

The Open Agriculture Journal

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RESEARCH ARTICLE

Silicon reduces Fusarium Head Blight Development in Barley

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Abstract:

Background:

Silicon (Si) can directly or indirectly enhance plant resistance to fungal pathogens, but no report is available concerning the effectiveness of Si in decreasing Fusarium Head Blight (FHB) disease on barley (*Hordeum vulgare* L.).

Objective and Methods:

The evaluation of Si supplied to barley incorporated into the soil and as a foliar spray against four FHB species under controlled conditions was investigated. In addition, the potential resistance mechanisms related to the reduction of Disease Incidence (DI) and Disease Severity (DS) measured at 7, 14, 21, and 28 days post-inoculation (dpi) were proposed. Four Si concentrations of both a SiO₂ powder incorporated into the soil as a solid source, *i.e.*, of 0.00, 0.50, 1.50 and 3.00 g/kg and a liquid formulation of Si as a foliar spray, *i.e.*, of 0.0, 0.8, 1.7 and 3.4 ppm were tested to study their effect on the development of FHB fungi on two barley moderately resistant “MR” and susceptible “S” cultivars.

Results:

All concentrations of Si did not significantly reduce DI and DS at 7 dpi. The disease reduction was observed with the application of 1.50 g/kg of soil and 1.7 ppm at 14 dpi and increased with time until 28 dpi, however, the other rates had no significant effect. At 28 dpi, solid and foliar treatments reduced DI by 26.6% and 22.9%, respectively, on “MR” cultivar, and by 19.4% and 19.5%, respectively, on “S” cultivar and decreased DS by 20.4% and 19.5%, respectively, on “MR” plants and by 18.8% and 18.4%, respectively, on “S” plants.

Conclusion:

No effects of Si were observed during the initial infection stage; our results suggest that Si triggers defense processes in barley plants in the latest infection stages to diminish DI and DS by affecting mycotoxins synthesis. Si inputs can be a valuable tool in integrated FHB management by reducing the disease development on barley.

Keywords: Disease management, *Hordeum vulgare*, Foliar spraying, Incorporation into the soil, Concentration, Resistance, Silicon-accumulator.

Article History

Received: December 15, 2020

Revised: March 16, 2021

Accepted: April 6, 2021

1. INTRODUCTION

Barley (*Hordeum vulgare* L.) is the fourth most produced cereal crop globally and is cultivated in temperate climate regions. Nearly 140 million tonnes per year are produced worldwide, which are principally used as animal feed (70%) and for beer production (27%) [1]. Barley is susceptible to a wide array of harmful fungal diseases. Fusarium Head Blight (FHB) is an economically important disease of barley and other small grain cereals (*i.e.*, wheat, oat, rye, and triticale) [2]. FHB reduces yield and impairs grain quality particularly due to the

accumulation of Dangerous Mycotoxins (DON), which are harmful to human and animal health. Consequently, the contamination by DON makes barley-harvested kernels unacceptable for the malting and brewing industry [3]. Numerous Fusarium species differing in their predominance and mycotoxin spectra have been associated with FHB disease. By far, the most prevalent species are *F. graminearum* and *F. culmorum*, found in all barley-growing areas [4, 5]. During warm and wet conditions, FHB fungi can penetrate the rachis and spread via direct floret-floret contamination at anthesis. Disease symptoms are recognized by necrotic patches, bleaching of the florets, and discoloured kernels (tan, orange, brown, pink or red) scattered throughout the head [6].

The development and deployment of resistant barley

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cultivars are the simplest and the most effective approach in integrated disease management for decreasing the negative effects of FHB [7]. Two primary categories of polygenic resistance determined quantitatively by several Quantitative Trait Loci (QTLs) in barley to disease infection are generally recognized as Type I (resistant to initial penetration of the pathogen) and Type II (resistant to fungal spread within a spike), with Type I as the predominant type [8]. However, the lack of FHB resistant barley cultivars makes it difficult to achieve complete control of disease due to (1) potential presence of fungal inoculum on crop residues, such as ascospores, macroconidia, chlamydospores, and hyphal fragments, (2) possible persistence of favorable environmental conditions during the flowering stage for FHB infection, (3) complex inheritance of QTLs resistance and (4) significant cultivar-by-environment interaction effects [6 - 8]. Given this evidence, new strategies in the context of integrated disease management need to be developed to diminish losses due to the FHB pathogen complex.

Silicon (Si) has been well-documented to play a vital role in enhancing growth, development, and yield for a wide array of field crops, particularly under various abiotic (*i.e.*, nutrient imbalance, salinity, metal toxicity, water deficit, waterlogging, radiation damage, UV, and temperature extremes) and biotic (*i.e.*, pathogens and insect pests) stresses [9 - 14]. Silicon is also regarded as an environment-friendly compound in relation to soil, fertilizers, and plant nutrition [15]. Several mechanisms have been suggested to explore the enhanced fungal resistance in plants by silicon. Firstly, the polymerization of silicon beneath the cuticle and in the cell walls increases the physical barrier to fungal pathogens. Secondly, silicon plays a metabolic function in the plant–fungal interaction by enhancing the activities of plant defensive enzymes, leading to increased accumulation of defensive compounds, such as phenolics and phytoalexins, and in turn enhancing plant resistance to fungal pathogens. Thirdly, silicon can induce systemic resistance in plants [11, 13, 14, 16]. Thus, the decrease in disease symptom expression is due to the effect of silicon on some components of plant resistance [11].

Barley is a silicon-accumulator monocot having more than 1% dry weight; its absorption capacity from the soil is recorded in the range of 50–150 kg Si/ha [15]. However, there is little information about the potential positive effects of silicon on the resistance of barley to fungal pathogens, except against powdery mildew (*Blumeria graminis*) [17]. The decrease in the powdery mildew disease was due to the fact that Kunoh and Ishizaki study focused on barley leaf tissue [17]. The effect of silicon deposition on leaves might have been stronger against foliar pathogens because silicon played both: physical and biochemical roles [11, 13, 14, 16]. Until recently, no report is available in the literature examining the effectiveness of silicon in reducing the development of FHB fungi on barley. However, in the wheat–FHB association, Yobo *et al.* found that granulated and foliar potassium silicate has a limited potential to decrease *F. graminearum* severity on day 12, after pathogen inoculation as compared to the pathogen-inoculated control treatment in bread (*Triticum aestivum* L.) plants [18].

There is a need to understand how silicon affects the two

components of barley resistance, Type I and Type II, in growth chambers where all biotic and abiotic conditions are strictly controlled. In order to develop a deeper understanding of the pivotal role of silicon in decreasing the negative disease effects in the barley–FHB system, here we aimed to (1) investigate the effect of soluble silicon applied to moderately resistant and susceptible barley plants via incorporation into the soil and via foliar spraying on the reduction of bleaching of spikes (disease incidence) and spikelets (disease severity) caused by four FHB species, *i.e.*, *F. culmorum*, *F. solani*, *F. verticillioides* and *F. equiseti* in a growth chamber, and (2) propose the potential resistance mechanisms related to the reduction of the bleaching of spikes and spikelets.

2. MATERIALS AND METHODS

2.1. Establishment of Aggressiveness Assays under Controlled Conditions

2.1.1. Fungal Isolates, Inoculum Preparation, and Barley Cultivars

Sixteen fungal isolates representing four *Fusarium* species (*F. culmorum* (F1, F2, F3, F28, and F30), *F. verticillioides* (synonym *F. moniliforme*) (F15, F16, F21 and F27), *F. solani* (F7, F20, F26, F29, F31 and F35), and *F. equiseti* (F43)) were sampled from wheat heads showing obvious FHB disease symptoms. Isolates were collected during the 2015 growing season in several localities of the Ghab Plain, one of the principal Syrian wheat production areas. All isolates were morphologically identified based on macroscopic features, such as pigmentations and growth rates over the surface of potato dextrose agar (PDA, HiMedia, HiMedia Laboratories) in 9-cm Petri plates, as well as their microscopic characteristics involving the size of macroconidia, and the presence of microconidia and chlamydospores [19 - 22]. Recently, the 16 FHB fungal isolates were molecularly analyzed by random amplified polymorphic DNA [Sakr, unpublished data]. For ensuring adequate aggressiveness on the tested barley plants, pathogenic responses were quantified with the main *Fusarium* species present in Syria because FHB causative agents had not been collected from the Syrian barley fields until now [19 - 23]. These isolates were stored in sterile distilled water at 4°C or by freezing at -16°C [24].

