Recent Advances in Understanding and Controlling Fusarium Diseases of Alliums

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Abstract: Allium species are known for their culinary, medicinal, and ornamental purposes. Fusarium basal rot is one of the most damaging soilborne fungal diseases of Allium species and poses a significant threat to yield, quality, and storage life worldwide. Various species of Fusarium have been identified as causal agents for Fusarium basal rot, depending on the Allium species involved. Diverse disease management practices have been implemented to mitigate the impact of Fusarium basal rot. This review article provides a comprehensive overview of the recent progress in detecting different species of Fusarium involved in Fusarium basal rot and strategies to control them in affected Allium species involving chemical, biological, and cultural methods. It covers the latest advancements in host plant resistance research from traditional breeding to modern molecular techniques and studying secondary metabolites involved in defense mechanisms against Fusarium basal rot.

Keywords: Allium; Fusarium; fungicides; biocontrol agents; soil amendments; host plant resistance; screening; quantitative trait loci; defense-related genes; saponins

1. Introduction

Allium species, contained in the Alliaceae family, are among the earliest domesticated plants, valued for their historical significance and culinary use [1]. The major cultivated Allium species include onion (Allium cepa L.), garlic (A. sativum L.), shallot (A. cepa L. Aeggregatum group or A. cepa L. var. ascalonicum), leek (A. porrum L.), chive (A. schoenoprasum L.), Chinese chive (A. tuberosum Rottl. ex K. Spreng. or A. ramosum L.), Chinese garlic (A. macrostemon Bunge) and Welsh or Japanese bunching onion (A. fistulosum L.) [2–4]. Wild species that are considered an important source of disease resistance include A. ledebourianum Roem. & Schult., A. galanthum Kar. & Kir., A. altaicum Pall., A. caesium Schrenck, A. proliferum (Moench) Schrad. ex Willd., A. nutans L., A. taviilovii Popv & Vved., and A. tricoccum Ait. [3,4]. Ornamental species comprise A. giganteum Regel. & Schult., A. galanthum Kar. & Kir., A. alliucum Pall., A. caesium Schrenck, A. proliferum (Moench) Schrad. ex Willd., A. nutans L., A. taviilovii Popv & Vved., and A. tricoccum Ait. [3,4]. Ornamental species comprise A. giganteum Regel, A. aflatunense B. Fedtsch., A. karatachiense Regel, A. oreophilum C.A. Mey., A. moly L., A. insubricum Boiss. & Reut., A. macleanii Baker [5]. Within the Allium genus, significant variations exist in the genome size of different species, and Alliums in general are known to possess some of the largest genomes among the plant species. Among Alliums, onions are recognized for having a large genome size of ~16 billion bp [6]. Wild garlic (A. ursinum L.) surpasses this with a genome size of 30.9 billion bp [7]. Allium species feature a diverse biochemical composition, abundant in several bioactive compounds including organosulfur compounds and polyphenols with health benefits such as antioxidant [8], anti-inflammatory [8], antimicrobial [9], anti-obesity [10], anti-diabetic [11], hepatoprotective [11], anticancer [12], cardioprotective [13], neuroprotective [14], and immunomodulatory properties [15].

The production of Allium crops is impacted adversely by several biotic stresses, including insects and fungal, bacterial, and viral diseases [16–18]. A soilborne fungal disease, Fusarium basal rot (FBR), is a major threat to the production of Allium species, causing...
significant yield losses worldwide. The primary *Fusarium* species responsible for FBR can vary depending upon the *Allium* species involved (Table 1). However, FBR is linked to various complexes of several species, such as *Fusarium oxysporum* Schlechtend.:Fr. f. sp. *cepae* W.C. Snyder & H. N. Hans. (FOC), *F. proliferatum* (Matsush.) Nirenberg, *F. solani* (Mart.) Sacc., *F. acuminatum* Ellis & Everhart, *F. equiseti* (Corda) Saccardo, *F. culmorum* (W.G. Smith) Saccardo, *F. falciforme* (Carrión) Summerbell & Schroers, *F. brachygibbosum* Padwick, *F. redolens* Wollenweber, *F. verticillioides* (Saccardo) Nirenberg, *F. acutatum* Nirenberg & O’Donnell, and *F. anthophilium* (Braun) Wollenweber [19–25]. It should be noted that *Fusarium* species names in this review are taken directly from the original literature and thus may not accurately reflect current knowledge of *Fusarium* taxonomy and phylogeny. Indeed, recent research findings have led to significant changes in *Fusarium* taxonomy [26,27].

Bulbing onions, including Welsh onions, are primarily attacked by FOC [28]. Two species, *F. acuminatum* and *F. verticillioides* were found to infect garlic [29], and *F. culmorum* was isolated from infected leek plants [30,31]. FBR has been reported at diverse locations, including Israel [32,33], Vietnam [24], Finland [21], Iran [20], and Burkina Faso [34]. In Mexico, FBR has been reported in onions involving two species, *F. falciforme* and *F. brachygibbosum* [23]. In Italy, France, and Spain, *F. proliferatum* has been identified as the predominant species infecting garlic cloves, causing a loss of up to 30% in Italy [35–37]. The exact losses attributed to FBR have not been extensively reported. However, in shallots, a reduction of about 45% in yield post-harvest and a loss of 12–30% of bulbs during storage have been observed [38].

**Table 1.** *Fusarium* species infecting different *Allium* species.

<table>
<thead>
<tr>
<th><em>Fusarium</em> Species Causing Rot</th>
<th>Affected <em>Allium</em> Species</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Fusarium oxysporum</em> f. sp. <em>cepae</em> (FOC)</td>
<td><em>Allium cepa, Allium sativum, Allium cepa Aggregatum group, Allium fistulosum</em></td>
<td>[19,21,24,28,32–34,38]</td>
</tr>
<tr>
<td><em>Fusarium proliferatum</em></td>
<td><em>Allium cepa, Allium sativum, Allium fistulosum</em></td>
<td>[20,21,24,29,32–34–37]</td>
</tr>
<tr>
<td><em>Fusarium solani</em></td>
<td><em>Allium cepa, Allium sativum, Allium fistulosum</em></td>
<td>[20,24,29,34]</td>
</tr>
<tr>
<td><em>Fusarium acuminatum</em></td>
<td><em>Allium sativum</em></td>
<td>[29]</td>
</tr>
<tr>
<td><em>Fusarium equiseti</em></td>
<td><em>Allium cepa</em></td>
<td>[22]</td>
</tr>
<tr>
<td><em>Fusarium culmorum</em></td>
<td><em>Allium porrum</em></td>
<td>[30,31]</td>
</tr>
<tr>
<td><em>Fusarium falciforme</em></td>
<td><em>Allium cepa</em></td>
<td>[23,34]</td>
</tr>
<tr>
<td><em>Fusarium brachygibbosum</em></td>
<td><em>Allium cepa</em></td>
<td>[23]</td>
</tr>
<tr>
<td><em>Fusarium redolens</em></td>
<td><em>Allium cepa</em></td>
<td>[20,21]</td>
</tr>
<tr>
<td><em>Fusarium verticillioides</em></td>
<td><em>Allium sativum</em></td>
<td>[29]</td>
</tr>
<tr>
<td><em>Fusarium acutatum</em></td>
<td><em>Allium cepa</em></td>
<td>[32–34]</td>
</tr>
<tr>
<td><em>Fusarium anthophilium</em></td>
<td><em>Allium cepa</em></td>
<td>[32]</td>
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</table>

