). Fusarium Ear Blight (Scab) in Small Grain Cereals—A Review. *Plant Pathology*, *44*(2), 207–238. https://doi.org/10.1111/J.1365-3059.1995.TB02773.X

Pasquali, M., Beyer, M., Logrieco, A., Audenaert, K., Balmas, V., Basler, R., Boutigny, A. L., Chrpová, J., Czembor, E., Gagkaeva, T., González-Jaén, M. T., Hofgaard, I. S., Köycü, N. D., Hoffmann, L., Levic, J., Marin, P., Miedaner, T., Migheli, Q., Moretti, A., … Vogelgsang, S. (2016). A European Database of *Fusarium graminearum* and *F. culmorum trichothecene* Genotypes. *Frontiers in Microbiology*, *7*(APR), 182512. https://doi.org/10.3389/fmicb.2016.00406

Paterson, A. H. (2002). What has QTL Mapping Taught us About Plant Domestication? *New Phytologist*, *154*(3), 591–608. https://doi.org/10.1046/J.1469-8137.2002.00420.X

Pathak, V. M., Verma, V. K., Rawat, B. S., Kaur, B., Babu, N., Sharma, A., Dewali, S., Yadav, M., Kumari, R., Singh, S., Mohapatra, A., Pandey, V., Rana, N., & Cunill, J. M. (2022). Current Status of Pesticide Effects on Environment, Human Health and it's Eco-Friendly Management as Bioremediation: A Comprehensive Review. *Frontiers in Microbiology*, *13*, 962619. https://doi.org/10.3389/fmicb.2022.962619

Paulitz, T. C. (2006). Low Input No-Till Cereal Production in the Pacific Northwest of the U.S.: The Challenges of Root Diseases. *European Journal of Plant Pathology*, *115*(3), 271–281. https://doi.org/10.1007/s10658-006-9023-6

Piacentini, K. C., Rocha, L. O., Savi, G. D., Carnielli-Queiroz, L., Fontes, L. D. C., & Correa, B. (2019). Assessment of Toxigenic *Fusarium* Species and Their Mycotoxins in Brewing Barley Grains. *Toxins 2019, Vol. 11, Page 31*, *11*(1), 31. https://doi.org/10.3390/TOXINS11010031

Pieterse, C. M. J., Leon-Reyes, A., Van Der Ent, S., & Van Wees, S. C. M. (2009). Networking by Small-Molecule Hormones in Plant Immunity. *Nature Chemical Biology 2009 5:5*, *5*(5), 308–316. https://doi.org/10.1038/nchembio.164

Pieterse, C. M. J., Zamioudis, C., Berendsen, R. L., Weller, D. M., Van Wees, S. C. M., Bakker, P. A. H. M., NI, C. M. J. P., NI, C. Z., NI, R. L. B., NI, S. V., & NI, P. A. H. M. B. (2014). Induced Systemic Resistance by Beneficial, Microbes. *52*, 347–375. https://doi.org/10.1146/ANNUREV-PHYTO-082712-102340

Pingali, P. L. (2012). Green Revolution: Impacts, Limits, and the Path Ahead. *Proceedings of the National Academy of Sciences of the United States of America*, *109*(31), 12302. https://doi.org/10.1073/PNAS.0912953109

Poole, G. J., Harries, M., Hüberli, D., Miyan, S., MacLeod, W. J., Lawes, R., & McKay, A. (2015). Predicting Cereal Root Disease in Western Australia Using Soil DNA and Environmental Parameters. *Phytopathology*, *105*(8), 1069–1079. https://doi.org/10.1094/PHYTO-07-14-0203-R

Prashar, P., & Vandenberg, A. (2017). Genotype-Specific Responses to the Effects of Commercial *Trichoderma* Formulations in Lentil (*Lens culinaris* ssp. *culinaris*) in the Presence and Absence of the Oomycete Pathogen *Aphanomyces euteiches*. *Biocontrol Science and Technology*, *27*(10), 1123–1144. https://doi.org/10.1080/09583157.2017.1376035

Price, A. L., Patterson, N. J., Plenge, R. M., Weinblatt, M. E., Shadick, N. A., & Reich, D. (2006). Principal Components Analysis Corrects for Stratification in Genome-Wide Association Studies. *Nature Genetics*, *38*(8), 904–909. https://doi.org/10.1038/NG1847

R Core Team. (2023). R: A Language and Environment for Statistical Computing.

Rahman, M., Davies, P., Bansal, U., Pasam, R., Hayden, M., & Trethowan, R. (2020). Marker-Assisted Recurrent Selection Improves the Crown Rot Resistance of Bread Wheat. *Molecular Breeding*, *40*(3), 1–14. https://doi.org/10.1007/s11032-020-1105-1

Rauwane, M. E., Ogugua, U. V., Kalu, C. M., Ledwaba, L. K., Woldesemayat, A. A., & Ntushelo, K. (2020). Pathogenicity and Virulence Factors of *Fusarium graminearum* Including Factors Discovered Using Next Generation Sequencing Technologies and Proteomics. *Microorganisms*, *8*(2). https://doi.org/10.3390/MICROORGANISMS8020305

Rebeka, G., Shimelis, H., Laing, M. D., Tongoona, P., & Mandefro, N. (2013). Evaluation of Sorghum Genotypes Compatibility with *Fusarium oxysporum* under Striga Infestation. *Crop Science*, *53*(2), 385–393. https://doi.org/10.2135/CROPSCI2012.02.0101

Roberti, R., Badiali, F., Pisi, A., Veronesi, A., Pancaldi, D., & Cesari, A. (2006). Sensitivity of *Clonostachys rosea* and *Trichoderma* spp. as Potential Biocontrol Agents to Pesticides. *Journal of Phytopathology*, *154*(2), 100–109. https://doi.org/10.1111/J.1439-0434.2006.01069.X

Roberti, R., Veronesi, A. R., Cesari, A., Cascone, A., Di Berardino, I., Bertini, L., & Caruso, C. (2008). Induction of PR Proteins and Resistance by the Biocontrol Agent *Clonostachys rosea* in Wheat Plants Infected with *Fusarium culmorum*. *Plant Science*, *175*(3), 339–347. https://doi.org/10.1016/J.PLANTSCI.2008.05.003

Roca-Couso, R., Flores-Félix, J. D., & Rivas, R. (2021). Mechanisms of Action of Microbial Biocontrol Agents Against *Botrytis cinerea*. *Journal of Fungi 2021, Vol. 7, Page 1045*, *7*(12), 1045. https://doi.org/10.3390/JOF7121045

Rocher, F., Alouane, T., Philippe, G., Martin, M. L., Label, P., Langin, T., & Bonhomme, L. (2022). *Fusarium graminearum* Infection Strategy in Wheat Involves a Highly Conserved Genetic Program That Controls the Expression of a Core Effectome. *International Journal of Molecular Sciences*, *23*(3). https://doi.org/10.3390/ijms23031914

Rodriguez, M. A., Cabrera, G., Gozzo, F. C., Eberlin, M. N., & Godeas, A. (2011). *Clonostachys* rosea BAFC3874 as a *Sclerotinia sclerotiorum* Antagonist: Mechanisms Involved and Potential as a Biocontrol Agent. *Journal of Applied Microbiology*, *110*(5), 1177–1186. https://doi.org/10.1111/J.1365-2672.2011.04970.X

Rush, T. A., Shrestha, H. K., Gopalakrishnan Meena, M., Spangler, M. K., Ellis, J. C., Labbé, J. L., & Abraham, P. E. (2021). Bioprospecting *Trichoderma*: A Systematic Roadmap to Screen Genomes and Natural Products for Biocontrol Applications. *Frontiers in Fungal Biology*, *2*, 716511. https://doi.org/10.3389/ffunb.2021.716511

