

Mycelial Compatibility Grouping and Aggressiveness of *Sclerotinia sclerotiorum* on Different Hosts in North of Iran

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ABSTRACT

In this study, genetic structure and pathogenic diversity of *Sclerotinia sclerotiorum*, the causal agent of white cottony stem rot, were assessed through Mycelial Compatibility Groupings (MCGs) and comparison of isolate virulence. 65 isolates from six different hosts from three northern provinces of Iran (Golestan, Mazandaran and Gilan) were placed at 39 different mycelial compatibility groups. The observed MCGs differed within three regions. From all MCGs, 61.53% and 15.38% belonged to Mazandaran and Golestan and 10.25% and 12.82% were common between Mazandaran and Gilan and Mazandaran and Golestan, respectively. Shannon diversity index (Ho) of MCGs for the whole regions was 1.509 (Htot). Partition of total diversity (Htot) showed that 68% corresponded to a variation in diversity within *S. sclerotiorum* populations. Study on virulence was tested using a stem inoculation technique under greenhouse conditions. Evaluating of severity by two methods Stem Lesion Length and the Area Under Disease Progress Curve (AUDPC) showed that isolate virulence varied within the four clade. Moreover, in most cases the differences in virulence of isolates within MCGs were significant. The data indicated that populations of *S. sclerotiorum* obtained from the studied regions were composed of a heterogeneous mix of MCGs, therefore the population structure of this pathogen as well as variations in virulence of isolates must be considered in disease management systems in these regions.

Keywords: MCG, *Sclerotinia sclerotiorum*, Shannon Index and AUDPC.

INTRODUCTION

A pernicious pathogen, *Sclerotinia sclerotiorum* (Lib.) de Bary has a wide geographic distribution and a diverse host range, including many agronomic crops (Hartman, *et al.*, 1999).

In Iran, *Sclerotinia* stem rot is the most destructive and harmful disease of canola, especially in favourable climatic conditions such as those found in the northern

flats of the Caspian Sea (Pakdaman and Mohammadi Goltapeh, 2007). Incidence of this disease in canola fields of Iran has ranged from 12.3% to 54.4% (Barari, *et al.*, 2000)

S. sclerotiorum can spread by sexually and vegetatively produced propagules (Hartman, *et al.*, 1999). Sclerotia, the over-wintering vegetative form of the fungus, reside in the soil and germinate during the cropping season to produce apothecia which release millions of sexually produced, airborne ascospores. Up to 90% of the ascospores, the source of primary inoculum that infects flowers, remain within 100 m of the dispersal site (Ben-Yephet and Bitton, 1985). Additionally, mycelia-infected seed can provide a source of inoculum for continuity of the pathogen's life cycle

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(Hartman, *et al.*, 1998; Mueller, *et al.*, 1999). Although mycelia-infested seed can be dispersed widely, no reports are available to suggest that the thin-walled ascospores are viable after long-distance wind dispersal. Mode and range of pathogen dispersal are important considerations potentially impacting population structure and disease control (Kull *et al.*, 2004).

Intraspecific variation in virulence (Marciano *et al.*, 1983; Morral *et al.*, 1972) and in such morphological characteristics as pigmentation of the mycelium, ascus, ascospore, sclerotial size and production (Boland and Smith, 1991; Le Toumeau, 1979; Price *et al.*, 1975; Purdy, 1955) has been reported in *S. sclerotiorum*. To identify intraspecific variation of *S. sclerotiorum*, two presupposed unrelated criteria of mycelial compatibility groups (MCGs), and DNA fingerprinting have been employed (Kohn *et al.*, 1990; 1991).

Mycelial compatibility/incompatibility is a macroscopic assay of the self-/non-self- recognition system controlled by multiple loci, but knowledge of the underlying genetic mechanisms is limited in most filamentous fungi (Glass and Kaneko, 2003). When hyphae of isolates that differ at one or more of these loci fuse, compartmentalisation and cell lysis occur, leaving a reaction line/barrage zone of reduced growth between the two isolates. Well characterised interaction zones occur between vegetatively incompatible isolates of *S. sclerotiorum* (Kohn *et al.*, 1990). It has been suggested that MCGs represent genetically dissimilar individuals and each MCG is a particular genotype (Kohn *et al.*, 1990). Infrequently outbreeding in *S. sclerotiorum*, immigration of strains from other sites (Glass and Kuldau, 1992), genetic exchange, meiotic recombination (Carbone *et al.*, 1999), mitotic recombination, transitory selection, selective neutrality, and diversifying selection (Kohli *et al.*, 1992) are possible sources of the MCG diversity. MCGs' based grouping has been applied to *S.*

sclerotiorum strains isolated from a variety of hosts in different countries (Atallah *et al.*, 2004; Kull *et al.*, 2004; Sexton *et al.*, 2006; Mert-Turk *et al.*, 2007; Malvarez *et al.*, 2007).