Environmental conditions conducive to FBR development differ depending on the pathogen species. The incidence of *F. proliferatum* was associated with rainy weather, whereas *F. oxysporum* incidence was linked to dry conditions [35]. *Fusarium* species prefer a soil temperature range of 28–32 °C [39]. Symptoms of the disease may vary based on the *Allium* species involved, and the developmental stage of the plant. FBR can exhibit diverse symptoms that include leaf chlorosis, drying leaf tips, damping off of seedlings, basal plate rot, blossom-end rot, clove rot, bulb rot, and plant dieback [40]. Many soilborne pathogens are ubiquitous and can live in the soil for several years due to their survival structures, such as chlamydospores in *Fusarium*. They can be difficult to control because they can also live on dead plant material as saprophytes [41,42]. Several methods have been utilized to control soilborne fungal diseases such as FBR. These control methods have included seed treatments with chemicals or beneficial fungi, treating the soil with chemicals, growing resistant cultivars, adding organic material to the soil to boost beneficial microorganisms, and rotating crops frequently [43,44]. This review summarizes recent advances in methods for controlling *Fusarium* diseases of *Allium* species, including chemical, biological, and cultural approaches, in addition to advancements in breeding FBR-resistant cultivars. In the hope of developing FBR-resistant cultivars, traditional breeding methods
and modern molecular and metabolomics studies to unravel disease resistance mechanisms will be presented.

2. Detection of *Fusarium* Diseases in Alliums

To successfully manage a disease, it is imperative to recognize the species of *Fusarium* involved [45,46]. The initial and crucial step in identifying plant pathogens involves macro- and microscopic observations in vitro. This conventional method, particularly for *Fusarium* species, may encounter limitations, such as a lack of expert mycologists, the existence of closely related species, and the reliance on diverse identification means based on morphological characteristics that can vary based on environmental conditions during culturing [47]. Different techniques to detect *Fusarium* species in infected parts of *Allium* plants include visual and microscopic tools, molecular and biochemical techniques, and machine learning tools, including biosensors.

2.1. Conventional Detection

FBR symptoms in Alliums may be detected visually at three growth stages, i.e., seedling, field growing, and mature bulb stage [25,48,49]. Seedling screening involves observing the damping-off symptoms of seedlings [40]. In the growing phase, symptoms appear in the form of leaf-tip dieback, foliage yellowing, and death of the plant [40]. In onions, FOC attacks the basal plates of mature bulbs, causing brown discoloration that eventually spreads into the scales through secondary pathogenesis [40]. The accuracy of visual detection depends on individual expertise and is subjective. Therefore, fungal isolation and culturing on appropriate artificial growing media for microscopic identification of the sporulating structures, the mycelial morphology, and the growing pattern are important steps in determining the appropriate taxonomic classification of the species involved [49]. Morphological characterization of different *Fusarium* species has been conducted by growing fungal colonies on a potato dextrose agar (PDA) medium and including microscopic observations for hyphae and spore formation for each isolate [32]. Kalman et al. [32] acquired six isolates of different species (FOC, *F. proliferatum*, *F. acutatum*, and *F. anthophilium*) through single-spore culture from the roots and basal plates of two onion cultivars. The colonies displayed a spectrum of colors, varying between species and within the same species. Cultures experienced age-dependent changes, starting with a flat, white surface and developing distinct hues such as purple, white, gray, and occasionally light brown as they matured [32].

2.2. Molecular Detection

Polymerase chain reaction (PCR)-based detection of *Fusarium* species may be used in detection and identification. Recently, very sensitive PCR methods were developed to identify *Fusarium* species in Alliums. For instance, to detect latent infection of *F. oxysporum* in onions as low as 10%, Latvala et al. [50] developed a dual PCR test, that employed fungal-specific and onion-pathogenic *F. oxysporum*-specific Secreted In Xylem (SIX3) primers. When the dual PCR test was applied to a pooled sample containing onion and *Fusarium* DNA, this protocol detected 100 pg of *F. oxysporum* DNA. Sasaki et al. [51] found that the SIX3 gene of FOC in the onion basal plate is a homolog of the *F. oxysporum* f. sp. *lycopersici* (Sacc.) Snyder & Hansen SIX3 gene (91.4% similarity). A primer pair (P1) was therefore designed based on the SIX3 nucleotide differences between these two species. A real-time quantitative PCR (qPCR) with the primer pair P1 was only sensitive in detecting FOC isolated from onion roots and basal plates but not from Welsh onion tissue or other formae speciales of *F. oxysporum*.

Recently, Kalman et al. [32] confirmed different *Fusarium* species using PCR amplification and sequencing using the universal internal transcribed spacer (ITS), the fungus-specific T12 beta-tubulin, and *Fusarium* species-specific translation elongation factor 1 (TEF1) primers. The amplified fragments matched the expected sizes, and the sequences were highly similar (99.4–100%) to known *Fusarium* species. They relied on their highest
sequence similarity scores to identify FOC and successfully amplified a product of the expected length using newly designed FOC-SIX3 primers. They also identified *F. proliferatum* using the partial calmodulin gene sequence (CLPRO) specific to the species. Using inter-simple sequence repeat (ISSR)-PCR, isolates of the same species, *F. acutatum* (B5 and B7) and FOC (B8 and B14), exhibited similar DNA profiles. Dimant and Degani [52] reported a novel real-time qPCR molecular tracking of two FBR pathogens, FOC and *F. acutatum*, found inside onion basal plates. Morphological parameters and real-time qPCR molecular evaluations were conducted 65 and 115 days after sowing while the plants were treated with commercially available fungicides. Real-time qPCR was used to monitor the *Fusarium*-specific TEF1 gene and cytochrome oxidase (COX) gene of mitochondria using forward/reverse and COX F/R gene primers, respectively. This qPCR monitoring helped to identify highly effective fungicides. This method also suggested inter-specific antagonism between the two *Fusarium* species.