Ryan, A. D., Kinkel, L. L., & Schottel, J. L. (2004). Effect of Pathogen Isolate, Potato Cultivar, and Antagonist Strain on Potato Scab Severity and Biological Control. *Biocontrol Science and Technology*, *14*(3), 301–311. https://doi.org/10.1080/09583150410001665187

Salgado, J. D., Madden, L. V., & Paul, P. A. (2014). Efficacy and Economics of Integrating In-Field and Harvesting Strategies to Manage Fusarium Head Blight of Wheat. *98*(10), 1407–1421. https://doi.org/10.1094/PDIS-01-14-0093-RE

Sánchez-Montesinos, B., Santos, M., Moreno-Gavíra, A., Marín-Rodulfo, T., Gea, F. J., & Diánez, F. (2021). Biological Control of Fungal Diseases by *Trichoderma aggressivum f. europaeum* and Its Compatibility with Fungicides. *Journal of Fungi 2021, Vol. 7, Page 598*, *7*(8), 598. https://doi.org/10.3390/JOF7080598

Sangeetha, G., Usharani, S., & Muthukumar, A. (2009). Biocontrol with *Trichoderma* species for the management of postharvest crown rot of banana. *Phytopathologia Mediterranea*, *48*, 214–225.

Santos, M. G., Nunes da Silva, M., Vasconcelos, M. W., & Carvalho, S. M. P. (2023). Scientific and Technological Advances in the Development of Sustainable Disease management Tools: A Case Study on Kiwifruit Bacterial Canker. *Frontiers in Plant Science*, *14*, 1306420. https://doi.org/10.3389/fpls.2023.1306420

Schmidt, J., Dotson, B. R., Schmiderer, L., van Tour, A., Kumar, B., Marttila, S., Fredlund, K. M., Widell, S., & Rasmusson, A. G. (2020). Substrate and Plant Genotype Strongly Influence the Growth and Gene Expression Response to *Trichoderma afroharzianum* T22 in Sugar Beet. *Plants 2020, Vol. 9, Page 1005*, *9*(8), 1005. https://doi.org/10.3390/PLANTS9081005

Schmoll, M., & Schuster, A. (2010). Biology and biotechnology of *Trichoderma*. *Applied Microbiology and Biotechnology*, *87*(3), 787–799. https://doi.org/10.1007/s00253-010-2632-1

Schöneberg, A., Musa, T., Voegele, R. T., & Vogelgsang, S. (2015). The potential of antagonistic fungi for control of *Fusarium graminearum* and *Fusarium crookwellense* varies depending on the experimental approach. *Journal of Applied Microbiology*, *118*(5), 1165–1179. https://doi.org/10.1111/JAM.12775

Schroers, H.-J., Samuels, G. J., Seifert, K. A., & Gams, W. (1999). Classification of the Mycoparasite *Gliocladium roseum* in *Clonostachys* as *C. rosea*, its relationship to *Bionectria ochroleuca*, and notes on other *Gliocladium*-like fungi. *Mycologia*, *91*(2), 365–385. https://doi.org/10.1080/00275514.1999.12061028

Searle, S. R., Speed, F. M., & Milliken, G. A. (2024). Estimated Marginal Means, aka Least-Squares Means [R package emmeans version 1.10.1]. *American Statistician*, *34*(4), 216–221. https://doi.org/10.1080/00031305.1980.10483031

Sharma, A., Kumar, V., Shahzad, B., Tanveer, M., Sidhu, G. P. S., Handa, N., Kohli, S. K., Yadav, P., Bali, A. S., Parihar, R. D., Dar, O. I., Singh, K., Jasrotia, S., Bakshi, P., Ramakrishnan, M., Kumar, S., Bhardwaj, R., & Thukral, A. K. (2019). Worldwide Pesticide Usage and its Impacts on Ecosystem. *SN Applied Sciences*, *1*(11), 1–16. https://doi.org/10.1007/s42452-019-1485-1

Shields, M. W., Johnson, A. C., Pandey, S., Cullen, R., González-Chang, M., Wratten, S. D., & Gurr, G. M. (2019). History, Current Situation and Challenges for Conservation Biological Control. *Biological Control*, *131*, 25–35. https://doi.org/10.1016/J.BIOCONTROL.2018.12.010

Singh, B. K., Delgado-Baquerizo, M., Egidi, E., Guirado, E., Leach, J. E., Liu, H., & Trivedi, P. (2023). Climate Change Impacts on Plant Pathogens, Food Security and Paths Forward. *Nature Reviews Microbiology 2023 21:10*, *21*(10), 640–656. https://doi.org/10.1038/s41579-023-00900-7

Smiley, R. W., Gourlie, J. A., Easley, S. A., Patterson, L. M., & Whittaker, R. G. (2005). Crop Damage Estimates for Crown Rot of Wheat and Barley in the Pacific Northwest. *Plant Disease*, *89*(6), 595–604. https://doi.org/10.1094/PD-89-0595

Smith, K. P., & Goodman, R. M. (1999). Host Variation for Interactions with Beneficial Plant-Associated Microbes. *Annual Review of Phytopathology*, *37*(Volume 37, 1999), 473–491. https://doi.org/10.1146/annurev.phyto.37.1.473

Sood, M., Kapoor, D., Kumar, V., Sheteiwy, M. S., Ramakrishnan, M., Landi, M., Araniti, F., & Sharma, A. (2020). *Trichoderma*: The "Secrets" of a Multitalented Biocontrol Agent. *Plants*, *9*(6), 1–25. https://doi.org/10.3390/PLANTS9060762

Spadaro, D., & Droby, S. (2016). Development of Biocontrol Products for Postharvest Diseases of Fruit: The Importance of Elucidating the Mechanisms of Action of Yeast Antagonists. *Trends in Food Science & Technology*, *47*, 39–49. https://doi.org/10.1016/J.TIFS.2015.11.003

Spinoni, J., Vogt, J. V., Naumann, G., Barbosa, P., & Dosio, A. (2018). Will Drought Events Become More Frequent and Severe in Europe? *International Journal of Climatology*, *38*(4), 1718–1736. https://doi.org/10.1002/JOC.5291

Stenberg, J. A., Sundh, I., Becher, P. G., Björkman, C., Dubey, M., Egan, P. A., Friberg, H., Gil, J. F., Jensen, D. F., Jonsson, M., Karlsson, M., Khalil, S., Ninkovic, V., Rehermann, G., Vetukuri, R. R., & Viketoft, M. (2021). When is it Biological Control? A Framework of Definitions, Mechanisms, and Classifications. *Journal of Pest Science*, *94*(3), 665–676. https://doi.org/10.1007/s10340-021-01354-7

Sun, Z. Bin, Sun, M. H., Zhou, M., & Li, S. D. (2017). Transformation of the Endochitinase Gene Chi67-1 in *Clonostachys rosea* 67-1 Increases its Biocontrol Activity Against *Sclerotinia sclerotiorum*. *AMB Express*, *7*(1). https://doi.org/10.1186/S13568-016-0313-X

Sun, Z. B., Li, S. D., Ren, Q., Xu, J. L., Lu, X., & Sun, M. H. (2020). Biology and Applications of *Clonostachys rosea*. *Journal of Applied Microbiology*, *129*(3), 486–495. https://doi.org/10.1111/JAM.14625

Sutton, J. C. (1982). Epidemiology of Wheat Head Blight and Maize Ear Rot Caused by *Fusarium graminearum*. *Canadian Journal of Plant Pathology*, *4*(2), 195–209. https://doi.org/10.1080/07060668209501326