For inoculum preparation, the isolates were placed on PDA Petri plates and incubated in an incubator (JSPC, JS Research Inc) for 10 days at 22°C under continuous darkness to allow mycelial growth and sporulation. Following growth, 10 ml of sterile distilled water were added to each dish, and the resulting spore suspensions were adjusted to 5×10^4 spores/ml for inoculation, following a count in a cell counting chamber Neubauer hemacytometer (Marienfeld).

FHB isolates were used to inoculate two barley cultivars: Arabi Aswad (AS) and Arabi Abiad (AB), showing varying quantitative resistance levels under *in vitro*, controlled, and field conditions (AB is more susceptible to FHB infection than AS) [19 - 22].

2.1.2. Evaluation of Aggressiveness of FHB Isolates and Resistance of Barley Plants using Artificial Head and Floret Inoculation Assays

Bleaching of spikes (disease incidence, DI) was quantified previously by Sakr for FHB fungi and barley plants using an artificial head inoculation assay in a growth chamber [22]. Here, we analyzed the bleaching of spikelets (disease severity, DS) of fungi and plants in order to use the highly pathogenic isolates for analyzing the effect of foliar and root application of silicon against FHB agents in barley. Methods for point inoculation of central spikelets were conducted to quantify DS of FHB isolates, as described previously by Sakr, to assess quantitative resistance component, DS in the wheat–FHB system [23].

Seeds of AS and AB were surface-sterilized with 5% sodium hypochlorite solution for 8 min and then washed 6 times with sterile distilled water. Subsequently, 8 seeds were sown into 20 × 15 cm pots filled with soil sterilized at 5 k Gray of Gamma Irradiation with Cobalt-60 (60 Co) source assisting a gamma irradiator (ROBO, Russia). We collected a clay soil (57% clay, 39% loam and 2% sand) from Sojji Agricultural Experiment Station (located east of Damascus, Syria, 33°30 N, 36°07 E) with the following traits: pH = 7.8, phosphorus = 13.4 ppm, potassium, sodium, calcium, magnesium = 1.81, 2.99, 33.1, 14 mg/100 g soil, respectively and organic matter = 1.25%. Each plastic pot contained 2 kg of air-dried, sieved (2 mm) soil. The experimental design was a completely

randomized design with 3 replicates for each isolate and cultivar. Three pots per isolate and cultivar were left uninoculated and served as controls. Following emergence, plants were thinned to five seedlings and nitrogen fertilizer, as urea (0.173 g/pot) was applied twice at the stages of plant emergence and tillering. The plants were watered (300 ml/pot) once a week until infection. When the spikes reached 50% anthesis, plants of a pot were sprayed with a spore suspension at 5×10^4 spores/ml for DI evaluations and injected into two adjacent florets (10 μ L at 5×10^4 spores/ml per floret) at the middle of each spike (without wounding) for DS ratings of 16 *Fusarium* isolates (Table 1). The control plants were sprayed with sterile distilled water. Inoculated spikes were covered with polyethylene bags for 48 h (100% relative humidity) to promote infection. The experiment was repeated twice. The 16 *Fusarium* isolates were individually inoculated on AS and AB barley plants in a growth chamber at 20°C day/night temperature and 16/8 h light/dark cycle to measure DI and DS, as indicators of the isolate's aggressiveness. DI (% of symptomatic spikes) was calculated as the percentage of spikes in a plant with visible FHB symptoms at 21 days post-inoculation (dpi). DS (% symptomatic spikelets/spike) was assessed as the mean percentage of the disease in infected heads at 21 dpi on a 1 – 9 scale, as described by Xue *et al.* [5], where 1 < 5%, 2 = 5 – 17%, 3 = 18 – 30%, 4 = 31 – 43%, 5 = 44 – 56%, 6 = 57 – 69%, 7 = 70 – 82%, 8 = 83 – 95% and 9 > 95% of the spikelets with FHB symptoms.

Table 1. Fusarium head blight severity (% symptomatic spikelets/spike) of 16 fungal isolates of four Fusarium head blight species on two cultivars of barley plants (Arabi Aswad and Arabi Abiad) under controlled conditions.

FHB Species	Fungal Isolates	Disease Severity (%)	
		Arabi Aswad	Arabi Abiad
<i>F. culmorum</i>	F1	29cde B	42d A
	F2	33cd A	25fg A
	F3	29cde B	58c A
	F28	20ef B	44d A
	F30	28de B	79a A
<i>F. solani</i>	F7	36bcd B	67bc A
	F20	17f B	36de A
	F26	16f B	28ef A
	F29	29cde B	76ab A
	F31	42b A	36de A
<i>F. verticillioides</i>	F35	58a A	26fg B
	F15	16f A	22fg A
	F16	38bc A	23fg B
	F21	15f B	32ef A
<i>F. equiseti</i>	F27	20ef A	18g A
	F43	66a A	36de B
-	-	P (F) isolates = 3.34E-21	
-	-	P (F) cultivars = 4.1E-11	
-	-	P (F) interactions = 3.59E-21	

According to the Fisher's test, values followed by the same letter are not significantly different at $p < 0.05$; lowercase letters refer to pathogenicity among fungal isolates within each barley cultivar and capital letters to quantitative resistance between the two cultivars within each *Fusarium* isolate, Probability (P (F)) ($p < 0.05$).

2.2. Foliar and Root Application of Silicon against FHB Agents in Barley

2.2.1. Plant Materials and Growth Conditions

Two barley cultivars, AS and AB, “MR” and susceptible “S” to FHB infection, respectively, were used as plant materials in this experiment. Surface-sterilized seeds of each cultivar were sown in 20 × 15 cm plastic pots filled with 2 kg of air-dried, sieved (2 mm) soil (above-mentioned sterilized soil). Barley plants were kept under chamber conditions (20°C day and night temperature, and 16 h of light per day). Following emergence, plants were thinned and fertilized (0.173 g/pot) to avoid nitrogen deficiency by providing urea at two dates: thinning and tillering. The plants were watered (300 ml/pot) once a week until infection.

2.2.2. Inoculation Procedure

Highly pathogenic isolates belonging to four FHB species (*F. culmorum*, *F. solani*, *F. verticillioides*, and *F. equiseti*) causing bleaching of spikes, *i.e.*, F3, F29, F16 and F43 on AS and F3, F29, F16, and F43 on AB and inducing bleaching of spikelets, *i.e.*, F2, F35, F16 and F43 on AS and F30, F29, F21 and F43 on AB, were used to inoculate barley plants [22]. The inoculum was prepared as follows: FHB isolates were grown individually on PDA in 9 cm-Petri plates, placed at an incubator under continuous darkness at 22°C for 10 days to allow mycelial and sporulation growth. Inocula were prepared by adding 10 ml of sterile distilled water to the cultures. Then, conidia were dislodged, harvested, and filtered through two layers of sterile cheesecloth to remove mycelia. Final conidial concentrations at 5×10^4 spores/ml for DI and DS experiments were based on haemocytometer counts. Barley plants were inoculated when each spike reached 50% anthesis with a spore suspension, for bleaching of spikes (DI) evaluations, and were injected into two adjacent florets (10 µL per floret) at the middle of each spike (without wounding), for bleaching of spikelets (DS) evaluations of FHB isolates; the entire spike was covered with a polythene bag for 48 h to create a high level of humidity to promote FHB infection. Non-inoculated plants were treated with sterile distilled water. Bleaching of spikes and spikelets was evaluated based on the visual assessment of blighting at 7, 14, 21, and 28 dpi when plants were at the soft dough stage. DI (% symptomatic spikes) was estimated as the percentage of spikes showing pathogenic symptoms. DS (% symptomatic spikelets/spike) was assessed as the percentage of diseased spikelets on the inoculated spikes with visually detectable disease symptoms on a nine-grade scale, according to Xue *et al.* [5].