2.3. Biochemical Detection

The ability of a plant to emit differential volatile compounds under normal and diseased conditions makes high-throughput screening and early detection of pathogen infection possible. Gas chromatography and mass spectroscopy (GC–MS) are commonly used techniques for this purpose. Besides GC–MS, other novel methods have recently been employed to detect volatile organic compounds (VOCs) as biomarkers for *Fusarium* detection. Wang et al. [53] used solid-phase microextraction (SPME) to identify 30 VOCs related to *F. oxysporum* and *F. proliferatum* activity during onion infection. The examination of VOCs revealed significant variations between the two species and among distinct isolates within each species. Ethanol, ethyl formate, ethyl acetate, 2-methyl-1-propanol, methyl thioacetate, n-propyl acetate, and 3-methyl-1-butanol were identified as potential biomarkers for *Fusarium* species in onion. Infantino et al. [54] identified nine VOCs that differentiated healthy garlic cloves from cloves infected with *F. proliferatum*. Additionally, a principal component analysis of different datasets (healthy/artificially inoculated/naturally infected cloves) proved this technique to be a useful tool for the early detection of disease development. In another experiment, VOC emissions from FOC-infected onion bulbs were studied post-inoculation to establish their relationship with increased metabolic activities [55], and 1-propanethiol, methyl propyl sulfide, and styrene were identified as potential VOC markers for infection. Also, this study demonstrated the extent of FBR infection revealed by VOC emissions that were validated by photos, multispectral image analysis, and real-time qPCR monitoring.

2.4. Imaging and Spectroscopic Techniques

Fungal diseases have long been diagnosed using various imaging and spectroscopic techniques that use specific light wavelengths to differentiate diseased and infected plant tissues. Mandal and Cramer [56] compared visual and digital image analysis to quantify FBR-infected and healthy basal plate tissue of onion cultivars from USDA germplasm. FOC-specific wavelength was first identified using confocal microscopy, which was subsequently used in a stereo fluorescence microscope to capture digital images of diseased and healthy tissues. A high-throughput image segmentation method produced reliable quantification comparable to the visual estimation of FBR infection. Tamburini et al. [57] used near-infrared spectroscopy (NIRS) to identify *F. proliferatum* levels in garlic cloves before planting. Calibration and cross-validation models provided promising predictive ability for disease in unknown peeled samples but failed in unpeeled samples. The presence of peel influenced spectral signature significantly and needs to be incorporated in future models. Recently, Kara et al. [58] demonstrated the utilization of Matrix-Assisted Laser Desorption/Ionization-Time-of-Flight Mass Spectrometry (MALDI-TOF MS) to diagnose different species of *Fusarium* including *F. culmorum*, *F. oxysporum*, *F. proliferatum*, *F. solani*, and *F. verticillioides*. MALDI-TOF MS has been applied for rapidly identifying fungi from
culture media, replacing time-consuming traditional methods, and offering a reliable and accurate alternative to microscopic and other molecular techniques.

2.5. Biosensors

The use of biosensors as a user-friendly, non-destructive, and robust technology for pathogen detection has been gaining momentum. Two recent examples used VOC-based biosensors to detect *Fusarium* in two *Allium* species. Makarichian et al. [59] used an electronic nose (E-nose) system that utilized nine metal oxide semiconductor (MOS) gas sensors to detect aromas of garlic infected with three fungal pathogens, including FOC. When the aroma profiles of the control and infected garlic cloves were compared, the volatile sensors were sensitive enough to detect tissue degradation only 4 days after inoculation compared with the 8 days required for visual disease symptoms to appear. When comparing three different methods of infection classification, such as support vector machine (SVM), linear discriminant analysis (LDA), and backpropagation neural networks (BPNNs), LDA was found to be mostly reliable, with 97.5% accuracy for early detection of all fungi. In the second example, Labanska et al. [60] used a PEN 3 electronic nose system in onion and shallot with 10 MOS gas sensors to distinguish between FOC-infected and healthy during post-harvest disease progression and to calculate the proportion of infected bulbs. The disease classification systems employed were LDA, SVM, and k-nearest neighbors (k-NN). The sensor system effectively distinguished FOC-infected from healthy bulbs stored at 25°C with varying proportions of the infection. LDA and optimized k-NN models showed 89.6% accuracy in classifying disease symptoms.

3. Recent Research on Methods to Control *Fusarium* Diseases in Alliums

3.1. Chemical Methods

Different fungicides as seed or soil treatments have been employed in the past [61], and new chemicals alone or in combination have been studied to control *Fusarium* diseases in Alliums. Previous studies have highlighted the effectiveness of various established fungicides against FOC in onions. Fungicides including Antracol (propineb, 70% WP; Bayer CropScience, Leverkusen, Germany), carbendazim (50% WP; BASF, Ludwigshafen, Germany), copper oxychloride (50% WP; Shenzhen King Quenson Industry Co., Ltd., Shenzhen, China), and Kingmil MZ (metalaxyl + mancozeb, 72% WP; King Wing Chemical Industry Co., Ltd., Shenzhen, China), were tested against FOC both in the lab and the pot experiment using a seedling inoculation method [62]. Carbendazim proved to be the most effective, followed by Antracol. Treatments with 10,000 ppm of Antracol, carbendazim, and copper oxychloride resulted in 100% seedling emergence, indicating their high efficacy. Significant differences in plant growth were observed between treated and untreated plants inoculated with FOC.