Syed Ab Rahman, S. F., Singh, E., Pieterse, C. M. J., & Schenk, P. M. (2018). Emerging Microbial Biocontrol Strategies for Plant Pathogens. *Plant Science : An International Journal of Experimental Plant Biology*, *267*, 102–111. https://doi.org/10.1016/J.PLANTSCI.2017.11.012

Tahir, H. A. S., Gu, Q., Wu, H., Niu, Y., Huo, R., & Gao, X. (2017). *Bacillus* volatiles adversely affect the physiology and ultra-structure of *Ralstonia solanacearum* and induce systemic resistance in tobacco against bacterial wilt. *Scientific Reports 2017 7:1*, *7*(1), 1–15. https://doi.org/10.1038/srep40481

Thambugala, K. M., Daranagama, D. A., Phillips, A. J. L., Kannangara, S. D., & Promputtha, I. (2020). Fungi vs. Fungi in Biocontrol: An Overview of Fungal Antagonists Applied Against Fungal Plant Pathogens. *Frontiers in Cellular and Infection Microbiology*, *10*, 604923. https://doi.org/10.3389/fcimb.2020.604923

Tian, Y., Tan, Y., Yan, Z., Liao, Y., Chen, J., De Boevre, M., De Saeger, S., & Wu, A. (2018). Antagonistic and Detoxification Potentials of *Trichoderma* isolates for control of zearalenone (ZEN) producing *Fusarium graminearum*. *Frontiers in Microbiology*, *8*(JAN), 325406. https://doi.org/10.3389/fmicb.2017.02710

Timper, P. (2014). Conserving and Enhancing Biological Control of Nematodes. *Journal of Nematology*, *46*(2), 75. /pmc/articles/PMC4077175/

Toledo, A. V., Virla, E., Humber, R. A., Paradell, S. L., & Lastra, C. C. L. (2006). First Record of *Clonostachys rosea* (Ascomycota: *Hypocreales*) as an entomopathogenic fungus of *Oncometopia tucumana* and *Sonesimia grossa* (Hemiptera: *Cicadellidae*) in Argentina. *Journal of Invertebrate Pathology*, *92*(1), 7–10. https://doi.org/10.1016/J.JIP.2005.10.005

Trail, F. (2009). For Blighted Waves of Grain: *Fusarium graminearum* in the Postgenomics Era. *Plant Physiology*, *149*(1), 103. https://doi.org/10.1104/PP.108.129684

Tucci, M., Ruocco, M., De Masi, L., De Palma, M., & Lorito, M. (2011). The Beneficial Effect of *Trichoderma* spp. on Tomato is Modulated by the Plant Genotype. *Molecular Plant Pathology*, *12*(4), 341–354. https://doi.org/10.1111/J.1364-3703.2010.00674.X

Tudi, M., Ruan, H. D., Wang, L., Lyu, J., Sadler, R., Connell, D., Chu, C., & Phung, D. T. (2021). Agriculture Development, Pesticide Application and Its Impact on the Environment. *International Journal of Environmental Research and Public Health*, *18*(3), 1–24. https://doi.org/10.3390/IJERPH18031112

Tzelepis, G., Dubey, M., Jensen, D. F., & Karlsson, M. (2015). Identifying Glycoside Hydrolase Family 18 genes in the Mycoparasitic Fungal Species *Clonostachys rosea*. *Microbiology (United Kingdom)*, *161*(7), 1407–1419. https://doi.org/10.1099/mic.0.000096

Umer, M., Mubeen, M., Iftikhar, Y., Ali Shad, M., Usman, H. M., Sohail, M. A., Atiq, M. N., Abbas, A., & Ateeq, M. (2021). Role of Rhizobacteria on Plants Growth and Biological Control of Plant Diseases: A Review. *Plant Protection*, *5*(1), 59–73. https://doi.org/10.33804/PP.005.01.3565

Valan Arasu, M., Vijayaraghavan, P., Al-Dhabi, N. A., Choi, K. C., & Moovendhan, M. (2023). Biocontrol of *Trichoderma gamsii* Induces Soil Suppressive and Growth-Promoting Impacts and Rot Disease-Protecting Activities. *Journal of Basic Microbiology*, *63*(7), 801–813. https://doi.org/10.1002/JOBM.202300016

van de Wouw, M., van Hintum, T., Kik, C., van Treuren, R., & Visser, B. (2010). Genetic Diversity Trends in Twentieth Century Crop Cultivars: A Meta Analysis. *TAG. Theoretical and Applied Genetics. Theoretische Und Angewandte Genetik*, *120*(6), 1241–1252. https://doi.org/10.1007/S00122-009-1252-6

van der Lee, T., Zhang, H., van Diepeningen, A., & Waalwijk, C. (2015). Biogeography of *Fusarium graminearum* Species Complex and Chemotypes: A Review. *Food Additives & Contaminants: Part A*, *32*(4), 453–460. https://doi.org/10.1080/19440049.2014.984244

van Dijk, M., Morley, T., Rau, M. L., & Saghai, Y. (2021). A Meta-Analysis of Projected Global Food Demand and Population at Risk of Hunger for the Period 2010–2050. *Nature Food 2021 2:7*, *2*(7), 494–501. https://doi.org/10.1038/s43016-021-00322-9

Van Loon, L. C., Bakker, P. A. H. M., & Pieterse, C. M. J. (1998). Systemic Resistance Induced by Rhizosphere Bacteria. *Annual Review of Phytopathology*, *36*, 453–483. https://doi.org/10.1146/ANNUREV.PHYTO.36.1.453

Verma, M., Brar, S. K., Tyagi, R. D., Surampalli, R. Y., & Valéro, J. R. (2007). Antagonistic Fungi, *Trichoderma* spp.: Panoply of Biological Control. *Biochemical Engineering Journal*, *37*(1), 1–20. https://doi.org/10.1016/J.BEJ.2007.05.012

Vogelgsang, S., Beyer, M., Pasquali, M., Jenny, E., Musa, T., Bucheli, T. D., Wettstein, F. E., & Forrer, H. R. (2019). An Eight-year Survey of Wheat Shows Distinctive Effects of Cropping Factors on Different *Fusarium* Species and Associated Mycotoxins. *European Journal of Agronomy*, *105*, 62–77. https://doi.org/10.1016/J.EJA.2019.01.002

von der Goltz, J., Dar, A., Fishman, R., Mueller, N. D., Barnwal, P., & McCord, G. C. (2020). Health Impacts of the Green Revolution: Evidence from 600,000 Births Across the Developing World. *Journal of Health Economics*, *74*. https://doi.org/10.1016/J.JHEALECO.2020.102373

Wang, J., & Zhang, Z. (2021). GAPIT Version 3: Boosting Power and Accuracy for Genomic Association and Prediction. *Genomics, Proteomics & Bioinformatics*, *19*(4), 629–640. https://doi.org/10.1016/J.GPB.2021.08.005

Wang, M., Xue, J., Ma, J., Feng, X., Ying, H., & Xu, H. (2020). *Streptomyces lydicus* M01 Regulates Soil Microbial Community and Alleviates Foliar Disease Caused by *Alternaria alternata* on Cucumbers. *Frontiers in Microbiology*, *11*, 523552. https://doi.org/10.3389/fmicb.2020.00942

Wang, M., Yan, J., Zhao, J., Song, W., Zhang, X., Xiao, Y., & Zheng, Y. (2012). Genome-wide Association Study (GWAS) of Resistance to Head Smut in Maize. *Plant Science*, *196*, 125–131. https://doi.org/10.1016/J.PLANTSCI.2012.08.004

Wickham, H., Averick, M., Bryan, J., Chang, W., D', L., Mcgowan, A., François, R., Grolemund, G., Hayes, A., Henry, L., Hester, J., Kuhn, M., Lin Pedersen, T., Miller, E., Bache, S. M., Müller, K., Ooms, J., Robinson, D., Seidel, D. P., … Yutani, H. (2019). Welcome to the Tidyverse. *Journal of Open Source Software*, *4*(43), 1686. https://doi.org/10.21105/JOSS.01686