2.2.3. Silicon Application

In this study, a SiO₂ powder (Kieselsaure, Carl Roth GmbH + Co. KG) with a minimum silicon content of 99% was used as the silicon source. A SiO₂ powder at four concentrations (0.00, 0.50, 1.50 and 3.00 g/kg) was dispersed as a solid source to the soil prior to planting, and then the seeds were watered (300 ml/pot). The liquid formulation of silicon was first applied as a foliar spray at the three-leaf stage at the rates of 0.0, 0.8, 1.7, and 3.4 ppm. Separate drenches as 250

ml per pot were applied once a week, with silicon concentrations during the period ranging from 7 to 28 dpi

2.2.4. Experimental Design

The experiments were laid out in a completely randomized design with five replications. Each replication consisted of one-pot containing 2 kg of soil and five barley plants per experimental unit. The experiment was repeated twice. The experiments were conducted to quantify the effect of the solid and liquid formulation of silicon on head blight DI and DS with eight treatments for both AS (“MR”) and AB (“S”) and four highly pathogenic FHB isolates: (1) inoculation with FHB pathogens and no silicon application for root treatment, as control, (2) inoculation with FHB pathogen and addition of 0.50 g Si powder, (3) inoculation with FHB pathogens and addition of 1.50 g Si powder, (4) inoculation with FHB pathogens and addition of 3.00 g Si powder, (5) inoculation with FHB pathogens and no silicon application for foliar treatment, as control, (6) inoculation with FHB pathogens and addition of 0.8 ppm Si, (7) inoculation with FHB pathogens and addition of 1.7 ppm Si, and (8) inoculation with FHB pathogens and addition of 3.4 ppm Si.

2.3. Statistical Analyses

The experimental data were shown as means ± standard deviation and subjected to the analysis of variances (ANOVA) using DSAASAT, 2015, version 1.514, Department of Agriculture and Environmental Science, University of Perugia, Italy. The differences were compared using Fisher’s least significant difference test at the 5% level of significance. Comparison among FHB isolates and between AS and AB treated with silicon was made by the contrast procedure using DSAASAT add-in version 2011.

3. RESULTS

3.1. Evaluation of Aggressiveness of FHB Isolates and Resistance of Barley Plants

Distinctive FHB symptoms generated by the 16 fungal isolates were obvious and simple to record in the inoculated spikelets, whereas no symptoms were existent in the control plants (Table 1). FHB symptoms were seen after 7 dpi, and pathogenicity of FHB isolates and disease resistance in AS and AB was scored 21 dpi. On AS, the values for DS ranged from ~16% for the least pathogenic isolates F20 and F26 (*F. solani*), and F15 and F21 (*F. verticillioides*) to 66% for the most pathogenic isolate F43 (*F. equiseti*). On AB, the values for DS ranged from 18% for the least pathogenic isolate F27 (*F. verticillioides*) to 79% for the most pathogenic isolate F30 (*F. culmorum*). Point inoculation of central spikelets carried out to quantify FHB resistance showed statistically significant differences in the resistance of AS and AB, calculated as the average percentage of affected spikelets per spike. The fraction of plants exhibiting FHB symptoms varied from 15% to 66% on AS and from 18% to 79% on AB. The fungus/host interaction for FHB DS was significant. Although AS and AB were differently affected by all tested isolates except for F2 (*F. culmorum*), F31 (*F. solani*), and F15 and F27 (*F. verticillioides*), AB seemed to exhibit more DS scores than AS.

Thus, AS seemed to be more resistant as measured by DS than AB.

3.2. Comparison among Root and Foliar Applications of Silicon

Values (%) of disease incidence (DI) and disease severity (DS) in AS and AB barley plants treated with several rates of solid and foliar formulation of silicon have been shown in Tables 2 and 3, respectively. The initial head blight symptoms, identified as bleaching of spikes and spikelets, became obvious on plants supplied or not with silicon as they were rated at 7 dpi. Non-inoculated barley plants treated with sterile distilled water did not show FHB symptoms (Fig. 1). The distinctive FHB symptoms manifested and rapidly developed to progressive bleaching of spikes and spikelets over time until 28 dpi. At 7 dpi, not all root and foliar silicon applications led to a significant reduction ($p > 0.05$) in DI and DS on both AS and AB, regardless of FHB isolates. However, application of SiO₂

powder at a concentration of 1.5 g/kg and liquid silicon at a concentration of 1.7 ppm consistently decreased DI and DS compared to the control at 14 dpi, and a greater decrease in DI and DS was observed over time at 21 and 28 dpi (Tables 2 and 3). Compared to the control, mean values for the decrease in DI and DS of the four tested isolates with respect to root (R) and foliar (F) applications at 14, 21 and 28 dpi were 15.6 ± 0.8%, 20.7 ± 3.4% and 23.6 ± 2.8% (DI/R), 15.0 ± 0.6%, 20.6 ± 3.3% and 22.9 ± 3.2% (DI/F), 12.0 ± 0.6%, 17.4 ± 3.1% and 20.4 ± 2.3% (DS/R), 12.3 ± 0.6%, 17.0 ± 2.7% and 19.5 ± 2.9% (DS/F) for AS and 12.7 ± 0.6%, 16.2 ± 2.0% and 19.4 ± 2.3% (DI/R), 13.1 ± 0.8%, 15.7 ± 2.1% and 19.5 ± 1.9% (DI/F), 11.0 ± 0.7%, 15.4 ± 0.5% and 18.8 ± 0.8% (DS/R), 11.0 ± 0.5%, 15.5 ± 1.4% and 18.4 ± 1.4% (DS/F) for AB. Regarding AS and AB, no significant DI and DS differences among other root applications at the rates of 0.50 and 3.00 g/kg and foliar treatments at the rates of 0.8 and 3.4 ppm were exhibited with respect to three time periods (in days) post-inoculation (14, 21 and 28 dpi).

Table 2. Disease incidence (% symptomatic spikes) of four Fusarium head blight species in two cultivars of barley plants (Arabi Aswad and Arabi Abiad) treated with root and foliar applications of silicon grown under controlled conditions.