Recent studies have focused on identifying the most effective fungicide or new combinations of fungicides to control *F. proliferatum*. In garlic, a study was designed to evaluate the efficacy of three fungicides against *F. proliferatum* and their effect on crop yield with foliar application during crop growth [63]. Seven isolates of the fungus showed different sensitivities to the evaluated fungicides, Flint Max (tebuconazole 50% + trifloxystrobin 25%; Bayer CropScience) being the most effective with EC$_{50}$ of <1 ppm, followed by Luna Experience (fluopyram 20% + tebuconazole 20%; Bayer CropScience), with an EC$_{50}$ of <3 ppm. An in vitro assay exhibited mycelial growth inhibition of 46.01% and 61.61% using 1 ppm of Luna Experience and Flint Max, respectively, with increased inhibition upon increasing the dose to 100 ppm. The fungicide Cabiro Duo (dimethomorph 7.2% + pyraclostrobin 4%; BASF) exhibited an inhibition of about 70% at the highest concentration of 1000 ppm. However, tested fungicides exhibited a limited effectiveness in controlling postharvest rot. A separate study examined the efficacy of six distinct combinations of triazole-class fungicides at various concentrations for controlling *F. proliferatum* and *F. oxysporum* in garlic using in vitro and in vivo experiments [64]. In vivo, garlic cloves were treated with fungicides before planting and then were inoculated with a spore suspension. Among
the six combinations, propiconazole + prochloraz (100%) (BUMPER P, ADAMA Italia, Grassobio, Italy) exhibited the highest inhibition of fungal growth (100%).

3.2. Biological Methods

Various biological agents have demonstrated efficacy in managing FBR across different Allium species (Table 2). In particular, several Trichoderma species with botanical extracts and other beneficial microbes have shown their usefulness as potential biological control agents. However, more experimentation on optimizing the doses, exploring new combinations, and further comparative studies with fungicides including field experiments are needed to have a long-term impact on disease management in Alliums.

Table 2. Biological control agents evaluated against Fusarium species infecting Alliums.

<table>
<thead>
<tr>
<th>Biological Control Agents Alone or in Combinations</th>
<th>References</th>
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<tbody>
<tr>
<td>Trichoderma hamatum, Trichoderma atroviride, Trichoderma gamsii, and Trichoderma harzianum</td>
<td>[65]</td>
</tr>
<tr>
<td>Trichoderma harzianum + fermented Legundi leaf (Vitex trifolia)</td>
<td>[66]</td>
</tr>
<tr>
<td>Saccharomyces cerevisiae and Candida tropicalis</td>
<td>[67]</td>
</tr>
<tr>
<td>Trichoderma viride + arbuscular mycorrhizal fungi (AMF)</td>
<td>[68]</td>
</tr>
<tr>
<td>Trichoderma viride + Pseudomonas fluorescens + Glomus mosseae</td>
<td>[69]</td>
</tr>
<tr>
<td>Several rhizospheric bacterial isolates of Bacillus species</td>
<td>[70]</td>
</tr>
<tr>
<td>Local plant growth-promoting rhizobacteria (PGPR) + mycorrhizae, and phosphate-solubilizing bacteria (PSB)</td>
<td>[71, 72]</td>
</tr>
<tr>
<td>Saccharomyces cerevisiae and Candida tropicalis</td>
<td>[67]</td>
</tr>
<tr>
<td>Trichoderma viride + Pseudomonas fluorescens + Glomus mosseae</td>
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<td>[71, 72]</td>
</tr>
<tr>
<td>Bacillus cereus, Enterobacter xiangfangensis, Bacillus thuringiensis, Alcaligenes faecalis, Pseudomonas putida, and</td>
<td>[73]</td>
</tr>
<tr>
<td>Citrobacter freundii</td>
<td></td>
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<tr>
<td>Pseudomonas protegens, Bacillus subtilis, and Enterobacter cloacae</td>
<td>[74]</td>
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</table>

Trichoderma is one of the most widely used biological agents to control Fusarium diseases of Alliums worldwide. Bunbury-Blanchette et al. [65] used different Trichoderma species, i.e., T. hamatum, T. harzianum, T. gamsii, and T. atroviride that were isolated from local soil to control FBR in onions caused by FOC. They observed several antagonistic effects in the culture bioassays, such as creating a hyphal barrier and inhibiting FOC growth. To enhance the effectiveness of Trichoderma species, several researchers combined other botanical extracts to control FBR successfully. Astiko and Sudantha [66] fermented Legundi leaf (Vitex trifolia) with T. harzianum to control FBR in shallot involving FOC. With only 2.5 mL per plant, the Trichoderma–Legundi biofungicide mixture effectively suppressed 2.73% and 66.57% of FBR in two cultivars, and maximum control was achieved at 10 mL per plant. The effectiveness was likely due to the activities of the secondary metabolites, such as terpenes, alkaloids, steroids, tannins, flavonoids, and saponins.

In addition to Trichoderma, yeast and bacterial strains were also evaluated to control FBR of onion. Saccharomyces cerevisiae and Candida tropicalis effectively inhibited 57.74% and 51.18% of mycelial growth of FOC in vitro [67]. When these two yeasts were tested in the greenhouse, field, and storage conditions with two inoculation methods, i.e., soil application (FOC and yeast) and seedling soaking, both methods produced results comparable to the fungicides. Also, two studies compared the effect of Trichoderma with arbuscular mycorrhizal fungi (AMF) (Glomus mosseae and G. fasiculatum) and bacterial (Pseudomonas fluorescens and Bacillus subtilis) treatments to control FBR in onions. It was found that the combinations of T. viride alone and with the AMF were the most effective in suppressing FOC [68]. In another study, T. viride and Pseudomonas species reduced pathogen mycelial growth by 82.86% and 80.82%, respectively. In comparison, an enhanced reduction of 89.49% in FBR incidence was observed in a greenhouse experiment when these two treatments were combined with the AMF [69].

Several studies found soilborne bacteria to be effective in controlling FBR. Seven local rhizosphere bacterial isolates of Bacillus species were studied as biological control agents for inhibiting FOC in wakegi onion (Allium × proliferum (Moench) Schrad. ex Willd). The bacterial isolates effectively reduced FBR incidence by 68.04%, with a low disease incidence of 31.5% compared with the uninoculated control. This antagonism of Bacillus species to
FOC was due to antibiosis [70]. When applied in combination, other local isolates of plant growth-promoting rhizobacteria (PGPR) and mycorrhizae delayed the FOC incubation period and increased the growth and yield of infected onion plants [71]. Bektas and Kusek [72] utilized isolates of local phosphate-solubilizing bacteria (PSB) to observe their effect on controlling onion basal rot and found 71.5 to 75.7% mycelial growth inhibition in vitro and 77.8% disease suppression in a pot experiment. In another study, Kara et al. [73] employed 18 putative bacterial isolates from healthy onion bulbs, roots, and leaves to control root and basal plate rot of onion caused by *F. proliferatum*. *Bacillus cereus, Enterobacter xiangfangensis, Bacillus thuringiensis, Alcaligenes faecalis, Pseudomonas putida,* and *Citrobacter freundii* were found to be effective. *B. cereus* was the most effective, with 50.56% inhibition of *F. proliferatum*. The effectiveness of these bacterial isolates against *Fusarium* species was due to their indole acetic acid (IAA), siderophore, and protease activities. Zhou [74] compared the efficacy of three strains, i.e., *Pseudomonas protegens*, *B. subtilis*, and *Enterobacter cloacae*, along with a seaweed (*Ascophyllum nodosum* (ANE)) extract individually or in combinations to control onion FBR. In vitro tests documented the maximum effectiveness of *E. cloacae* CAL2 against FOC. On the other hand, in vivo, the application of *E. cloacae* CAL2, ANE, and combinations thereof did not show any positive effects on onion biomass, regardless of the presence or absence of the pathogen. Interestingly, the inclusion of 0.5% ANE negatively affected plant biomass parameters, especially reducing shoot and root fresh weights in onion plants.