Wilkinson, G. N., & Rogers, C. E. (1973). Symbolic Description of Factorial Models for Analysis of Variance. *Journal of Applied Statistics*, *22*(3), 392–399. https://doi.org/10.2307/2346786

Wilson, P. S., Ketola, E. O., Ahvenniemi, P. M., Lehtonen, M. J., & Valkonen, J. P. T. (2008). Dynamics of Soilborne *Rhizoctonia solani* in the Presence of *Trichoderma harzianum*: Effects on Stem Canker, Black Scurf and Progeny Tubers of Potato. *Plant Pathology*, *57*(1), 152–161. https://doi.org/10.1111/J.1365-3059.2007.01706.X

Winter, M., Samuels, P. L., Otto-Hanson, L. K., Dill-Macky, R., & Kinkel, L. L. (2019). Biological Control of Fusarium Crown and Root Rot of Wheat by *Streptomyces* isolates – It's Complicated. *Phytobiomes Journal*, *3*(1), 52–60. https://doi.org/10.1094/PBIOMES-11-18-0052-R

Xu, F., Yang, G., Wang, J., Song, Y., Liu, L., Zhao, K., Li, Y., & Han, Z. (2018). Spatial Distribution of Root and Crown Rot Fungi Associated with Winter Wheat in the North China Plain and its Relationship with Climate Variables. *Frontiers in Microbiology*, *9* (MAY), 350491. https://doi.org/10.3389/fmicb.2018.01054

Yang, J., Manolio, T. A., Pasquale, L. R., Boerwinkle, E., Caporaso, N., Cunningham, J. M., De Andrade, M., Feenstra, B., Feingold, E., Hayes, M. G., Hill, W. G., Landi, M. T., Alonso, A., Lettre, G., Lin, P., Ling, H., Lowe, W., Mathias, R. A., Melbye, M., … Visscher, P. M. (2011). Genome Partitioning of Genetic Variation for Complex Traits Using Common SNPs. *Nature Genetics*, *43*(6), 519–525. https://doi.org/10.1038/NG.823

Yao, X., Guo, H., Zhang, K., Zhao, M., Ruan, J., & Chen, J. (2023). *Trichoderma* and its Role in Biological Control of Plant Fungal and Nematode Disease. *Frontiers in Microbiology*, *14*. https://doi.org/10.3389/FMICB.2023.1160551

Yohalem, D. S., Nielsen, K., Green, H., & Jensen, D. F. (2004). Biocontrol Agents Efficiently Inhibit Sporulation of *Botrytis aclada* on Necrotic Leaf Tips but Spread to Adjacent Living Tissue is not Prevented. *FEMS Microbiology Ecology*, *47*(3), 297–303. https://doi.org/10.1016/S0168-6496(03)00299-X

Yu, Y., Gui, Y., Li, Z., Jiang, C., Guo, J., & Niu, D. (2022). Induced Systemic Resistance for Improving Plant Immunity by Beneficial Microbes. *Plants 2022, Vol. 11, Page 386*, *11*(3), 386. https://doi.org/10.3390/PLANTS11030386

Zamoum, M., Goudjal, Y., Sabaou, N., Barakate, M., Mathieu, F., & Zitouni, A. (2015). Biocontrol Capacities and Plant Growth-Promoting Traits of Endophytic Actinobacteria Isolated from Native Plants of Algerian Sahara. *Journal of Plant Diseases and Protection*, *122*(5–6), 215–233. https://doi.org/10.1007/BF03356555

Zehra, A., Raytekar, N. A., Meena, M., & Swapnil, P. (2021). Efficiency of Microbial Bio-Agents as Elicitors in Plant Defense Mechanism under Biotic Stress: A Review. *Current Research in Microbial Sciences*, *2*, 100054. https://doi.org/10.1016/J.CRMICR.2021.100054

Zhai, M. M., Qi, F. M., Li, J., Jiang, C. X., Hou, Y., Shi, Y. P., Di, D. L., Zhang, J. W., & Wu, Q. X. (2016). Isolation of Secondary Metabolites from the Soil-Derived Fungus *Clonostachys rosea* YRS-06, a Biological Control Agent, and Evaluation of Antibacterial Activity. *Journal of Agricultural and Food Chemistry*, *64*(11), 2298–2306. https://doi.org/10.1021/acs.jafc.6b00556

Zhang, F., Ge, H., Zhang, F., Guo, N., Wang, Y., Chen, L., Ji, X., & Li, C. (2016). Biocontrol Potential of *Trichoderma harzianum* Isolate T-aloe Against *Sclerotinia sclerotiorum* in Soybean. *Plant Physiology and Biochemistry*, *100*, 64–74. https://doi.org/10.1016/J.PLAPHY.2015.12.017

Zhang, N., Yuan, S., Zhang, Q., Liu, W., Zhou, Y., & Yang, W. (2022). Screening Fungicides for Controlling Wheat Crown Rot Caused by *Fusarium pseudograminearum* Across Hebei

Province in China. *Agriculture (Switzerland)*, *12*(10), 1643. https://doi.org/10.3390/AGRICULTURE12101643/S1

Zhu, Y., Chen, H., Fan, J., Wang, Y., Li, Y., Chen, J., Fan, J. X., Yang, S., Hu, L., Leung, H., Mew, T. W., Teng, P. S., Wang, Z., & Mundt, C. C. (2000). Genetic Diversity and Disease Control in Rice. *Nature*, *406*(6797), 718–722. https://doi.org/10.1038/35021046

Supplementary Material

Supplementary table 1 List of the 190 genotypes used in this study for the *Clonostachys rosea* bioassay. Genotypes marked in bolt were used in the bioassay with treatment *Fusarium graminearum* and *Trichoderma afroharzianum* (FgTa).

nordID	genID	cultivar	release_year	country	accession_type
NGB16916	593001	Galicia	2010	Denmark	Cultivar
NGB1	593002	Iduna	1911	Sweden	Cultivar
NGB10	593003	Äring II	1936	Sweden	Cultivar
NGB11	593004	Äring III	1940	Sweden	Cultivar
NGB11316	593005	Kalle	1990	Norway	Cultivar
NGB11317	593006	Rida	1976	Norway	Cultivar
NGB11425	593007	Starke II - LR	1968	Denmark	Unknown
NGB12	593008	Eroica	1943	Sweden	Cultivar
NGB12242	593010	Pansar I	1915	Sweden	Cultivar
NGB12243	**593011**	**Ergo II**	**1949**	**Sweden**	**Cultivar**
NGB12244	593012	Konge III	1939	Denmark	Cultivar
NGB13	593013	Virtus	1945	Sweden	Cultivar
NGB13023	593014	Ritmo		Netherlands	Unknown
NGB13430	593015	Finnish Winter Wheat (Pi181455)		Finland	Unknown
NGB13442	593016	Winter Wheat From Bohuslän			Unknown
NGB13444	593017	-		Sweden	Landrace
NGB13445	593018	Ångermanland		Sweden	Landrace
NGB13446	593019	Tystofte smaahvede	1909	Denmark	Cultivar
NGB13479	593020	Stava	1995	Sweden	Cultivar
NGB13576	593021	Urban	1981	Germany	Cultivar
NGB13659	593022	Bjørke	1997	Norway	
NGB14	593023	Aros	1947	Sweden	Cultivar
NGB14114	593024	Gunbo	1997	Sweden	Cultivar
NGB14115	593025	Mjölner	1996	Sweden	
NGB14116	593026	Rental	1993	Sweden	Cultivar