An important consideration that may impact cultivar resistance evaluations and long-term disease control is variability in MCG and isolate aggressiveness and the potential for interactions. "Pathogen aggressiveness" is defined as the relative ability to colonize the host and cause damage, and "virulence" as the relative capacity to produce disease (Agrios, 1999; Shurtleff and Averre, 1997). Greenhouse inoculation studies on canola suggested that some clones are more aggressive in forming lesions (Errampalli and Kohn, 1995; Kimmer, 2003).

Several studies on evaluating soybean resistance to *S. sclerotiorum* showed that results of resistance evaluations from greenhouse and field experiments were of low correlation and also, results from these studies across field locations are mixed (Kim *et al.*, 1999; Nelson *et al.* 1991; Wegulo *et al.* 1998). These may be due in part to disregard for differences in MCG frequency among fields and variation in virulence of *S. sclerotiorum* isolates (Kull *et al.*, 2004).

The objectives of this research were (1) to identify MCGs and evaluate variability in *S. sclerotiorum* populations from different hosts in three provinces of Iran (2) to determine virulence of isolates and their correlation with the identified MCGs.

Materials and Methods

Sclerotinia populations

In total, 260 *S. sclerotiorum* isolates from six different hosts (*Vicia faba*, *Brassica napus*, *Lactuca sativa*, *Lychopersicum esculentum*, *Cucumis sativus* and *Sinapis arvensis*) were collected from 52 fields in three regions in northern Iran that represented the most

important canola-growing areas. These regions included: Guilan, Mazandaran and Golestan Provinces that are mostly located around the Caspian Sea in the north of Iran. Fields were mostly small and their sizes ranged from 0.5 to a maximum of 3 ha. Plants sampled were separated by a minimum distance of 50 steps (25 m), and Five infected plants from each farm were choosed and transferred to the laboratory, representing a total geographical area of about 10000 km². One sclerotium from each plant was surface-sterilised, cultured on potato dextrose agar (PDA; Merck) and incubated at 21 to 24°C in darkness for 2 days. Each isolate was purified by transferring the single hyphal tip onto fresh medium, and generated sclerotia were stored at -20°C until used (Willettts and Wong, 1980; Cubeta *et al.*, 1997; Atallah *et al.*, 2004).

Mycelial Compatibility Grouping

Isolates were manipulated in all possible combinations on PDA, amended with 75 µl L⁻¹ (Schafer and Kohn, 2006) of Wilton's red food coloring per liter of culture medium. (Wilton Crop., USA) according to a previously planned (pyramid design) procedure (Atallah *et al.*, 2004). All pairings were scored after incubation in the dark at room temperature (20-22°C) for 7 and 14 days with each pairing performed triple.

Pairings were scored as mycelium compatible by the fusion of mycelia when no reaction line was observed within the interaction zone between paired isolates.

Pairings were scored as mycelium incompatible by an obvious red line on the bottom side of petri dishes or when a thin to wide band of uniform, aerial mycelium or both was observed between adjacent paired isolates (Kohn *et al.*, 1990). To facilitate MCG determination of all isolates, at first a subset of isolates from each field was selected and paired in all possible combinations and then representatives from each MCG were paired with

representatives of each of the other MCGs (Table 1). In the meantime, self-self pairings were performed as well.

Assessment and Comparison of MCG Diversity

The Shannon-Wiener's index, h_0 , was calculated for each population as follows (Shannon and Wiener, 1949):

$$H_0 = -\sum (p_i \ln p_i)$$

where p_i was the frequency of i th MCG. Frequency was defined as the ratio between the number of isolates belonging to the i th MCG and the number of isolates in the sample. To correct for differences in sample size among populations, MCG diversity values were normalized by the maximum diversity in each populations, so that $H_0 = h_0 / \ln k$. Where k was the sample size. Total MCG diversity (H_{tot}) was partitioned into within, and among population components (Goodwin *et al.*, 1992).

Virulence Assessment

Virulence of isolates was determined using a stem inoculation technique, By using petals inoculated with colony of *S. sclerotiorum* (Ziqin *et al.*, 2008.). Haola 401 rape seed, was used in all virulence tests. Blossom colonization was achieved by placing autoclaved blossoms on top of a growing colony of *S. sclerotiorum* on PDA medium for 3 to 4 days.