### 3.3. Cultural Methods

Control methods for soilborne pathogens that utilize changes in cultural practices seek to manipulate the soil environment directly or indirectly through tillage, irrigation practices, crop rotation, and green manure crops as soil amendments. In the past, the implementation of crop rotation was a common strategy to reduce pathogen levels and mitigate FBR. However, its effectiveness may be compromised, as pathogens can persist in the soil or on the roots of symptomless alternative hosts for prolonged periods, limiting the efficacy of this approach [40].

Recently, two studies investigated the efficacy of different plant species as soil amendments to control FBR. In Ethiopia, green manure amendments that included *Brassica* crops, cabbage (*Brassica oleracea*), Ethiopian mustard (*B. carinata*), rapeseed (*B. napus*), and garden cress (*Lepidium sativum*) were evaluated for controlling FBR [75]. For an in vitro assay, sterilized leaves of these crops were macerated, 0.20 mL was poured into PDA petri dishes, and an FOC plug from a culture plate was transferred onto the plate to measure the radial growth of FOC at 24, 48, and 72 h after incubation. For a greenhouse pot experiment, wheat seeds inoculated with FOC were incorporated into sterilized soil at a rate of 10 g/kg. After raising green manure crops for 50 days, the chopped pieces were incorporated at 75 g/kg of soil, filled in the pots, and allowed to degrade for 45 days before planting shallot bulbs from a susceptible cultivar. For the in vitro study, Ethiopian mustard exhibited the highest inhibition, followed by rapeseed. Disease incidence was significantly reduced by incorporating these green manure amendments in the pot experiment. Both rapeseed and Ethiopian mustard treatments led to a lower incidence of FBR in bulbs compared with the control treatment. Another study assessed the potential of *Chenopodium album* leaves using 1%, 2%, and 3% (w/w) dry leaf biomass treatments as a soil amendment for controlling basal rot in onion caused by FOC [76]. In a pot trial, using a 3% leaf biomass of *C. album* significantly reduced disease incidence by 63% and mortality by 89% compared with the positive control with 100% disease incidence and 92% mortality. To determine the effects of various fractions of methanolic extract on fungal growth, four different solvents were used—n-hexane, chloroform, ethyl acetate, and n-butanol—for a test tube assay with 1 mL of medium and one drop of FOC spores. The chloroform fraction was the most effective in solubilizing the bioactive compounds, exhibiting the strongest antifungal activity by reducing fungal biomass by 96–100%. Two unidentified compounds, A (Rf 0.17) and B (Rf 0.44), isolated from this fraction via thin-layer chromatography (TLC), exhibited significant
antifungal activity. Further research is required to identify the chemical structure and function of these compounds.

3.4. Breeding for FBR Resistance

The development of disease-resistant cultivars is the most cost-effective and sustainable approach to manage any plant disease [77]. Different breeding approaches have been utilized to develop FBR-resistant cultivars and determine the genetic mechanisms of disease resistance (Figure 1).

![Breeding for FBR resistance](image)

**Figure 1.** Different breeding approaches to understand the genetic mechanisms of FBR resistance and to develop FBR-resistant onion cultivars.

3.4.1. Conventional Breeding

Various mechanisms for the inheritance of FBR resistance in onion have been proposed. In 1989, Bacher identified two partially dominant genes (Foc1 and Foc2) that had an additive effect on FBR resistance [78]. In 1991, Tsutsui suggested that a single dominant gene and possibly cytoplasmic genes were involved in FBR resistance [79]. Other researchers have also suggested that FBR resistance could be a multigenic inheritance pattern [48]. Despite numerous findings, the inheritance of FBR resistance is not completely understood yet. Nonetheless, it is recognized as a quantitative mode of inheritance controlled by multiple genes with additive effects [80].

Screening disease-resistant plants using natural infection or artificial inoculation methods is categorized under the conventional breeding approach. Several studies have been conducted to evaluate *Allium* germplasm for FBR susceptibility. Utilizing three virulent FOC isolates (FOC-CBT3, FOC-CBT7, FOC-CBT12), a diverse *Allium* group consisting of onion and garlic accessions was evaluated for FBR resistance with two check cultivars [81]. Transplanted seedlings were inoculated at 10 and 21 days after transplanting by pouring 40 mL of a conidial suspension in pots at a final concentration of $3.0 \times 10^7$ spores/mL. Except for CBT-Ac77, all onion accessions tested were susceptible to FBR, and six garlic accessions exhibited disease levels ranging from 67% to 96% based on their disease severity index (DSI) score. One accession each from garlic (CBT-As153) and Welsh onion (NIC 20231) exhibited complete resistance against the three FOC isolates.

In another study, 85 onion accessions obtained from 23 countries, including susceptible and moderately-resistant cultivars, were tested for FBR susceptibility [82]. A seedling screening was utilized by inoculating silica sand with a virulent FOC isolate, CSC-515, at a final concentration of $1.0 \times 10^4$ spores/g of sterile sand. For mature bulbs, screening involved transversely cutting basal plates and inoculating them with agar plugs of the same
isolate at a concentration of $3.0 \times 10^4$ spores/mL. After 20 days of incubation, FBR severity and incidence were calculated. A significant variation in FBR susceptibility was found among the accessions at both the seedling and the mature bulb stage. Accession PI 656956 demonstrated higher seedling survival than the susceptible check cultivar and exhibited lower FBR severity (4.3 on a 1 to 9 scale) and incidence (41.7%). In a related study that used the same mature bulb screening inoculated with a concentration of $3.0 \times 10^4$ spores/mL, populations of initial and advanced FBR-resistant selections were evaluated for FBR susceptibility. The most advanced selected populations exhibited a reduced disease severity compared with the initial population of the cultivars [83]. Specifically, the most advanced selected populations of the “NuMex Sweetpak” cultivar surpassed the moderately-resistant check “Serrana” in levels of FBR resistance. Additionally, high levels of resistance were observed in cultivated A. fistulosum and A. schoenoprasum accessions, whereas wild Alliums, such as, A. roylei and A. galanthum have shown intermediate levels of resistance [25]. However, there are currently no studies reporting the transfer of FBR resistance from wild to cultivated Alliums.