NGB14117	593027	Stava	1995	Sweden	Cultivar
NGB14118	593028	Rudolf rubin	1921	Sweden	Cultivar
NGB14286	593029	S-5		Sweden	Landrace
NGB15	593030	Eroica II	1951	Sweden	Cultivar
NGB15070	593031	Kirsten	1997	Denmark	Cultivar
NGB15071	593032	Lone	1994	Denmark	Cultivar
NGB15072	**593033**	**Brandt**	**1999**	**Denmark**	**Cultivar**
NGB15075	593034	Karat	2000	Denmark	Cultivar
NGB15076	593035	Arlo		Denmark	Cultivar
NGB16	593036	Banco	1953	Sweden	Cultivar
NGB16675	593037	Saxild	2002	Denmark	Cultivar
NGB16679	593038	Abba	2002	Denmark	Cultivar
NGB16852	593039	Konsul	1994	Sweden	Cultivar
NGB16853	593040	Rektor	1981	Denmark	Cultivar
NGB16909	593041	Probat	2000	Denmark	Cultivar
NGB16910	593042	Stakado	1995	Denmark	Cultivar
NGB17	593043	Ertus	1953	Sweden	Cultivar
NGB17135	593044	Sampo	1933	Finland	Cultivar
NGB17137	593045	Väinö		Finland	Cultivar
NGB17141	593046	Pitkävihneinen maatiainen		Finland	Landrace
NGB17142	593047	Kökar		Finland	Landrace
NGB18	593048	Starke	1959	Sweden	Cultivar
NGB18629	593049	Olympia	1941	Finland	Cultivar
NGB19	593050	Trond	1960	Sweden	Cultivar
NGB2	593051	Standard	1921	Sweden	Cultivar
NGB20	593052	Thor	1961	Sweden	Cultivar
NGB21	593053	Norre	1962	Sweden	Cultivar
NGB21864	593054	Otso	1989	Finland	Cultivar
NGB22	593055	Starke II	1968	Sweden	Cultivar
NGB23	593056	Holme	1972	Sweden	Cultivar
NGB23170	593057	Kuikka		Finland	Landrace
NGB23171	593058	Istäsuomalainen		Finland	Landrace
NGB23345	593059	Alrø	1999	Denmark	Cultivar

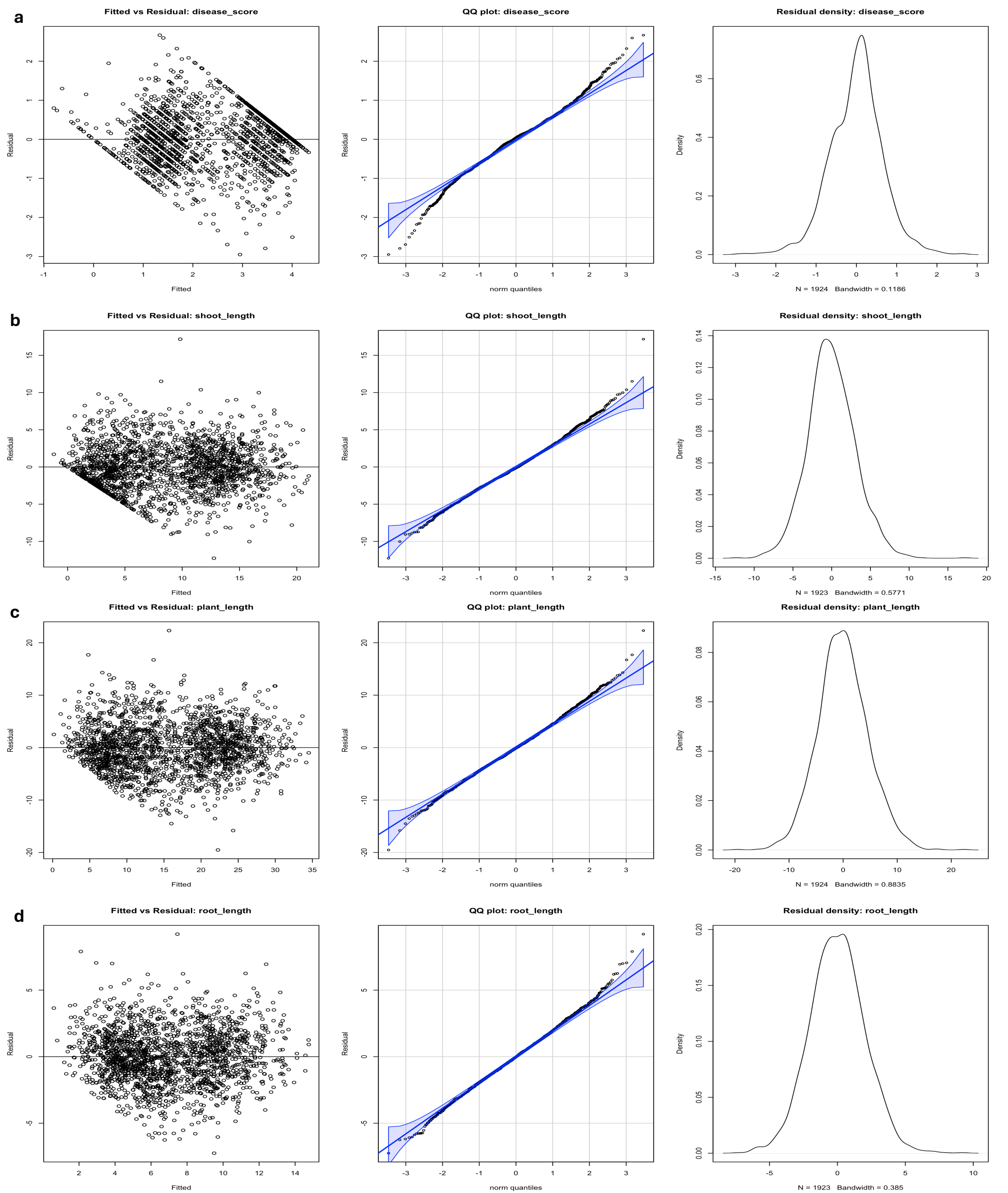
NGB23346	593060	Dirigent	1999	Denmark	Cultivar
NGB23347	**593061**	**Facet**	**1995**	**Sweden**	**Cultivar**
NGB23348	593062	Frimegu	1995	Denmark	Cultivar
NGB23349	593063	Hybris	1998	Denmark	Cultivar
NGB23350	593064	Junker	1988	Sweden	Cultivar
NGB23351	593065	Miller	2000	Denmark	Cultivar
NGB23352	593066	Pentium	1996	Denmark	Cultivar
NGB23353	593067	Revelj	2000	Sweden	Cultivar
NGB23356	593068	Skjaldar	1976	Norway	Cultivar
NGB23357	593069	Solist	1999	Denmark	Cultivar
NGB23358	593070	Terra	1994	Denmark	Cultivar
NGB23359	593071	Ure	1991	Denmark	Cultivar
NGB23360	**593072**	**Wasmo**	**1999**	**Denmark**	**Cultivar**
NGB23363	593073	Cardos	1998	Germany	Cultivar
NGB23364	593074	Gefion	1998	Denmark	Cultivar
NGB23678	593075	Loyal		Sweden	Cultivar
NGB23679	593076	Ambition		Denmark	Cultivar
NGB23681	**593077**	**Mariboss**		**Denmark**	**Cultivar**
NGB23682	593078	Hereford		Denmark	Cultivar
NGB23780	593079	Agrestis	2001	Denmark	Cultivar
NGB24	593080	Walde	1945	Sweden	Cultivar
NGB2434	593081	Folke	1981	Sweden	Cultivar
NGB2435	593082	Holger	1981	Sweden	Cultivar
NGB25	593083	Sture	1975	Sweden	Cultivar
NGB26	593084	Helge	1980	Sweden	Cultivar
NGB3	**593085**	**Jarl**	**1925**	**Sweden**	**Cultivar**
NGB31181	593086	Cymbal	2012	Sweden	Cultivar
NGB31730	593087	Penta Sejet	2001	Denmark	Cultivar
NGB334	593088	Linna	1965	Finland	Cultivar
NGB343	593089	Nisu	1966	Finland	Cultivar
NGB344	**593090**	**Vakka**	**1959**	**Finland**	**Cultivar**
NGB347	593091	Aura	1976	Finland	Cultivar