Cultivar	FHB*	dpi**	Silicon Treatments							
			Root Applications (g/kg)				Foliar Applications (ppm)			
			0.00	0.50	1.50	3.00	0.0	0.8	1.7	3.4
Arabi Aswad	FC	7	11.6 ± 0.9a	12.0 ± 0.7a	11.6 ± 0.5a	11.2 ± 0.8a	12.0 ± 1.0a	11.6 ± 0.5a	11.4 ± 0.5a	11.4 ± 1.1a
		14	22.2 ± 0.8a	22.2 ± 0.8a	18.8 ± 1.0b	22.0 ± 0.7a	21.2 ± 0.8a	22.2 ± 0.8a	18.5 ± 1.0b	22.0 ± 1.0a
		21	34.2 ± 0.8a	33.6 ± 1.1a	25.8 ± 0.8b	34.4 ± 1.1a	34.0 ± 0.7a	34.4 ± 0.5a	26.0 ± 1.2b	34.6 ± 0.5a
		28	40.6 ± 0.5a	39.4 ± 0.9a	30.0 ± 0.7b	40.4 ± 0.5a	40.4 ± 0.5a	39.4 ± 0.9a	29.7 ± 1.0b	40.6 ± 0.5a
	FS	7	9.0 ± 0.7a	9.4 ± 0.9a	9.4 ± 0.9a	9.4 ± 0.9a	9.4 ± 0.9a	9.4 ± 0.5a	9.0 ± 0.7a	9.2 ± 0.8a
		14	30.2 ± 0.8a	30.4 ± 0.5a	26.2 ± 0.4b	30.4 ± 0.5a	30.8 ± 0.8a	30.2 ± 0.8a	26.0 ± 1.0b	30.6 ± 0.5a
		21	44.8 ± 0.8a	44.0 ± 1.2a	36.8 ± 0.8b	44.4 ± 0.9a	45.2 ± 1.1a	44.6 ± 1.1a	36.9 ± 1.0b	44.6 ± 1.1a
		28	50.2 ± 0.4a	50.6 ± 0.5a	40.2 ± 1.3b	50.6 ± 0.9a	49.8 ± 0.8a	49.8 ± 1.1a	39.2 ± 1.3b	49.6 ± 0.9a
	FV	7	15.0 ± 1.0a	15.0 ± 0.7a	15.2 ± 0.8a	15.0 ± 0.7a	14.8 ± 0.8a	14.2 ± 0.8a	14.8 ± 0.8a	15.0 ± 0.7a
		14	15.4 ± 1.1a	14.8 ± 0.8a	13.1 ± 1.0b	15.0 ± 1.0a	15.0 ± 1.2a	15.4 ± 1.1a	13.1 ± 0.8b	15.0 ± 1.4a
		21	38.0 ± 0.7a	37.8 ± 1.1a	31.4 ± 1.0b	38.2 ± 0.8a	37.0 ± 1.2a	38.2 ± 0.8a	31.5 ± 1.2b	37.2 ± 0.8a
		28	55.8 ± 0.8a	56.2 ± 0.8a	44.8 ± 0.7b	55.4 ± 1.1a	55.6 ± 0.9a	55.4 ± 1.1a	44.1 ± 0.7b	56.0 ± 1.0a
	FE	7	13.6 ± 0.5a	13.4 ± 0.5a	13.8 ± 0.8a	13.4 ± 0.9a	13.6 ± 0.9a	13.2 ± 0.8a	13.0 ± 0.7a	13.0 ± 0.7a
		14	19.0 ± 1.0a	19.8 ± 0.8a	16.1 ± 0.8b	19.4 ± 0.5a	19.6 ± 0.5a	19.0 ± 1.0a	15.7 ± 0.8b	18.6 ± 0.9a
		21	51.2 ± 0.8a	51.2 ± 1.3a	39.6 ± 0.9b	51.0 ± 0.7a	51.2 ± 1.3a	51.8 ± 1.3a	39.6 ± 1.1b	50.8 ± 0.8a
		28	52.6 ± 0.5a	52.2 ± 0.8a	39.0 ± 0.6b	52.8 ± 1.1a	52.2 ± 0.4a	52.2 ± 0.8a	38.9 ± 0.8b	52.2 ± 1.1a
Arabi Abiad	FC	7	19.2 ± 0.8a	19.4 ± 0.9a	19.6 ± 1.1a	19.4 ± 1.1a	19.6 ± 1.1a	19.6 ± 1.1a	19.6 ± 1.1a	19.6 ± 1.1a
		14	45.0 ± 0.7a	45.2 ± 0.8a	38.6 ± 0.5b	44.8 ± 0.4a	44.4 ± 0.9a	44.0 ± 1.4a	38.4 ± 0.5b	44.0 ± 1.0a
		21	84.6 ± 1.1a	85.2 ± 0.4a	68.6 ± 0.5b	84.8 ± 1.1a	84.6 ± 0.9a	84.8 ± 0.8a	68.6 ± 0.5b	84.2 ± 0.4a
		28	78.0 ± 0.7a	78.2 ± 0.8a	61.0 ± 0.7b	78.4 ± 1.1a	78.2 ± 0.4a	78.6 ± 0.9a	60.8 ± 0.8b	78.4 ± 0.9a
	FS	7	14.8 ± 0.8a	15.0 ± 1.0a	15.4 ± 1.1a	15.2 ± 0.8a	15.2 ± 1.3a	15.0 ± 0.7a	15.0 ± 1.1a	15.2 ± 1.3a
		14	33.8 ± 0.8a	34.0 ± 1.2a	29.4 ± 0.9b	34.2 ± 0.8a	34.0 ± 1.0a	33.6 ± 0.9a	29.4 ± 0.3b	33.6 ± 1.3a
		21	64.8 ± 0.8a	65.0 ± 0.7a	55.2 ± 0.8b	65.2 ± 0.8a	65.2 ± 0.8a	65.4 ± 0.5a	55.4 ± 0.9b	65.0 ± 1.0a
		28	60.6 ± 0.5a	60.8 ± 0.4a	50.4 ± 0.5b	60.8 ± 0.4a	60.8 ± 0.8a	60.4 ± 0.5a	49.6 ± 0.5b	60.8 ± 1.5a

(Table 2) contd....

Cultivar	FHB*	dpi**	Silicon Treatments							
			Root Applications (g/kg)				Foliar Applications (ppm)			
			0.00	0.50	1.50	3.00	0.0	0.8	1.7	3.4
Arabi Abiad	FV	7	12.0 ± 0.7a	12.2 ± 1.1a	12.2 ± 0.8a	12.4 ± 1.1a	12.2 ± 0.4a	12.4 ± 0.5a	11.6 ± 0.5a	12.2 ± 1.1a
		14	20.6 ± 0.5a	21.0 ± 0.7a	18.0 ± 1.0b	20.8 ± 0.4a	20.8 ± 1.1a	21.0 ± 0.7a	18.5 ± 0.4b	20.6 ± 1.1a
		21	36.6 ± 0.5a	36.2 ± 0.8a	31.2 ± 0.8b	36.8 ± 0.4a	37.2 ± 0.8a	37.0 ± 0.7a	31.6 ± 0.5b	37.0 ± 0.7a
		28	41.6 ± 0.9a	41.4 ± 0.9a	34.0 ± 0.7b	41.8 ± 1.1a	41.0 ± 0.7a	41.2 ± 0.8a	34.2 ± 0.8b	41.0 ± 1.2a
	FE	7	9.8 ± 0.8a	9.6 ± 1.1a	10.2 ± 0.8a	10.0 ± 0.7a	10.2 ± 0.6a	10.0 ± 0.7a	10.2 ± 0.8a	9.6 ± 0.9a
		14	15.6 ± 0.5a	15.8 ± 0.8a	13.4 ± 0.5b	15.8 ± 0.8a	16.0 ± 0.7a	16.2 ± 0.8a	13.9 ± 0.1b	15.8 ± 1.1a
		21	19.6 ± 0.5a	19.8 ± 0.8a	15.8 ± 0.4b	19.8 ± 0.8a	20.0 ± 1.4a	20.4 ± 0.9a	17.5 ± 0.5b	20.2 ± 1.1a
		28	30.2 ± 0.8a	30.0 ± 1.0a	23.8 ± 1.1b	29.8 ± 1.3a	30.2 ± 0.8a	30.4 ± 1.1a	23.8 ± 0.4b	30.2 ± 1.1a

Abbreviations: *FHB: Fusarium head blight species, FC: *F. culmorum*, FS: *F. solani*, FV: *F. verticillioides*, FE: *F. equiseti*, **dpi: days post-inoculation. Values were represented as means ± standard deviation of five replicates. Values for the same cultivar, same FHB species, and same period of days after inoculation among root and foliar application in the same line with the same letter were not significantly different based on Fisher's test at $p < 0.05$.

Table 3. Disease severity (% symptomatic spikelets/spike) of four Fusarium head blight species in two cultivars of barley plants (Arabi Aswad and Arabi Abiad) treated with root and foliar applications of silicon grown under controlled conditions.