3.4.2. Molecular Breeding

A molecular genetics approach to disease resistance development is comprised of marker-assisted selection using tightly linked markers to the genes or quantitative trait loci (QTLs) involved in FBR resistance, integrating other molecular techniques with conventional breeding approaches to accelerate genetic gains and understanding the involvement of defense-related genes and other molecules. Studies have also explored the function of defense-related genes in response to FOC infection and their expression profiles. In addition, investigations into the role of microRNAs (miRNAs) have contributed to understanding the mechanisms of disease resistance [84].

Several studies have been conducted recently to identify the QTLs controlling FBR resistance. In a patent by Black et al. [85], a region on linkage group 2 (chromosome 4) was discovered that carries a codominant FBR-resistance allele from the resistant parent. They identified a genomic region characterized by loci NQ0345038 and NQ0257326, linked to FBR resistance. It is essential to explore the involvement of different QTLs in diverse populations for confirmation of results. In another study, Taylor et al. [86] used 892 published Kompetitive allele-specific PCR (KASP) single-nucleotide polymorphism (SNP) markers to genotype genetic variation across 91 founder onion accessions and 765 selected polymorphic markers for further analysis after imputation. They found that chromosomes 1, 6B, and 8 may be involved in FBR resistance, with linked markers mapped to c00676_1004 (149.8 cM), i34519_442 (30.2 cM), and i30594_1021 (1.1 cM), respectively. In a recent study, Straley et al. [80] investigated the sources of FBR resistance through mapping and estimating the genetic effects of resistance. They established segregating families by crossing FBR-resistant and FBR-susceptible inbreds and performed genotype by sequencing (GBS) for F$_2$ and F$_3$ plants. Three QTLs were identified on chromosomes 2 and 4 from segregating families produced from the F$_3$ generation. The codominant FBR resistance was strongly correlated with the marker isotig38484_281 on chromosome 2B. The chromosome 4A marker isotig44683_192 was also linked to FBR resistance, while the W440 allele at the marker isotig31106_505 on chromosome 4C was associated with FBR susceptibility. The study also revealed that the diversity among FOC isolates may hinder the identification of FBR-resistant QTLs, but the identified SNP markers may be useful for future research to understand the mechanism behind FBR resistance.

Different genes of various pathways involved in a plant’s defense activate upon infection. To understand the role of defense-related genes, interactions of the susceptible Takstar F$_1$ hybrid of onion with isolates of two Fusarium species, FOC and F. proliferatum, were investigated at both the seedling and the bulb stage [87]. In this study, the expression of different defense-related genes was analyzed. Reverse-transcription quantitative PCR analysis revealed distinct expression patterns for phenylalanine ammonia-lyase (PAL1, PAL2), lipoxygenase (LOX2), chalcone synthase (CHS), anthocyanidin synthase (ANS), and
pectin methylesterases (PME). The expression patterns of defense-related genes in seedlings were largely influenced by both Fusarium species and the aggressiveness of their isolates, indicating variation in the kinetic patterns of defensive mechanisms. Consequently, plant defensive mechanisms were unable to hinder the Fusarium infection or proliferation in seedlings. A positive correlation between differentially expressed genes and FBR severity was found using a bulb assay, which confirmed the ineffective defense mechanisms against FBR. These findings suggested that the expression of genes cannot solely determine the activation of defense mechanisms; rather, this is influenced by various factors, including environmental conditions, pathogen isolates, and the overall physiological state of the plant.

To delve into more details of plant defensive mechanisms, post-transcriptional regulators (metabolite synthesis) such as the miRNAs, especially in developmental processes, are being researched [88]. In a study by Mahanty et al. [84], miRNA profiles were compared for two types of onions: one FBR-resistant (Ac77) and the other FBR-susceptible (Arka Kirthiman (AK)). They identified 37 differentially expressed miRNAs that played an important role in defending onions against FOC. Three unique miRNAs (ace-miRn3a, ace-miRn8, and ace-minRn11) were identified because of their higher expression levels. One miRNA, ace-miRn3a, exhibited a 5.57-fold increase in its transcript abundance in the resistant line but a 0.41-fold decrease in the susceptible one after infection. Interestingly, the upregulated expression of ace-miRn3a in Ac77 contributed to improved FBR resistance. The levels of its target gene (ACCL_237), that makes transport inhibitor response 1 (TIR1) type F-box proteins, were decreased in Ac77 and AK lines when ace-miRn3a was overexpressed. This result suggests that ace-miRn3a could be used to engineer FOC-resistant onions by reducing fungal colonization by suppressing TIR1 F-box proteins. In a recent study, researchers compared transcript levels of certain genes, including coronatine-insensitive1 (COI1), TIR1, and ethylene response factor (ERF1), and their respective miRNAs in both a susceptible cultivar of A. cepa (Ghermeze Azarshahr) and a resistant wild onion relative (A. asarense R.M. Fritsch & Matin) to identify the major genes involved in the onion defense response against FOC [89]. The study revealed dynamic changes in gene expression during onion’s defense response to FOC following infection. ERF1 gene expression levels increased initially in Ghermeze Azarshahr compared with the control (non-inoculated), later decreased at 48 h post-inoculation (hpi), and increased again at 72 hpi. Contrarily, in A. asarense, the levels decreased initially, increasing at 48 hpi and then again were reduced at 72 hpi. Similarly, COI1 expression increased in Ghermeze Azarshahr at 24 and 72 hpi compared with A. asarense. TIR1 gene expression varied inconsistently, with an increase in Ghermeze Azarshahr at 24 and 48 hpi, but a significant reduction was observed in A. asarense at 24 and 72 hpi. Moreover, miR-393 and miR-774 did not influence the expression of TIR1 and ERF1, respectively, but miR-5629 inversely regulated COI1, being up-regulated in A. asarense and down-regulated in Ghermeze Azarshahr at 72 hpi. These findings highlighted the reciprocal relationship between miRNAs and target gene expression, indicating a regulatory role in the onion defense mechanism.

