Integrating Onion in Crop Rotation to Control Sclerotium rolfsii

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ABSTRACT

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Including onions in crop rotation reduced damage in crops susceptible to *Sclerotium rolfsii*. In peanuts grown after onions, disease incidence was reduced 62% and pod yield was increased 15–52% in field experiments. Onion cultivars showed distinct differences in their ability to reduce the incidence of *S. rolfsii* infection. Disease suppression detected 4 mo after planting onions in infested soil lasted for a year. Sclerotia buried in an onion field were 42% less viable than the control. Onion bulb extract or root exudates inhibited both sclerotial germination and mycelial growth. The inhibitory compound, with a molecular weight of less than 5,000, was heat-sensitive. Integrating onions in crop rotation in *S. rolfsii*-infested fields is potentially an inexpensive means of disease control.

Sclerotium rolfsii Sacc. is one of the major pathogens of peanuts, tomatoes, and other field crops. Disease buildup in monoculture of susceptible crops is very common because of massive production of sclerotia by the pathogen (2). Fumigation with eradicants, e.g., methyl bromide, and soil solarization (7,8) are the most effective control measures for S. rolfsii but are too expensive (5,8). Resistant crops were integrated in crop rotation to reduce incidence of S. rolfsii infection (6,16). Disease incidence was reduced by 3-4 yr of rotation of wheat and corn (4).

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Members of the Lilaceae produce antimicrobial compounds. Garlic oil inhibited growth and sclerotium production in *Rhizoctonia solani* Kühn (13) and growth and spore production in 10 other fungi (9). Agrawal (1) reported the in vitro inhibitory effect of onion root and bulb extracts on the growth of several rhizosphere fungi. Parkinson and Clarke (12) showed that the microflora levels in rhizospheres of onion and garlic were significantly lower than those of other plants.

We report the introduction of onion in crop rotation to reduce damage caused by S. rolfsii to crops in commercial fields and the in vitro effect of onion exudates on the fungus.

MATERIALS AND METHODS

Field experiments and survey. Two field experiments were conducted in

naturally infested soils in central Israel, where peanuts and tomatoes have been grown frequently for the last two decades. Field experiments were made in five replicates of each treatment in a randomized block design, each plot consisting of five beds in an area of 7×20 m. The field plots, located in Kfar-Kassem (experiment 1) and Kalansua (experiment 2), consisted of in alluvial vertisol soil (30% sand, 17.5% silt, 52% clay, and 0.5% organic matter; pH 7.95). Onion seeds (Allium cepa L. 'Grano') (Hazera Seed Co., Haifa, Israel), planted on 20–22 November 1981 in the treated plots, were harvested at the end of May 1982. The control plots were not cropped to onions. Fields were cultivated along the previous rows to prevent soil movement between the plots, and beds were prepared on the original locations. Peanuts (Arachis hypogaea L. 'Shulamit') were sown on 7-11 June 1982. Plants were irrigated and treated with pesticides according to the standard practice in this region. Peanut plants completely destroved by S. rolfsii were removed and their numbers recorded during the season. At harvesttime, 140 days after planting, plants along 12 m from the three central rows of each plot were uprooted and examined for root and pod diseases.

Presence of S. rolfsii in diseased plants was verified by isolating the pathogen. Pods were dried in open air, and the yield of each plot was determined after 2 wk. In experiment 1, 150 beans were sown between two peanut rows at the centers of the beds. Disease incidence was recorded

after 45 days. All bean plants were uprooted on the 45th day.

Greenhouse experiments. The potential for general S. rolfsii populations in soil to induce disease was estimated using beans (Phaseolus vulgaris L. 'Brittle Wax') as a test plant. Six polypropylene boxes ($12 \times 9 \times 6$ cm), each containing 0.5 kg of soil, were planted with 10 bean seeds. Soil from both field and greenhouse experiments was assayed. Boxes were maintained in the greenhouse at 24-30 C for 21 days. Diseased plants were recorded until the end of the experiments.

The ability of four onion cultivars (Grano, Early Grano, Ben-Shemen, and Ori) (Hazera Seed Co., Haifa, Israel) to reduce $S.\ rolfsii$ disease in the soil was tested in naturally infested soil. Onion plants grown in 12 polypropylene boxes $(11\times20\times20\,\mathrm{cm})$, each containing 3 kg of soil, were uprooted 2, 4, and 6 mo after planting. $S.\ rolfsii$ disease incidence was estimated as described.

Thirty S. rolfsii sclerotia that had developed on synthetic medium (SM) (10) were mixed with 5 g of field soil and placed in nylon net bags, which were buried 15 cm deep in the onion field plots. The contents of these bags were sampled

during the growing season. Sclerotia were aseptically placed on SM agar plates prepared according to Elad et al (5) and their viability recorded.