NGB348	593092	Jyvä	1965	Finland	Cultivar
NGB4	593093	Ankar	1928	Sweden	Cultivar
NGB4494	593094	Borstvete från Gotland		Sweden	Landrace
NGB473	593095	Sigyn II	1972	Norway	Cultivar
NGB4770	593096	Als	1923	Denmark	Cultivar
NGB4783	**593097**	**Storvik sjundeå**		**Finland**	**Landrace**
NGB4799	593098	Atchena K.62		Afghanistan	Landrace
NGB5	593101	Saxo	1929	Sweden	Cultivar
NGB5147	593102	Squarehead II	1909	Sweden	Cultivar
NGB5151	593103	Deh Kundi K.244		Afghanistan	Landrace
NGB5152	593104	Gusalek K.17		Afghanistan	Landrace
NGB5153	593105	Hunsballe R	1955	Denmark	Cultivar
NGB6	593106	Ankar II	1928	Sweden	Cultivar
NGB6383	593107	Skandia	1935	Sweden	Cultivar
NGB6388	593108	Lading skæghvede		Denmark	Landrace
NGB6392	593109	Kabel K.238		Afghanistan	Landrace
NGB6691	593110	Lantvete från Halland		Sweden	Landrace
NGB6692	593111	Lantvete från Uppsala		Sweden	Landrace
NGB6693	593112	Kotte	1950	Sweden	Cultivar
NGB6694	593113	Extra squarehead	1900	Sweden	Cultivar
NGB6695	593114	Bore	1902	Sweden	Cultivar
NGB6696	593115	Grenadier II	1907	Sweden	Cultivar
NGB6697	593116	Extra squarehead II	1909	Sweden	Cultivar
NGB6698	593117	Pudel	1910	Sweden	Cultivar
NGB6699	**593118**	**Renodlat sammetsvete**	**1910**	**Sweden**	**Cultivar**
NGB6700	593119	Sol	1911	Sweden	Cultivar
NGB6701	593120	Sol II	1916	Sweden	Cultivar
NGB6702	593121	Thule II	1917	Sweden	Cultivar
NGB6703	593122	Pansar III	1919	Sweden	Cultivar
NGB6704	593123	Svea I	1924	Sweden	Cultivar
NGB6705	**593124**	**Riddar**	**1922**	**Sweden**	**Cultivar**
NGB6706	593125	Birgitta	1922	Sweden	Cultivar

NGB6707	593126	Pansar III	1923	Sweden	Cultivar
NGB6708	593127	Kron	1925	Sweden	Cultivar
NGB6709	593128	Stål	1927	Sweden	Cultivar
NGB6710	593129	Sol III	1929	Sweden	Cultivar
NGB6712	593130	Bore II	1931	Sweden	Cultivar
NGB6713	593131	Gyllen II	1935	Sweden	Cultivar
NGB6714	593132	Thule III	1936	Sweden	Cultivar
NGB6715	593133	Sol IV	1937	Sweden	Cultivar
NGB6716	593134	Gyllen II	1938	Sweden	Cultivar
NGB6717	593135	Skandia II	1939	Sweden	Cultivar
NGB6718	593136	Gluten	1939	Sweden	Cultivar
NGB6719	593137	Borg	1943	Sweden	Cultivar
NGB6720	593138	Skandia III B	1955	Sweden	Cultivar
NGB6721	593139	Hansa Svalöf	1945	Sweden	Cultivar
NGB6722	593140	Pärl II	1946	Sweden	Cultivar
NGB6723	593141	Odin	1949	Sweden	Cultivar
NGB6724	593142	Robur	1949	Sweden	Cultivar
NGB6725	593143	Svale	1955	Sweden	Cultivar
NGB6726	593144	Diana	1957	Sweden	Cultivar
NGB6727	593145	Ölve	1959	Sweden	Cultivar
NGB6728	**593146**	**Seba**	**1969**	**Sweden**	**Cultivar**
NGB6729	**593147**	**Virgo**	**1968**	**Sweden**	**Cultivar**
NGB6730	593148	Solid	1973	Sweden	Cultivar
NGB6731	593149	Hildur	1976	Sweden	Cultivar
NGB6773	**593150**	**Hankkijan ilves**	**1984**	**Finland**	**Cultivar**
NGB7	593151	Äring	1932	Sweden	Cultivar
NGB7027	593152	Dania	1926	Denmark	Cultivar
NGB7034	593153	Mendel	1950	Sweden	Cultivar
NGB7043	593154	Bagelgrom K.87		Afghanistan	Landrace
NGB7044	593155	Kabel K.162		Afghanistan	Landrace
NGB7045	593156	Kabel K.165		Afghanistan	Landrace
NGB7183	593157	Små II, Tystofte	1915	Denmark	Cultivar
NGB7184	593158	Storaks Abed	1967	Denmark	Cultivar
NGB7193	593159	Gusalek K.10 A		Afghanistan	Landrace
NGB7194	593160	Vama K.40 A		Afghanistan	Landrace

NGB7195	593161	Øtofte I.56	1956	Denmark	Cultivar
NGB7482	593162	Kosack	1984	Sweden	Cultivar
NGB7483	593163	Sleipner	1988	Sweden	Cultivar
NGB7484	593164	Rurik	1986	Sweden	Cultivar
NGB8	593165	Ergo	1934	Sweden	Cultivar
NGB8189	593166	Dronning	1940	Sweden	Cultivar
NGB8194	593167	Konge II	1939	Denmark	Cultivar
NGB8197	593168	Stand tystofte	1907	Denmark	Cultivar
NGB8198	593169	Lantvete från Värmland		Sweden	Landrace
NGB8199	593170	Gammalt Svenskt lantvete		Sweden	Landrace
NGB8672	593171	Salut	1982	Sweden	Cultivar
NGB8933	593172	Borg Abed	1966	Denmark	Cultivar
NGB8937	593173	Bankuta		Sweden	Cultivar
NGB8946	593174	Brødtorp Pajo		Denmark	Landrace
NGB8957	593175	Enger		Norway	Landrace
NGB8968	593176	Haukiala Pirola		Finland	Landrace
NGB8973	593177	Ideal	1929	Denmark	Cultivar
NGB8999	**593178**	**Sammets**	**1910**	**Sweden**	**Cultivar**
NGB9	593179	Standard II	1936	Sweden	Cultivar
NGB9016	**593181**	**Trifolium 14**	**1925**	**Denmark**	**Cultivar**
NGB9017	593182	Tystofte Stakket	1967	Denmark	Cultivar
NGB9020	**593184**	**Varma Tammisto**	**1933**	**Finland**	**Cultivar**
NGB9057	593185	Hallandsvete		Sweden	Landrace
NGB9062	593186	Mendel II	1952	Sweden	Cultivar
NGB9078	593188	Kabel K.161		Afghanistan	Landrace
NGB9079	593189	Pandshir K.156 A		Afghanistan	Landrace
NGB9080	**593190**	**Pandshir K.157**		**Afghanistan**	**Landrace**
NGB9118	593191	Nana	1975	Denmark	Cultivar
NGB9119	593192	Sarah	1976	Denmark	Cultivar
NGB9122	**593193**	**Anja**	**1980**	**Denmark**	**Cultivar**
NGB9123	593194	Kraka	1980	Denmark	Cultivar
NGB9925	593195	Portal	1990	Germany	Cultivar