Similar studies have also been documented in garlic, exploring defense-related genes and the involvement of miRNAs in modulating their defense mechanisms against FBR. A previously identified resistant garlic line CBT-As153 was used to isolate nucleotide-binding-site-leucine-rich-repeats (NBS-LRR) resistance gene analogues (RGAs), structurally similar to known resistance (R) genes, associated with FBR resistance and to assess their nature, degree of sequence variation, and genetic index [90]. Defense signal molecules such as methyl jasmonate (MeJA), abscisic acid (ABA), salicylic acid (SA), and H2O2, and the transcriptional induction kinetics of putative A. sativum resistance gene analogs (AsRGA) associated with FBR resistance were investigated following FOC infection. Most plant disease-resistance (R) genes contain a leucine-rich repeat structure and a highly conserved nucleotide-binding site, that can be used to identify potential candidate genes for FBR resistance. From CBT-As153, 28 NBS sequences were identified as AsRGAs by degenerative primers based on the NBS conserved motif of NBS LRR resistance proteins. Six classes of non-Toll interleukin receptor (non-TIR) subfamily were identified among the AsRGAs.
AsRGAs were found to be differentially expressed in the stem, leaves, and roots, with the higher expression of AsRGA29 in the stem of CBT-As153. Further, the expression of AsRGA29 was more than 20 times higher in CBT-As153 than that of *A. roylei* and *A. fistulosum*. Additionally, the mRNA levels of AsRGA29 significantly increased in response to FOC infection and treatment with the four defensive signaling molecules, SA, MeJA, H$_2$O$_2$, and ABA. These results explain the crucial role of AsRGA29 in mediating many defense-signaling pathways to protect garlic from FOC. This investigation will support the creation of RGA-based markers for genetic mapping in garlic and other Alliums. The specific miRNAs involved in garlic’s defense against FOC remain unexplored. Another investigation was conducted to address this gap by sequencing a small-RNA library derived from FOC-infected resistant garlic cultivar plants [91]. The analysis revealed 45 miRNAs, comprising 39 conserved and six novel sequences, responsive to FOC. These miRNAs were categorized into distinct expression patterns in the susceptible and resistant garlic lines (CBT-As11 and CBT-As153, respectively). Transgenic garlic plants overexpressing miR164a, miR168a, and miR393 exhibited increased resistance against FOC, as demonstrated by reduced fungal growth and increased expression of defense-related genes. Future studies can further investigate the mechanisms underlying FBR resistance and explore the potential of manipulating miRNA expression to improve FBR resistance.

### 3.4.3. Metabolomics-Assisted Breeding

Metabolomics plays an important role in plant breeding for disease resistance by providing unique chemical fingerprints or metabolites. By identifying biomarkers through metabolomics, susceptible and resistant genotypes can be effectively distinguished, aiding in the selection of plants with natural defense against diseases [92,93]. Moreover, investigating metabolic pathways can uncover molecular mechanisms underlying disease resistance. Plants synthesize different primary and secondary metabolites with varying structures [94]. Primary metabolites are important for a plant’s growth and development. On the other hand, secondary metabolites are involved in plants’ defense mechanisms [95,96]. There is a trade-off between these two metabolites, because higher amounts of secondary metabolites contribute towards resistance to biotic and abiotic stresses, negatively impacting the production of primary metabolites [97,98]. Plants produce secondary metabolites such as terpenoids, steroids, alkaloids, glycosides, and saponins [99–101]. Saponins are classified as phytoanticipins, which means that their synthesis in the plant is natural, and the levels change due to external stimuli [102]. Saponins are known as antifungal compounds because their mode of action against fungi comprises the formation of pores and the loss of membrane integrity via making complexes with fungal sterols [103–105] which leads to the leakage of cell contents or the induction of programmed cell death in sensitive fungal cells [106]. The chemical structure of saponins is comprised of two components, aglycone (sapogenin) and glycone (sugar moieties). Saponins can either be steroidal or triterpenoid aglycones attached to sugar moieties [107–109].

*Allium* species are known to produce steroidal saponins with antifungal properties [108, 110–115]. Saponin levels vary across different parts of an onion plant with the root–basal stem plate exhibiting higher content [116–118]. The greater amounts of saponins in the root–basal stem plate could be responsible for defending plants against soilborne pathogens. Different species of *Allium* have been explored to evaluate the antifungal activity of crude and individual saponins. The antifungal activity of saponins against several isolates of FOC was studied in *A. roylei* [116]. A crude saponin extract caused variable growth inhibition for four isolates of FOC (9.11% to 41.33%). The variability in growth inhibition could be due to the ability of some isolates to generate enzymes that degrade saponins into nontoxic molecules. To explore the potential association between FBR resistance and *A. roylei* genes, *A. roylei* was utilized to generate alien addition lines within *A. fistulosum* (genome FF) with extra chromosomes from *A. roylei* (genome RR) [119]. Total saponins were extracted from the roots of *A. fistulosum*, *A. roylei*, an amphidiploid (FFR), and an allotriploid (FFR) to test their antifungal activity at a final concentration of 1000 ppm. Saponins from the allotriploid
exhibited significantly higher antifungal activity against two FOC isolates than \textit{A. fistulosum}. The crude saponins were in much greater quantity in the allotriploid plant roots when compared with the roots of both \textit{A. fistulosum} and the amphidiploid plant. The increased saponin content in the FFR allotriploid could be attributed to the saponin biosynthesis controlled by introgressed genes from \textit{A. roylei}.

Dependency on total saponin levels can be misleading, as certain specific saponins may increase upon infection to protect resistant genotypes, while others, even if their levels rise, may not confer protection. Hence, it is imperative to identify the specific saponins involved in FBR resistance. A spirostane saponin aginoside isolated from \textit{A. nigrum} L. was evaluated for its antifungal activity against seven different soilborne fungi and exhibited partial inhibition of FOC at 400 ppm \cite{118}.