Cultivar	FHB*	dpi**	Silicon Treatments							
			Root Applications (g/kg)				Foliar Applications (ppm)			
			0.00	0.50	1.50	3.00	0.0	0.8	1.7	3.4
Arabi Aswad	FC	7	9.2 ± 0.8a	9.6 ± 1.1a	9.4 ± 0.9a	9.8 ± 0.4a	9.6 ± 0.9a	9.4 ± 0.9a	9.4 ± 0.4a	9.2 ± 0.8a
		14	18.4 ± 1.1a	18.8 ± 1.3a	16.2 ± 0.4b	18.8 ± 0.8a	18.6 ± 1.3a	18.4 ± 0.9a	16.4 ± 0.9b	18.0 ± 0.7a
		21	33.4 ± 0.5a	33.0 ± 0.7a	26.2 ± 0.8b	33.4 ± 0.5a	33.2 ± 0.8a	33.4 ± 1.1a	25.6 ± 0.5b	33.2 ± 0.8a
		28	35.0 ± 0.7a	34.8 ± 0.4a	26.8 ± 0.4b	35.0 ± 0.7a	35.0 ± 0.7a	34.8 ± 1.1a	27.0 ± 0.7b	34.6 ± 0.5a
	FS	7	14.8 ± 0.8a	15.0 ± 1.2a	15.0 ± 1.0a	15.2 ± 0.8a	15.4 ± 0.5a	15.2 ± 0.8a	15.0 ± 1.0a	14.8 ± 0.8a
		14	32.0 ± 0.7a	31.6 ± 1.1a	28.0 ± 0.7b	32.4 ± 0.5a	32.2 ± 0.8a	31.8 ± 0.8a	28.2 ± 1.3b	31.6 ± 1.1a
		21	57.6 ± 0.5a	58.0 ± 0.7a	49.4 ± 1.1b	57.6 ± 0.5a	57.0 ± 1.2a	56.6 ± 1.1a	48.6 ± 0.5b	56.8 ± 1.1a
		28	55.8 ± 0.8a	55.4 ± 0.5a	45.6 ± 0.5b	55.4 ± 0.5a	55.0 ± 0.7a	54.8 ± 0.8a	44.8 ± 0.8b	45.6 ± 1.5a
	FV	7	12.0 ± 1.0a	12.6 ± 0.5a	12.8 ± 0.8a	12.2 ± 1.3a	12.2 ± 1.3a	12.4 ± 0.9a	12.2 ± 1.3a	12.0 ± 1.0a
		14	21.0 ± 1.2a	21.2 ± 1.1a	18.2 ± 0.4b	20.8 ± 0.8a	21.4 ± 1.1a	21.0 ± 1.0a	18.8 ± 0.4b	20.6 ± 0.5a
		21	38.0 ± 0.7a	38.4 ± 0.5a	32.6 ± 0.5b	38.0 ± 0.7a	38.6 ± 0.9a	38.2 ± 0.4a	32.6 ± 0.5b	39.0 ± 1.0a
		28	44.2 ± 0.8a	43.8 ± 0.8a	37.4 ± 0.5b	43.8 ± 0.8a	44.0 ± 0.7a	43.8 ± 1.1a	35.8 ± 1.1b	45.6 ± 0.5a
	FE	7	15.0 ± 0.7a	15.2 ± 0.8a	14.6 ± 1.1a	14.6 ± 1.1a	14.6 ± 0.5a	14.8 ± 0.8a	15.2 ± 1.1a	15.0 ± 0.7a
		14	35.2 ± 0.8a	34.8 ± 0.8a	31.0 ± 0.7b	35.4 ± 0.5a	34.8 ± 0.8a	34.8 ± 0.4a	31.6 ± 1.1b	35.6 ± 0.5a
		21	65.2 ± 0.8a	65.4 ± 0.5a	53.4 ± 0.9b	65.2 ± 0.8a	65.0 ± 1.0a	64.6 ± 0.9a	53.4 ± 0.5b	64.6 ± 0.9a
		28	60.8 ± 0.8a	60.4 ± 1.1a	47.2 ± 0.8b	60.6 ± 0.9a	60.6 ± 0.9a	60.8 ± 1.1a	47.2 ± 0.4b	60.4 ± 1.1a
Arabi Abiad	FC	7	24.6 ± 0.5a	25.0 ± 0.7a	24.4 ± 0.5a	24.2 ± 0.8a	24.0 ± 1.2a	24.8 ± 0.8a	24.6 ± 0.5a	24.8 ± 0.8a
		14	32.6 ± 1.1a	33.0 ± 0.7a	28.8 ± 0.4b	32.8 ± 1.3a	32.0 ± 0.7a	32.2 ± 0.8a	28.4 ± 0.5b	31.8 ± 0.4a
		21	79.4 ± 0.5a	79.2 ± 0.4a	65.6 ± 0.9b	79.0 ± 0.7a	78.6 ± 0.5a	78.8 ± 0.8a	65.6 ± 0.9b	78.4 ± 0.9a
		28	74.0 ± 0.7a	74.2 ± 0.4a	58.2 ± 0.8b	73.8 ± 1.1a	74.4 ± 0.5a	74.0 ± 1.2a	58.0 ± 0.7b	74.6 ± 0.5a
	FS	7	20.6 ± 0.5a	21.0 ± 0.7a	20.4 ± 0.5a	20.2 ± 0.8a	20.2 ± 0.8a	20.8 ± 0.8a	20.4 ± 0.5a	20.4 ± 0.9a
		14	41.0 ± 0.7a	40.8 ± 1.1a	36.4 ± 0.5b	41.2 ± 0.4a	40.8 ± 0.8a	41.0 ± 0.7a	36.5 ± 0.5b	40.6 ± 0.9a
		21	75.8 ± 0.4a	75.8 ± 0.8a	64.4 ± 0.5b	75.4 ± 0.9a	75.6 ± 0.9a	75.8 ± 0.8a	64.8 ± 0.4b	75.4 ± 1.1a
		28	80.0 ± 0.7a	80.2 ± 0.4a	65.2 ± 0.8b	79.8 ± 0.8a	80.2 ± 0.8a	80.0 ± 0.7a	65.8 ± 1.1b	80.4 ± 0.9a
	FV	7	14.4 ± 0.5a	14.8 ± 0.8a	14.8 ± 0.8a	14.0 ± 1.2a	14.6 ± 0.9a	14.8 ± 0.8a	14.6 ± 0.9a	14.2 ± 0.4a
		14	19.8 ± 0.8a	20.0 ± 0.7a	17.6 ± 0.5b	20.0 ± 0.7a	19.8 ± 0.4a	20.0 ± 0.7a	17.4 ± 0.5b	19.6 ± 0.5a
		21	31.4 ± 0.9a	31.8 ± 0.4a	27.4 ± 0.5b	31.0 ± 1.0a	32.0 ± 0.7a	32.2 ± 0.4a	27.4 ± 0.5b	31.8 ± 0.8a
		28	34.6 ± 0.5a	34.8 ± 0.4a	28.7 ± 0.3b	34.2 ± 1.3a	34.6 ± 0.5a	34.4 ± 0.9a	28.8 ± 0.3b	34.8 ± 0.4a
	FE	7	15.8 ± 0.8a	16.0 ± 0.7a	16.0 ± 1.2a	16.0 ± 0.7a	16.2 ± 0.8a	16.0 ± 0.7a	16.2 ± 0.8a	15.6 ± 1.1a
		14	24.0 ± 0.7a	24.2 ± 1.1a	21.4 ± 0.9b	24.2 ± 1.1a	23.4 ± 0.9a	23.8 ± 0.4a	21.0 ± 0.7b	23.2 ± 0.8a
		21	35.2 ± 0.8a	35.4 ± 0.9a	29.6 ± 0.4b	34.8 ± 0.8a	35.6 ± 0.5a	35.4 ± 0.9a	29.6 ± 0.5b	35.4 ± 0.5a
		28	39.6 ± 0.5a	39.8 ± 0.4a	32.5 ± 0.4b	40.0 ± 0.7a	39.6 ± 0.5a	39.4 ± 0.9a	32.0 ± 0.7b	39.8 ± 0.8a

Abbreviations: *FHB: Fusarium head blight species, FC: *F. culmorum*, FS: *F. solani*, FV: *F. verticillioides*, FE: *F. equiseti*, **dpi: days post-inoculation. Values were represented as means ± standard deviation of five replicates. Values for the same cultivar, same FHB species, and same period of days after inoculation among root and foliar application in the same line with the same letter were not significantly different based on Fisher's test at $p < 0.05$.

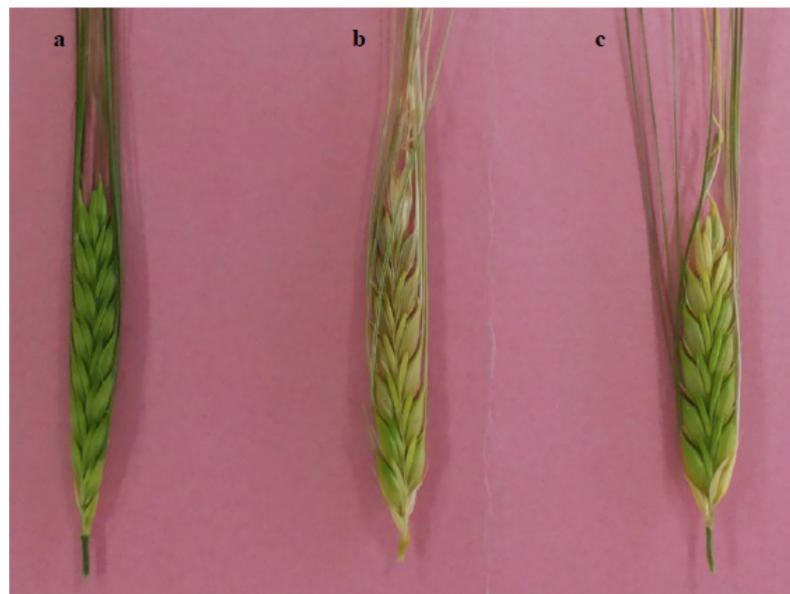


Fig. (1). Root application with silicon enhances barley resistance to Fusarium head blight. FHB disease suppression on Arabi Abiad barley heads in response to adding a 1.50 g/kg of silicon to the soil at 21st day post-inoculation in a growth chamber; (a) a non inoculated barley head treated with sterile distilled water, (b) a barley head inoculated with FHB pathogen using an artificial head inoculation assay and no silicon application for root treatment and (c) a barley head inoculated with FHB pathogen using an artificial head inoculation assay and addition of 1.50 g Si powder to soil.