Experiments with onion extracts. Onion bulb extract was prepared by mashing onions (cultivar Grano) in a food processor and sieving them through four layers of cheesecloth, then through Whatman No. 1 filter paper. The extract was sterilized by filtering through a 0.45- μ m Millipore filter at about 25 C.

Onion root exudates were prepared by rooting bulbs (2-3 cm in diameter) on top of Erlenmeyer flasks. The developing roots grew into distilled water during the 3 wk of incubation at room temperature. Solutions of onion root exudates were sterilized by filtering through a Millipore filter.

Onion bulb extracts or root exudates at different concentrations were mixed (1.5 ml) with SM to produce 15-ml aliquots of growth medium. Sclerotia of S. rolfsii or mycelial disks from the edges of a 72-hr culture were inoculated into the growth media. Linear growth and sclerotial germination were measured daily during the 5-day incubation period. Mycelium from 3-day-old liquid cultures of S. rolfsii

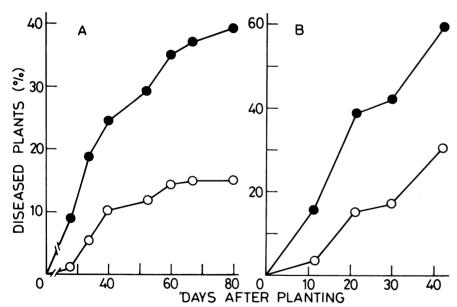


Fig. 1. Effect of onions in field crop rotation on incidence of disease caused by *Sclerotium rolfsii* on (A) peanuts and (B) beans. \bullet = Control and o = onion grown in the previous winter. Means at all sampling dates were statistically different (P = 0.05) according to Duncan's multiple range test.

Table 1. Disease incidence and yields of peanut crops following a crop of onions'

	Sclerotium rolfsii-			
Field location	Onion crop in winter	diseased peanut plants ^z (%)	Pod infestation by S. rolfsii ^z (%)	Pod yield (kg/ha)
Kfar-Kassem	_	45 a	32.3 a	2,530 a
	+	17 Ь	22.7 a	3,850 Ь
Taibe	_	41 a	40.8 a	3,870 a
	+	15 b	21.5 b	4,440 a

⁵ Onion cultivar Grano was planted on 20–22 November 1981 and harvested on 20–22 May 1982. Peanuts were planted on 7–11 June 1982. Disease was recorded on harvest day, 27–31 October 1982.

was oven-dried at 60 C for 6 days to measure dry weight.

Dialysis of onion root exudates was carried out in an ultrafiltration cell (Amicon Co., Denvers, MA) with a YM5 membrane, which lets through components with molecular weights of less than 5,000. The aliquots that passed through this membrane were added to SM for comparison with untreated root exudates.

RESULTS

Field experiments. Results from two experiments made in fields naturally infested with S. rolfsii showed that onions (cultivar Grano) grown during the winter season reduced disease incidence in peanuts the following summer 60-73% during the growing season (Fig. 1A) and 62% on harvest day compared with the control (Table 1). Incidence of diseased pods was reduced 30-47% in both experiments, and peanut yield increased 15-52% (Table 1). When beans were planted in the same plots, disease reduction was also observed in the plots where onions were grown previously (Fig. 1B). Soil samples collected from both field experiments during the onion and peanut growing seasons were planted to beans in the greenhouse. In samples taken from the fifth month of onion growth until the end of the peanut growing season, incidence of disease caused by S. rolfsii was reduced 47-78% compared with samples from control plots. Viability of S. rolfsii sclerotia, buried in the plots during the oniongrowing season, was 40% in the control compared with only 23% in onion plots. Soil samples taken during the peanutgrowing season (June-July 1982) and incubated for a year in the greenhouse at 25-33 C were planted to beans. Infestation by S. rolfsii, tested 21 days later, was 41-67% in control samples and 5-16% in plots preplanted with onions in both field experiments.

Effect of onion cultivar on disease incidence. Four onion cultivars differed in their ability to suppress incidence of *S. rolfsii* in the greenhouse. Onion plants were uprooted 2, 4, and 6 mo after planting and replaced by beans. Soil in which onion plants of the Ben-Shemen or Grano cultivars were grown for 4 mo yielded 100% healthy bean plants compared with 22.5-54.2% diseased bean plants in soils where other onion cultivars had been grown.

Effect of onion extract and root exudates on S. rolfsii. The effect of volatile compounds produced by onions on the linear growth of S. rolfsii was tested in petri plates. Surface-sterilized onion disks were attached to the inside of covers of plates in which S. rolfsii was grown on SM. After 5 days of incubation, linear growth of mycelia was decreased 89.6% in sealed plates but only 50% in nonsealed plates.