NGB9952	**593196**	**Tjelvar**	**1984**	**Sweden**	**Cultivar**
NGB9953	**593197**	**Tryggve**	**1990**	**Sweden**	**Cultivar**
Kranich	C1	Kranich	2007	Germany	
Nelson	C2	Nelson	2011	Germany	
Target	C3	Target	2010		



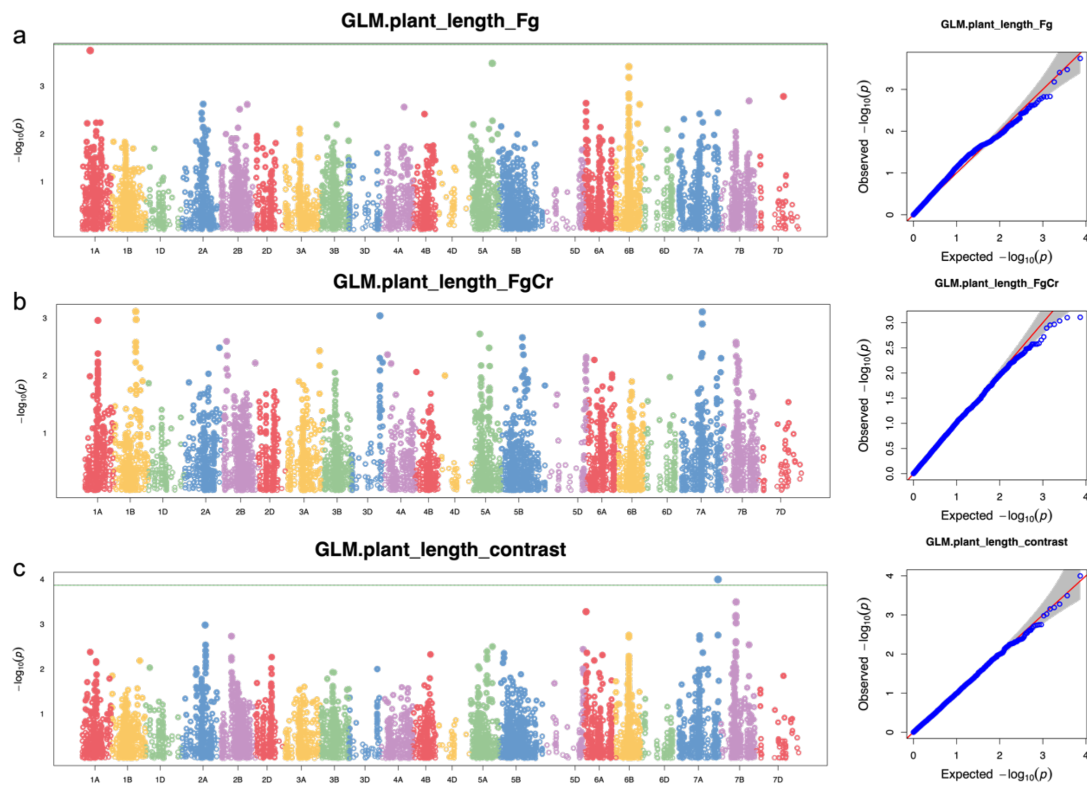
Suplementary Figure 1 Residuals vs fitted, Normal Q-Q and Density plots for Model of bioassay with treatment FgCr (*Clonostachys rosea*) in each trait. (a) disease score, (b) shoot length, (c) plant length, (d) root length.

Supplementary table 2 Summary of the significant SNPs marker identified which are associated as resistance indicator in GWAS analysis with 181 winter wheat genotypes.

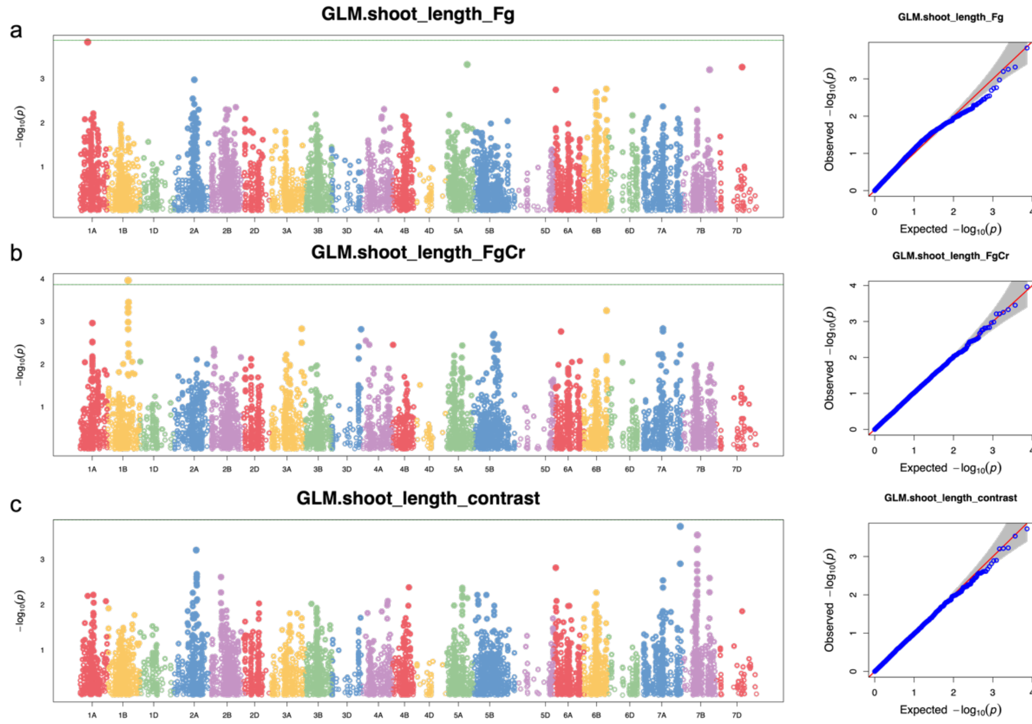
Trait_factor	**SNP marker name**	**Chr**	**Position (cM)**	***P*-value**	**MAF**
Plant length_contrast	Ra_c956_2318	7A	228	0.00010066160646946	0.161111111111111
Root length_Fg	Tdurum_contig15235_951	6B	65	0.000117016832676281	0.165745856353591
Shoot length_FgCr	Tdurum_contig10036_474	1B	112	0.000108659194439725	0.0773480662983425
Disease score_Fg	Excalibur_c7026_2635	1A	53	0.00011632709740174	0.0773480662983425
Disease score_Fg	BS00089497_51	2A	115	4.74112033107019e-05	0.124309392265193
Disease score_Fg	Kukri_c40121_373	2A	116	8.03233835758145e-05	0.237569060773481
Disease score_Fg	BS00096604_51	4B	71	3.33305376066823e-05	0.0607734806629834
Disease score_Fg	RFL_Contig2459_2314	4B	73	7.66706634796347e-05	0.0580110497237569
Disease score_ contrast	Excalibur_rep_c111629_239	7B	77	5.4175683358489e-05	0.116666666666667
Disease score_contrast	wsnp_Ex_rep_c109138_92064554	7B	77	7.19550692568248e-05	0.119444444444444
Disease score_contrast	BobWhite_c3564_81	7B	77	8.84923437955043e-05	0.122222222222222
Disease score_contrast	BS00021972_51	7B	77	8.84923437955043e-05	0.122222222222222
Disease score_contrast	BS00010557_51	7B	78	7.19550692568248e-05	0.119444444444444