Table 4. Comparisons among reductions in disease incidence and disease severity (% of control, inoculated with FHB pathogen and no addition of silicon) for four Fusarium head blight species with respect to three periods in days post-inoculation in two cultivars of barley plants (Arabi Aswad and Arabi Abiad) grown under controlled conditions.

Cultivar	Disease Incidence (% of control)				Cultivar	Disease Incidence (% of control)			
Arabi Aswad	Root Application (1.5 g/kg)				Arabi Abiad	Root Application (1.5 g/kg)			
	FC	FS	FV	FE		FC	FS	FV	FE
	14					14			
	16.0 ± 0.7a	15.2 ± 0.4a	15.8 ± 0.8a	15.4 ± 1.1a		13.0 ± 0.7a	12.4 ± 0.5a	12.6 ± 0.5a	12.6 ± 0.5a
	21					21			
	24.4 ± 0.5a	17.2 ± 0.8b	17.6 ± 0.5b	23.6 ± 0.5a		18.8 ± 0.8a	14.6 ± 0.5c	14.6 ± 0.5c	17.0 ± 1.2b
28				28					
26.4 ± 0.9a	21.4 ± 0.5b	20.6 ± 0.5b	26.0 ± 1.0a	21.8 ± 0.4a	17.4 ± 0.5b	17.2 ± 0.4b	21.2 ± 1.3a		
Arabi Aswad	Foliar Application (1.7 ppm)				Arabi Abiad	Foliar Application (1.7 ppm)			
	FC	FS	FV	FE		FC	FS	FV	FE
	14					14			
	15.4 ± 0.5a	14.8 ± 0.8a	14.6 ± 0.5a	15.2 ± 0.4a		13.6 ± 0.5a	12.8 ± 0.8a	12.6 ± 0.5a	13.2 ± 0.8a
	21					21			
	24.6 ± 0.5a	17.8 ± 0.4c	17.2 ± 0.8c	22.6 ± 0.9b		18.4 ± 0.9a	13.8 ± 0.8c	14.0 ± 0.7c	16.6 ± 0.5b
28				28					
26.2 ± 0.4a	19.6 ± 0.5b	20.0 ± 0.7b	25.8 ± 0.8a	21.6 ± 1.1a	17.4 ± 0.5b	18.2 ± 0.4b	20.6 ± 0.9a		
Cultivar	Disease severity (% of control)				Cultivar	Disease severity (% of control)			
Arabi Aswad	Root application (1.5 g/kg)				Arabi Abiad	Root application (1.5 g/kg)			
	FC	FS	FV	FE		FC	FS	FV	FE
	14					14			
	11.8 ± 0.4a	12.2 ± 0.4a	12.0 ± 0.7a	12.0 ± 0.7a		11.0 ± 0.7a	10.6 ± 0.5a	11.6 ± 0.5a	10.6 ± 0.5a
	21					21			
	21.8 ± 0.8a	14.8 ± 0.4c	14.4 ± 0.5c	18.4 ± 0.5b		16.6 ± 0.5a	14.4 ± 0.5b	14.8 ± 0.8b	16.0 ± 0.7a
28				28					
22.8 ± 0.8a	18.2 ± 0.8b	18.4 ± 0.5b	22.4 ± 0.5a	20.6 ± 0.5a	17.4 ± 0.5c	17.6 ± 0.9c	18.8 ± 0.8b		

(Table 4) contd....

Cultivar	Disease Incidence (% of control)				Cultivar	Disease Incidence (% of control)			
Arabi Aswad	Foliar Application (1.7 ppm)				Arabi Abiad	Foliar Application (1.7 ppm)			
	FC	FS	FV	FE		FC	FS	FV	FE
	14					14			
	12.0 ± 0.7a	12.4 ± 0.5a	12.6 ± 0.5a	12.0 ± 0.7a		11.2 ± 0.4a	10.8 ± 0.4a	11.2 ± 0.4a	10.6 ± 0.5a
	21					21			
	20.7 ± 0.8a	14.8 ± 0.8c	14.4 ± 0.5c	18.0 ± 0.7b		16.8 ± 0.8a	14.2 ± 0.4b	14.4 ± 0.5b	16.6 ± 0.5a
	28					28			
22.8 ± 0.8a	17.4 ± 0.5b	17.0 ± 0.7b	22.6 ± 0.9a	20.4 ± 0.5a	17.4 ± 0.9c	17.2 ± 0.4c	18.6 ± 0.5b		

Abbreviations: FC: *F. culmorum*, FS: *F. solani*, FV: *F. verticillioides*, FE: *F. equiseti*, 14, 21 and 28 correspond to periods in days post-inoculation. Values are means ± standard deviation of five replicates. Values for the same cultivar and same period of inoculation among four FHB species with respect to root and foliar application in the same line with the same letter are not significantly different based on Fisher's test at $p < 0.05$.

3.3. Comparison of the Responses of the Four FHB Species to Silicon Applications

Table 4 presents values (% of control) of reductions of DI and DS of four FHB species treated with amounts of soil and foliar formulations of silicon at the concentrations of 1.5 g/kg and 1.7 ppm, respectively. Contrast analysis revealed that DI and DS reductions were not significant among the four FHB species regardless of silicon treatment and barley cultivar at 14 dpi. With progressive inoculation of spikes and spikelets at 21 and 28 dpi on both AS and AB, contrast analysis showed that DI and DS reduction were the highest with *F. culmorum* followed by *F. equiseti* and the least with *F. solani* and *F. verticillioides*. Compared to the control, mean values for the reductions in DI and DS on AS and AB with respect to root (R) and foliar (F) applications at 21 and 28 dpi were 21.6 ± 2.9% and 24.1 ± 2.4% (DI/R), 21.5 ± 3.1% and 23.9 ± 2.4% (DI/F), 19.2 ± 2.7% and 21.7 ± 1.3% (DS/R), 18.8 ± 2.1% and 21.6 ± 1.4% (DS/F) for *F. culmorum* and 20.3 ± 3.4% and 23.3 ± 2.8% (DI/R), 19.6 ± 3.0% and 23.2 ± 2.7% (DI/F), 17.2 ± 1.3%

and 20.6 ± 1.9% (DS/R), 17.3 ± 0.9% and 20.6 ± 1.1% (DS/F) for *F. equiseti* and 15.9 ± 1.4% and 19.4 ± 2.1% (DI/R), 15.8 ± 2.1% and 18.5 ± 1.2% (DI/F), 14.6 ± 0.5% and 17.8 ± 0.7% (DS/R), 14.5 ± 0.7% and 17.4 ± 0.7% (DS/F) for *F. solani* and 16.1 ± 1.6% and 18.9 ± 1.8% (DI/R), 15.6 ± 1.7% and 19.1 ± 1.0% (DI/F), 14.6 ± 0.7% and 18.0 ± 0.8% (DS/R), 14.4 ± 0.5% and 17.1 ± 0.5% (DS/F) for *F. verticillioides*.

3.4. Comparison of the Responses of AS and AB Barley Plants to Silicon Applications

Values (% control) of reductions in DI and DS between "MR" cv. AS and "S" cv. AB infected with four FHB species and treated with amounts of soil, and foliar formulations of silicon at the concentrations of 1.5 g/kg and 1.7 ppm are shown in Table 5. Silicon application decreased DI and DS on AS and AB during the progressive infection starting from 14 dpi. Contrast analysis indicated that DI and DS reductions were higher in AS than DI and DS reductions in AB, irrespective of FHB species and silicon applications at 14, 21, and 28 dpi.