When water-soluble exudates of onion roots were collected from rooting onion

⁷ Means of each field experiment followed by the same letter are not statistically different (P = 0.05) according to Duncan's multiple range test.

Table 2. Growth of *Sclerotium rolfsii* in liquid synthetic medium amended with root exudates from two onion cultivars

Percentage of root extract	Dry wt of mycelium (mg/plate) ^y		
in SM ^x	Grano	Ben-Shemen	
100.0	5 b ^z	1 c	
50.0	17 b	49 b	
30.0	23 b	82 ab	
25.0	49 b	95 a	
12.5	57 b	101 a	
0.0	166 a	127 a	

^{*}Synthetic medium was mixed with onion root exudates to give the same nutrient concentration and filtered through a 0.45-μm Millipore filter.

bulbs, sclerotia that were placed on root exudate solution amended with SM constituents did not germinate compared with 73% germination on unamended SM. Liquid SM amended by graduated dilutions of root exudates was also inoculated with mycelial disks of S. rolfsii. Dry weight of S. rolfsii mycelium collected after 3 days of incubation and oven-dried at 60 C was lower in root exudates (Table 2).

Twenty sclerotia mixed with 30 g of natural or autoclaved soils amended with 3 ml of onion bulb extract were separated from the soil after 9 days of incubation and tested for viability on SM. Germination was 96 and 100% in unamended and amended autoclaved soils, respectively, whereas natural soil amended with onion extract resulted in 8% germination of sclerotia compared with 88% germination in unamended soil. Sclerotia from the amended soil were degraded.

Onion bulb extracts totally inhibited growth of Pythium aphanidermatum in SM, whereas growth of Rhizoctonia solani and Trichoderma harzianum was retarded 76 and 52%, respectively. Boiling destroyed the inhibitory effect of onion root exudates as well as that of onion bulb extracts. Freezing active onion solutions did not affect their inhibitory activity. Onion root exudates were dialyzed in an ultrafiltration cell and the solution that passed through the membrane inhibited growth of S. rolfsii. We concluded, therefore, that the molecular weight of the active compound in onion root exudates is less than 5,000.

DISCUSSION

A field survey was conducted in 11 peanut field plots in a heavily infested area to determine severity of disease caused by S. rolfsii. Fields with different crop rotations were compared. Fields planted to onions the preceding winter had only 0–12% diseased plants, whereas other fields not planted to onions had 75% diseased plants. Thus field experiments were designed to evaluate the effect of integrating onion in crop rotation to control S. rolfsii.

Growing onions in soil naturally infested with S. rolfsii reduced the disease incidence, resulting in higher peanut yields than in fields not cropped to onions. The use of crop rotation for controlling S. rolfsii was suggested in several studies using cereals and legumes (6,16). Both greenhouse and field experiments show that the inhibitory effect evident about 5 mo after the onions are planted lasts up to a year.

Sclerotia of S. rolfsii buried in onion plots were less viable than those in the control plots. Incubating sclerotia with onion extract significantly reduced their viability in natural but not in autoclaved soils. This suggests the possible role of soil microflora. According to Parkinson and Clarke (12), the microflora in the rhizosphere of onions and garlic is lower than that of other plants. The possibility, therefore, that S. rolfsii inoculum is predisposed by onion exudates and thus becomes susceptible to antagonistic microflora should not be excluded.

The inhibitory effect of various onion cultivars was observed in the greenhouse. The most effective cultivars were Grano and Ben-Shemen. Moreover, Grano was effective in the field, whereas Early Grano failed to reduce disease incidence.

Pariya and Chakraverty (11) found that onion extract inhibited mycelial growth of S. rolfsii and other fungi. On the other hand, onion oil stimulated germination of S. cepivorum sclerotia (3). Root exudates of onion cultivars Grano and Ben-Shemen inhibited sclerotial germination and mycelial growth of S. rolfsii even after dilution. The active compound, with a molecular weight of less than 5,000, though stable at room temperature, was destroyed by boiling as was shown for garlic extract by Tansey and Appleton (14) and for onion extract by Agrawal (1).

Onion itself is susceptible to S. rolfsii (15); however, during the winter season, temperatures do not favor disease development. Integrating onions in crop

rotation in S. rolfsii-infested fields may be a useful and inexpensive means of reducing disease incidence in susceptible crops such as peanuts, tomatoes, or beans.

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^yMycelia produced in each plate were ovendried at 60 C for 6 days and weighed.

Means in each column followed by a common letter are not statistically different (P = 0.05) according to Duncan's multiple range test.