Disease score_contrast	wsnp_Ku_rep_c68953_68153061	7B	78	9.9336415978717e-05	0.116666666666667
Disease score_contrast	Kukri_c20611_293	7B	78	0.000110281154788156	0.116666666666667

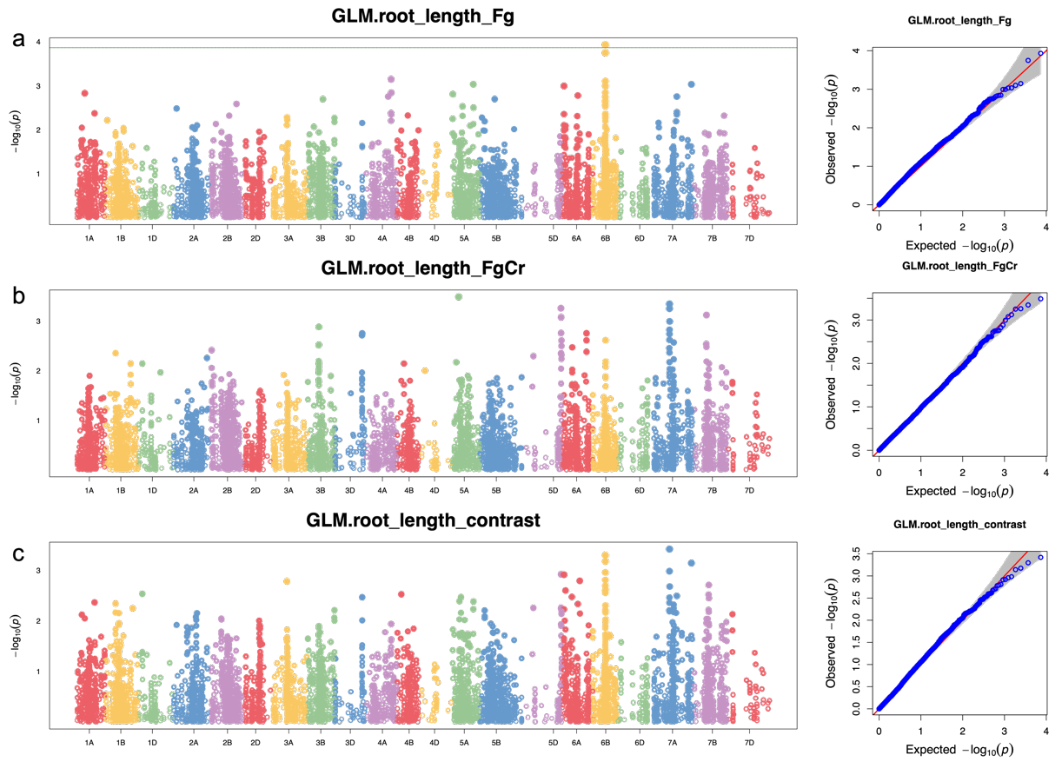
Chr: cromossome; MAF: minor allele frequency, SNP: Single-nucleotide polymorphism



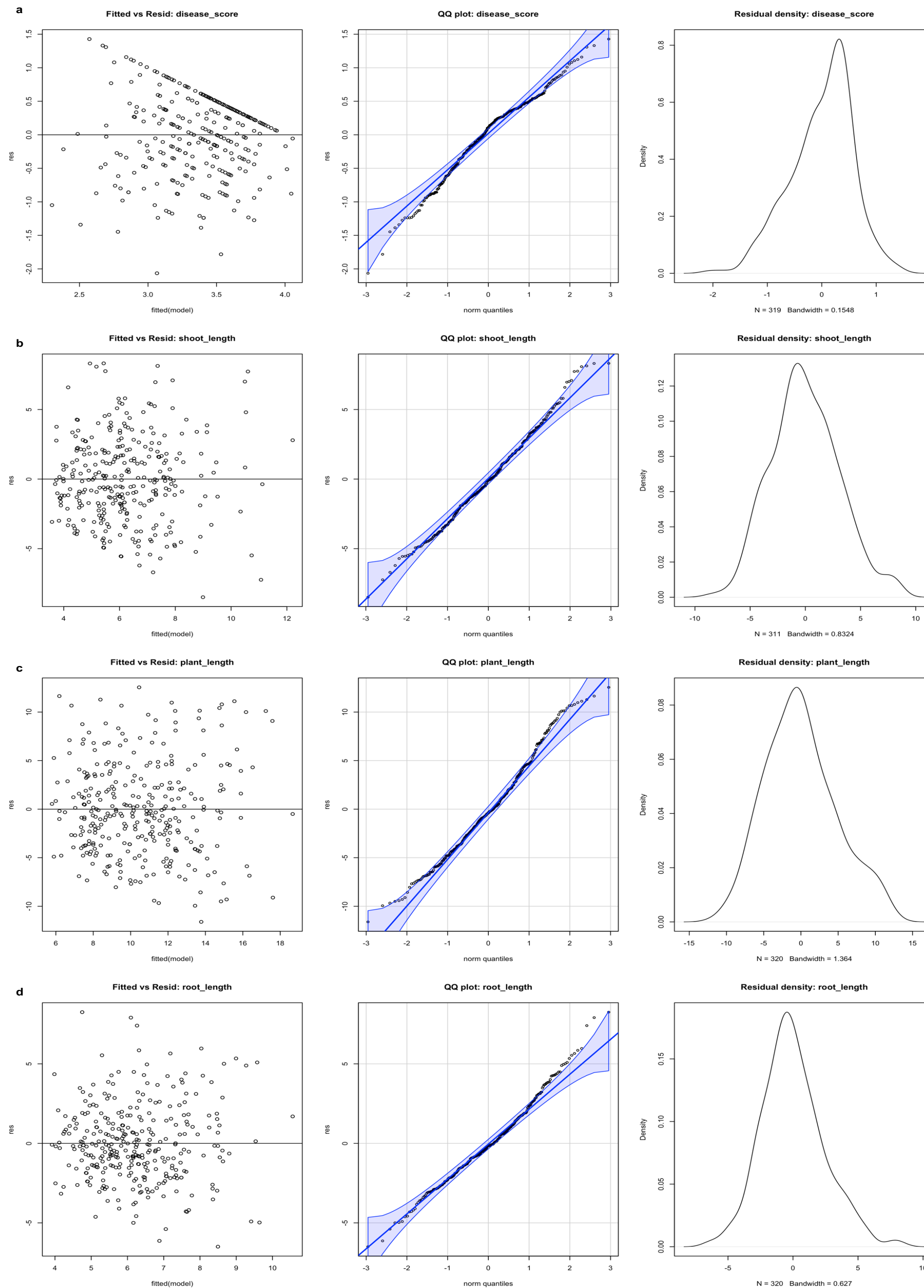
Suplementary Figure 2 Manhattan plot and corresponding QQ plots showing the association of SNP markers with the FFR plant length in treatment Fg (a), treatment FgCr (b) and disease score contrast or biocontrol efficacy (c). The solid horizontal line shows the significance threshold of −log10(P) = 3.85.



Suplementary Figure 3 Manhattan plot and corresponding QQ plots showing the association of SNP markers with the FFR shoot length in treatment Fg (a), treatment FgCr (b) and disease score contrast or biocontrol efficacy (c). The solid horizontal line shows the significance threshold of −log10(P) = 3.85.



Suplementary Figure 4 Manhattan plot and corresponding QQ plots showing the association of SNP markers with the FFR root length in treatment Fg (a), treatment FgCr (b) and disease score contrast or biocontrol efficacy (c). The solid horizontal line shows the significance threshold of −log10(P) = 3.85.



Suplementary Figure 5 Residuals vs fitted, Normal Q-Q and Density plots for Model of bioassay with treatment FgTa. (a) disease score, (b) shoot length, (c) plant length, (d) root length.