Table 5. Comparisons among reductions in disease incidence and disease severity (% of control, inoculated with FHB pathogen and no addition of silicon) between two barley cultivars (Arabi Aswad (AS) and Arabi Abiad (AB)) with respect to root and foliar applications of silicon infected with four Fusarium head blight species grown under controlled conditions.

dpi*	-	Disease Incidence (% of control)/Root Application (1.5 g/kg)	Disease Incidence (% of control)/Foliar Application (1.7 ppm)	
14	FC	AS	16.0 ± 0.7a	
		AB	13.0 ± 0.7b	
	FS	AS	15.2 ± 0.4a	
		AB	12.4 ± 0.5b	
	FV	AS	15.8 ± 0.8a	
		AB	12.6 ± 0.5b	
	FE	AS	15.4 ± 1.1a	
		AB	12.6 ± 0.5b	
	21	FC	AS	24.4 ± 0.5a
			AB	13.0 ± 0.7b
		FS	AS	17.2 ± 0.8a
			AB	12.4 ± 0.5b
FV		AS	17.6 ± 0.5a	
		AB	14.6 ± 0.5b	
FE		AS	23.6 ± 0.5a	
		AB	17.0 ± 1.2b	

(Table 5) contd....

dpi*	-	Disease Incidence (% of control)/Root Application (1.5 g/kg)		Disease Incidence (% of control)/Foliar Application (1.7 ppm)	
28	FC	AS	26.4 ± 0.9a	AS	26.2 ± 0.4a
		AB	21.8 ± 0.4b	AB	21.6 ± 1.1b
	FS	AS	21.4 ± 0.5a	AS	19.6 ± 0.5a
		AB	17.4 ± 0.5b	AB	17.4 ± 0.5b
	FV	AS	20.6 ± 0.5a	AS	20.0 ± 0.7a
		AB	17.2 ± 0.4b	AB	18.2 ± 0.4b
	FE	AS	26.0 ± 1.0a	AS	25.8 ± 0.8a
		AB	21.2 ± 1.3b	AB	20.6 ± 0.9b
		Disease Severity (% of control)/Root Application (1.5 g/kg)		Disease Severity (% of control)/Foliar application (1.7 ppm)	
14	FC	AS	11.8 ± 0.4a	AS	12.0 ± 0.7a
		AB	11.0 ± 0.7a	AB	11.2 ± 0.4a
	FS	AS	12.2 ± 0.4a	AS	12.4 ± 0.5a
		AB	10.6 ± 0.5b	AB	10.8 ± 0.4b
	FV	AS	12.0 ± 0.7a	AS	12.6 ± 0.5a
		AB	11.6 ± 0.5a	AB	11.2 ± 0.4a
	FE	AS	12.0 ± 0.7a	AS	12.0 ± 0.7a
		AB	10.6 ± 0.5b	AB	10.6 ± 0.5b
21	FC	AS	21.8 ± 0.8a	AS	20.7 ± 0.8a
		AB	16.6 ± 0.5b	AB	16.8 ± 0.8b
	FS	AS	14.8 ± 0.4a	AS	14.8 ± 0.8a
		AB	14.4 ± 0.5a	AB	14.2 ± 0.4a
	FV	AS	14.4 ± 0.5a	AS	14.4 ± 0.5a
		AB	14.8 ± 0.8a	AB	14.4 ± 0.5a
	FE	AS	18.4 ± 0.5a	AS	18.0 ± 0.7a
		AB	16.0 ± 0.7b	AB	16.6 ± 0.5a
28	FC	AS	22.8 ± 0.8a	AS	22.8 ± 0.8a
		AB	20.6 ± 0.5b	AB	20.4 ± 0.5b
	FS	AS	18.2 ± 0.8a	AS	17.4 ± 0.5a
		AB	17.4 ± 0.5a	AB	17.4 ± 0.9a
	FV	AS	18.4 ± 0.5a	AS	17.0 ± 0.7a
		AB	17.6 ± 0.9a	AB	17.2 ± 0.4a
	FE	AS	22.4 ± 0.5a	AS	22.6 ± 0.9a
		AB	18.8 ± 0.8b	AB	18.6 ± 0.5b

Abbreviations: *dpi: days post-inoculation, FC: *F. culmorum*, FS: *F. solani*, FV: *F. verticillioides*, FE: *F. equiseti*, AS: Arabi Aswad, AB: Arabi Abiad, Values are means ± standard deviation of five replicates. Values for the same period of inoculation and same FHB species between two barley cultivars with respect to root and foliar application in the same column with the same letter are not significantly different based on Fisher's test at $p < 0.05$.

4. DISCUSSION

Despite the urgent need to find alternative methods for genetic resistance for decreasing FHB infections on barley, no single strategy has yielded successful control [7]. There is a paucity of reports concerning the application of silicon as a protective measure for fungal pathogens in barley [17], and it has been proven only in the barley-*Blumeria graminis* pathosystem. In the case of head blight on barley, no study has investigated the potential impact of silicon. From a pathogenic point of view, we have, therefore, attempted to fill this gap by providing novel information on how silicon affects the two common quantitative components on moderately resistant "AS, MR" and susceptible "AB, S" barley, bleaching of spikes was used as an indicator of Type I resistance and bleaching of spikelets was used as a representative of Type II resistance, to four FHB pathogens. AS and AB were selected because they are currently the most important barley cultivars in Syria. AS is adapted to drier areas and is popular in northeast Syria. AB is adapted and primarily planted in the wetter areas in western and northwestern Syria. Both genetically different cultivars are

two-rowed with thin stems and high tillering ability [19 - 22]. For reasons still debated, it appears that silicon is particularly effective against biotrophic fungi [11, 13, 14, 16]. However, the use of silicon in this work has shown novel and important observations that could be exploited to fend off four hemibiotrophic *Fusarium* fungi, i.e., *F. culmorum*, *F. solani*, *F. verticillioides*, and *F. equiseti*. This brings further concrete evidence that silicon inputs could be a valuable tool in integrated management against FHB agents by reducing the disease development on barley.

In this investigation, three proposed mechanisms could elucidate the efficient decrease in the incidence (DI), and severity (DS) of FHB disease conferred by foliar and root silicon applications. Firstly, the hypothesis of fungicidal impact suggests that silicon directly affects FHB pathogens due to the fact that foliar sprays were continued during the period ranging from 7 to 28 dpi in which the disease evaluations were conducted, thus silicon was present on barley heads at the periods of primary infection and pathogen invasion. However, this mechanism was doubted by novel findings showing that

silicon did not hamper mycelial growth of the four FHB species used at any of the tested concentrations (1.67, 3.33, 5.00, or 6.67 mM soluble silicon). Being non-fungicidal, it is in agreement with other results [unpublished data]. The second and third postulated mechanisms are through improved overall mechanical strength and an outer protective layer at infection sites in barley heads and the priming of the plants for enhanced production of antifungal compounds [18]. These hypotheses are established on (1) FHB pathogens penetrate glumes, palea, and rachilla directly by constructing invasive mycelia which extend internally throughout vascular bundles in the spikelet [25, 26], and (2) it is unlikely that FHB species can invade the external surfaces of the floret due to the fact that they have strongly thickened lignified walls [25]. The second mechanism, silicon-enhanced cell wall fortification in bracts of inflorescence (silicon is present in glumes, palea, and rachilla [27] acts as a physical barrier against fungal infestation. However, this hypothesis can explain to a limited extent the lowering of FHB damage because 90% of absorbed silicon is approximately located in the epidermis cells of the barley leaves and their cell walls [28]. The third mechanism, germ tubes which successfully invaded the epidermis in barley heads, were controlled by the production of defense-related enzymes as well as the higher accumulation of antifungal components, reinforced by the existence of soluble silicon in the intracellular spaces inside the cells and in the cell wall, as well as in conducting vessels [11, 13]. Barley is known as a silicon absorber and accumulator plant [15]. This last hypothesis seems to be the best admitted and functional mechanism to reduce FHB bleaching of spikes and spikelets [18]. However, biochemical analyses are needed to explore which kind of antifungal compounds is related to the reduction of FHB symptoms on barley plants fed with silicon.

The two components quantified in the present work were negatively affected by silicon. The finding that there was a decrease in bleaching of spikes and spikelets is of great pathogenic value for two reasons. Firstly, the possible mycotoxins released by the four analyzed FHB pathogens, which may have had their ability to efficiently diffuse within the head, decreased. The damage to the cells was avoided due to the silicon deposition in the epidermis cells of the barley leaves, and their cell walls may stimulate the host to produce defense-related enzymes and antifungal compounds. It is known that the damage caused by mycotoxins released by the four tested *Fusarium* species causing head blight is due to the loss of chloroplast pigment and browning of the barley head [25]. Secondly, bleaching of heads by FHB species may be a consequence of synthesized DON or the clogging of phloem and xylem by the pathogens [25, 29]. In addition, mycotoxins are diffusible, *i.e.*, they move into barley tissues that are not colonized by FHB species [30]. The clogging of phloem and xylem vessels perturbs the movement of nutrients and water to uncolonized tissues, resulting in the premature death of spikelets [25]. The decrease in the bleaching of spikes and spikelets in silicon-applied plants indirectly suggests that although the fungi still gain full access to plant tissue, host colonization, diffusion of DON, and clogging of phloem and xylem vessels can be affected by the action of certain mechanisms of resistance.