Shallot is a close relative of bulb onion, and a few shallot genotypes were found to be resistant to FBR \cite{38}. The evaluation of antifungal properties of shallot metabolites on several isolates of FOC was explored for the first time \cite{120}. A shallot cultivar, \textit{A. fistulosum}, and a complete set of eight \textit{A. fistulosum}–shallot monosomic addition lines (MALs) were included in the study. Shallot roots exhibited a drastic increase in the levels of saponin content in root exudates 3 days after inoculation with FOC. Complete death of fungal cells was observed using a higher concentration (200 µg/mL) of crude saponins. The FF+2A line among eight MALs, which showed a specific saponin band derived from shallot on TLC, was the most resistant and contained chromosome 2A of shallot, indicating that it may contain genes related to FBR resistance. In another study, total saponins isolated from basal plate tissue of shallots were tested for antifungal activity and exhibited growth inhibition in all fungal isolates with differential response \cite{117}. Two spots on TLC with potential saponins were isolated and tested for antifungal activity in vitro and were recognized as alliospiroside A (ALA) (C_{38}H_{60}O_{12}) and alliospiroside B (ALB) (C_{39}H_{62}O_{13}). Both ALA and ALB demonstrated inhibitory effects on the growth of all tested plant pathogenic fungi in vitro, with ALA exhibiting superior antifungal activity. However, ALA’s efficacy was comparatively lower against \textit{Fusarium} species. The antifungal activity of this spirostanol saponin (ALA) decreased when its sugar chain was removed through acid hydrolysis, indicating that the presence of the sugar chain is crucial for its antifungal activity.

To understand the functional role of the shallot (AA) saponin in enhanced FBR resistance of \textit{A. fistulosum} (FF) against FOC, phytochemical analyses, including RNA-seq, were performed for the first time in Alliums \cite{109}. Shallot (AA), eight MALs, and \textit{A. fistulosum} (FF) were examined to identify candidate genes responsible for producing enzymes in the steroidal saponin biosynthesis pathway. TLC of crude saponin extracts from the roots of AA, MALs, and FF identified a spirostanol saponin compound in FF2A and AA, and two furostanol saponins in AA, FF1A, and FF2A. The highest saponin accumulation was observed in AA, FF1A, and FF2A roots compared with the roots of FF and other MALs. ALA was identified as a major bioactive shallot-specific saponin in FF2A, and its role in FBR resistance was observed in significant mycelial growth inhibition of 39.58% and 53.12% against two FOC strains. Moreover, 50 unigenes were discovered in all plant parts of AA and MALs involved in the biosynthesis of saponins. Comparing AA and MALs with FF, differential gene expression in different plant parts indicated a significant increase in the downstream pathway of saponin synthesis, which included cytochrome P450, glucosyltransferase, and beta-glucosidase genes located on chromosome 2A. This UniGene dataset is an important resource for further molecular and biochemical studies of saponins. By comprehending the genes related to saponin compounds and their biosynthesis pathway, generating plants with distinct saponin profiles with improved FBR resistance would be possible.

4. Conclusions

This review covered recent research in managing FBR in \textit{Allium} species using different methods ranging from chemicals to biocontrol agents alone or in combinations and using different crops as soil amendments. It also covered the progress made in developing detection tools to accurately identify various \textit{Fusarium} species in infected plant parts of
Alliums by employing morphological characterization, PCR techniques, volatile organic compounds as biomarkers, spectroscopic techniques, and biosensors. Additionally, advancements in improving FBR resistance, including conventional breeding, molecular techniques, and metabolomics analyses, have been included. Conventional breeding has advanced by employing artificial inoculation procedures to screen resistant germplasm. Molecular techniques have identified QTLs involved in FBR resistance and the functions of defense-related genes and miRNAs. Moreover, metabolomic analyses have explored the role of saponins as potent antifungal compounds within the plant's natural defense system, offering valuable insights into the mechanism underlying FBR resistance. However, further research is required to unravel the intricate pathways linking antifungal saponins to QTLs associated with FBR resistance.

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**References**


2. Fredotović, Ž.; Puizina, J. Edible *Allium* Species: Chemical Composition, Biological Activity and Health Effects. *Ital. J. Food Sci.* 2019, 31. [CrossRef]


33. Degani, O.; Dimant, E.; Gordani, A.; Graph, S.; Margalit, E. Prevention and Control of Fusarium spp., the Causal Agents of Onion (*Allium cepa*) Basal Rot. *Horticulturae* 2022, 8, 1071. [CrossRef]


35. Mondani, L.; Chiusa, G.; Battilani, P. Fungi Associated with Garlic During the Cropping Season, with Focus on *Fusarium proliferatum* and *F. oxysporum*. *Plant Health Prog.* 2021, 22, 37–46. [CrossRef]

52. Dimant, E.; Degani, O. Molecular Real-Time PCR Monitoring of Onion Fusarium Basal Rot Chemical Control. *J. Fungi* 2023, 9, 809. [CrossRef] [PubMed]
56. Mandal, S.; Cramer, C.S. Comparing Visual and Image Analysis Techniques to Quantify Fusarium Basal Rot Severity in Mature Onion Bulbs. *Horticulturae* 2021, 7, 156. [CrossRef]
64. Mondani, L.; Chiusa, G.; Battilani, P. Chemical and Biological Control of *Fusarium* Species Involved in Garlic Dry Rot at Early Crop Stages. *Eur. J. Plant Pathol.* 2021, 160, 575–587. [CrossRef]
70. Asrul, A. Potential of Local Bacillus spp. Isolates as Wilt Disease Biocontrol Agents for Fusarium oxysporum f. sp. cepae on Allium x Wakegi. Biodiversitas 2023, 24, 4989–4997. [CrossRef]

71. Anton Ciptady, M.; Nisa, C. Control of Fusarium Disease in Onion with Plant Growth Promoting Rhizobacteria (PGPR) and Mycorrhizae and Its Effect on Growth and Yield. J. Wetl. Environ. Manag. 2019, 7, 18–32. [CrossRef]


88. Sabzehzari, M.; Naghavi, M.R. Phyto-MiRNAs-Based Regulation of Metabolites Biosynthesis in Medicinal Plants. Gene 2019, 682, 13–24. [CrossRef]

89. Sabzehzari, M.; Naghavi, M.R. Phyto-MiRNAs-Based Regulation of Metabolites Biosynthesis in Medicinal Plants. Gene 2019, 682, 13–24. [CrossRef]

90. Khanzarinejad, B.; Dashti, F.; Buratti, E.; Mirzaie-asl, A.; Zafari, D.; Romano, M. Changes in the Expression of COI1, TIR1, and ERF1 Genes and Respectative MiRNAs in Fusarium Basal Rot-Stressed Onion. Gene 2024, 905, 148212. [CrossRef]


108. Lanzotti, V.; Romano, A.; Lanzuise, S.; Bonanomi, G.; Scala, F. Antifungal Saponins from Bulbs of White Onion, Allium cepa L. Phytochemistry 2012, 74, 133–139. [CrossRef] [PubMed]


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