During the initial infection stage, occurring up to 7 dpi, FHB fungi infection is established by forming several infection features, such as the foot structure, infection cushion, infection

hyphae, lobate appressorium, papillae silica cell, and runner hyphae or necrotic lesions surrounding them [31]. This initial infection stage is associated with other virulent factors, apart from trichothecene synthesis, such as secretion of hydrolytic enzymes [31]. Taking into account that no effects of silicon were observed on FHB incidence (DI) and severity (DS) during the initial infection stage, it seems that potential silicon mediated responses need more time to stimulate as observed during the infection of the heads after day 14 until 28 or virulent factors associated with this earlier stage do not stimulate plant defense mediated with silicon. Our results suppose that silicon triggers defense processes in barley plants, acting as an elicitor, in the latest infection stages to diminish DI and DS with a diversity depending on FHB species by affecting trichothecene synthesis and other virulent factors in fungi [31]. Therefore, the potential enhancement in the activity of these antifungal elements indicates the synthesis of plant defense compounds against external agents. It is widely accepted that plant's defense system is activated by the previous infestation of fungi, silicon fertilization, or both [11, 13, 14, 16]. In contrast to our findings, Yobo *et al.* reported that mycotoxins production impaired protein synthesis in heads, and this would have suppressed the ability of wheat plants to resist progressive infection via delaying plant disease responses in the latest infection stages rated at 22 and 28 dpi [18]. It is important to measure toxin characteristics directly, and their variations to the four tested *Fusarium* species in the presence and absence of silicon.

There is a direct link between the ability of a plant to absorb silicon and the benefits derived from it [15]. During our research, the content of silicon in leaf tissue seemed to be quite appropriate based on the innate physiological capability of this plant species to absorb this element from the soil solution and the potential uptake by leaves to negatively influence FHB fungi growth. As barley control plants were not fed with silicon, it can be suggested that variations in silicon content delivered to barley plants via root and foliar applications accounted for differences in the level of disease response observed in this study. At 28 dpi, solid and foliar treatments reduced the bleaching of spikes (DI) by 26.6% and 22.9%, respectively, on "MR" barley and by 19.4% and 19.5%, respectively, on "S" barley and decreased the bleaching of spikelets (DS) by 20.4% and 19.5%, respectively, on "MR" plants and by 18.8% and 18.4%, respectively, on "S" plants. Our results agree with those found by Yobo *et al.* [18]; they showed that a reduction of FHB DS in the presence of potassium silicate was found. The differences in silicon rates provided by Yobo *et al.* to wheat and applied herein to barley may be attributable to the contrasting isolates and host cultivars used in this study and previous work [18]. Our data revealed that "MR" and "S" barley plants treated with the highest (3.00 g and 3.4 ppm) and the lowest (0.5 g and 0.8 ppm) tested rates delivered through root and foliar applications performed similarly to the control plants. The lack of significantly lower silicon treatments may relate to insufficient accumulation of silicon. Our data have been substantiated by Dogramaci *et al.* through their study on Chili thrips *Scirtothrips dorsalis* populations on pepper plants [32]. Regarding higher silicon doses, the level of soluble silicon inside the plant probably exceeded the critical concentrations at which silicic acid polymerizes; here, the polymerized form of silicon is no longer physiologically active [33]. The content of silicon in leaf

tissues in barley plants treated with solid formulation and foliar sprays will be investigated in further studies that will aim to explore the role of silicon in reducing FHB symptoms. However, previous studies have found that the increased use of silicon enhanced host resistance to fungal diseases [34].

It is mainly accepted that silicon-mediated resistances were stimulated when plants were grown in silicon-enriched soil [11, 13]. More importantly, our data confirmed that barley plants treated with a solid source of silicon (1.50 g) suffered similarly lower levels of FHB disease as compared with those treated with foliar spray (1.7 ppm). Our findings are somewhat in accordance with Yobo's *et al.*'s study, which reported that granulated potassium silicate gave slightly better control of *F. graminearum* than the liquid one [18]. Thus, this observation is of great importance because of the potentially considerable quantities of silicon accumulated inside leaf tissues from foliar applications. However, no report has ever affirmed the efficient absorption of silicon in plant tissues following foliar sprays [35]. For the beneficial effects to manifest, it is commonly accepted that silicon should be absorbed by plant roots in the form of silicic acid, along with water through which it follows the transpiration stream to finally polymerize into insoluble silica, known as species-specific solid bodies (phytoliths) silica [15]. There is a significant body of literature showing that decrease of disease damage through foliar silicon treatment is the result of a direct effect on the pathogen rather than one alleviated by the plant. This was reviewed in Wang *et al.* [13] and Sakr [16].

The silicon effect seemed to be species-specific at 21 and 28 dpi. The four FHB species utilized in the current work are known as capable of mycotoxin production [6]. Thus, the ability of silicon to reduce bleaching of spikes and spikelets in varying amounts on barley proposes that *Fusarium* species differ in their abilities to resist potential silicon mediated responses. In addition, it seems that silicon accumulates much more in barley plants infected with *F. culmorum* followed by *F. equiseti*, and the least with *F. solani* and *F. verticillioides*. Such pathogen specificity has also been shown for certain fungal and viral infections [36, 37].

The present work showed that silicon significantly decreased DI and DS on both "MR" and "S" barley plants. Our results propose that silicon participates actively in enhancing the basal resistance of AS and AB to *Fusarium* species infection. However, the effects of the higher resistance in AS were pronounced under controlled conditions, significantly reducing DI and DS compared to AB, regardless of FHB species and silicon applications at 14, 21, and 28 dpi. We would point out that, generally, DI and DS were lower in AS plants treated with silicon than AB plants treated with silicon. Our findings are in accordance with those reported by Pazdiora *et al.* [38], who studied the same effect for wheat cultivars infected with *Pyrenophora tritici-repentis*, the causative agent of the tan spot. The greatest control of tan spot was obtained with the moderately resistant cultivar grown in soil amended with calcium silicate [38]. Intrinsic differences were observed between the two tested cultivars regarding Type I resistance; moderately resistant plants responded to silicon applications better than susceptible ones, irrespective of FHB species and silicon treatments at 14, 21, and 28 dpi. It seems that head infection is more applicable to distinguish specific responses between barley plants with different degrees of quantitative

resistance than spikelet infection determined for Type II resistance. Field experiments are in progress in our station to confirm the efficacy of soluble silicon in reducing FHB disease on AS and AB.

CONCLUSION

To summarize, methods used to protect barley plants against FHB infection focus principally on using genetic quantitative resistance. In the current work, the use of silicon inputs has highlighted novel and important phenomena that might be exploited in efforts to fend off FHB species. Our data provide the first evidence that root and foliar application of silicon can decrease both FHB DI and DS with a diversity depending of FHB species on "MR" and "S" barley cultivars. It is noteworthy that there exists an interesting observation within barley plants in terms of silicon absorption conferred by foliar spray. Whether this absorption is attributed to the presence of silicon transporters remains to be elucidated. The application of silicon would be an ideal environment-friendly policy and a valuable choice that may be used in an integrated disease management strategy, especially when completely FHB resistant barley cultivars are not available. Enhanced resistance occurred in barley plants with different levels of resistance to FHB infection.

ETHICS APPROVAL AND CONSENT TO PARTICIPATE

Not applicable.

HUMAN AND ANIMAL RIGHTS

Not applicable..

CONSENT FOR PUBLICATION

Not applicable.

AVAILABILITY OF DATA AND MATERIALS

Not applicable.

FUNDING

The author would like to thank the Atomic Energy Commission of Syria for its financial support.

CONFLICT OF INTEREST

The author declares no conflict of interest, financial or otherwise.

ACKNOWLEDGEMENTS

The author would like to thank the Atomic Energy Commission of Syria for providing assistance for this research. The unknown Reviewer is also thanked for constructive comments on this manuscript.

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