Each colonized blossom was lightly wrapped with gauze to hold the flower on the (is there any wounds or any scratch on stem surface/????) stem and to preserve moisture. Plants were subjected to an intermittent mist for 24 hours. Misting was stopped after 24 hours but the plants were kept at above 95% relative humidity for 48 hours. Ambient temperature was $16 \pm 2^\circ\text{C}$ at night and $27 \pm 2^\circ\text{C}$ during the day. On the third day post-inoculation, plants were transported to the greenhouse and kept at approximately 25°C during the day and 18°C at night for 3 additional days, and lesion lengths were

measured using Vernier calipers. Experimental design for all the isolate virulence tests (65 isolates) was a randomized, complete block at three replications and data was analyzed by one-way ANOVA using PROC GLM in SAS. Isolate was the test and lesion length was the observational unit. The data was not normally distributed and a rank transformation was used for the ANOVA test.

Two separate scoring systems were employed to assess the virulence of each isolate. Stem Lesion Length (SLL; cm) was measured 2, 4, and 6 days after inoculation. The Area Under Disease Progress Curve (AUDPC) was calculated as follows:

$$\text{AUDPC} = \left[\frac{(Y_1 + Y_2)}{2} (t_2 - t_1) \right] + \left[\frac{(Y_2 + Y_3)}{2} (t_3 - t_2) \right]$$

where Y_1 , Y_2 and Y_3 were lesion length 2, 4 and 6 days past inoculation, while t_1 , t_2 , and t_3 represented days of scoring.

Data Analysis

StemSL and AUDPC data were analyzed using Analysis of Variance (ANOVA) and employing MSTATC software. Duncan's Multiple Range Test (DMRT) was employed to determine the significant ($P=0.01$) differences among mean values.

RESULTS

MCG Diversity and Distribution

The MCG analysis of *S. sclerotiorum* populations in this study showed that in a given location there was a heterogeneous mix of MCGs, according to a previously published by barari and co-workers (2008; 2010 a,b;

2011; 2012) 39 mycelial compatibility groups were identified among 65 isolates representing 52 farm, which 66.6% were unique, defined as one MCG that constituted single isolate specimens. All the isolates were compatible within themselves. Pairs that were 100% compatible merged to form one colony with no distinct zone. Three levels of incompatibility were distinguished. Level 1 incompatibility (not completely compatible): when a sharp distinct thin band of mycelia was observed in the interaction zone; Level 2 incompatibility (not completely incompatible): reaction line was visible as abundant tufts, white patches of aerial mycelia in the reaction zone on the colony surface; and Level 3 incompatibility (100% incompatible): when a red reaction line observed between the interfering paired isolates. The observed MCGs differed within the three regions). From all MCGs, 61.53% and 15.39% belonged to Mazandaran and Golestan and 10.26% and 12.82% was common between Mazandaran and Gilan and Mazandaran and Golestan, respectively and There was no common group between Gilan and Golestan (Fig. 1).

Shannon diversity index of MCGs for three regions was found out 1.509 (H_{tot}), ranging from 0.579 for Gilan province to 1.456 for Mazandaran province and with a mean of 1.031 ($H_{\text{avr.}}$) (Table 3.1). Overall, diversity was higher in the population from Mazandaran than in those from Gilan and Golestan. Partition of total diversity showed that 68% corresponded to a variation of diversity within *S. sclerotiorum* populations, while only 32% of diversity was responsible for variability among those populations (Table 2).

Table 1. Investigated *S. sclerotiorum* isolates grouped through MCG, isolate code, and location.

Isolates code	Host	Site ¹	MCG	Isolates	Host	Site ¹	MCGs
R4	Lettuce	Dashtenaz ,M	1	R35	Rapeseed	Shirgah ,M	14
R19	Rapeseed	Amol ,M	1	R36	Rapeseed	Arateh ,M	15
R25	Rapeseed	Rezvanshahr,Gi	1	R38	Rapeseed	Kordekhail-Sari,M	16
R26	Rapeseed	Bandar Anzali,Gi	1	R39	Rapeseed	Shast kalateh,Go	17
R5	Lettuce	Kiakolla,M	2	R40	Rapeseed	Shast Kalateh, Go	18
R27	Rapeseed	Bandar Anzali,Gi	2	R41	Rapeseed	Kordkoi,Go	18
R6	Lettuce	Kiakolla,M	3	R65	Rapeseed	Bayekolla ,M	18
R37	Rapeseed	Nokandeh, Go	3	R42	Rapeseed	Kordkoi ,Go	19
R8	Lettuce	Amol ,M	4	R44	Rapeseed	Kordkoi-zare,Go	20
R13	Broad bean	Juibar,M	4	R45	Rapeseed	Kordkoi-zare,Go	21
R20	Rapeseed	Amol-hular,M	4	R48	Rapeseed	Behshahr ,M	21
R22	Rapeseed	Babol ,M	4	R46	Rapeseed	Behshahr ,M	22
R30	Rapeseed	Ghaemshahr,M	4	R50	Rapeseed	Kordkoi-Kar,Go.	23
R49	Rapeseed	Kordkoy,Go	4	R57	Rapeseed	Hullar ,M	23
R55	Rapeseed	Semeskandeh,M	4	R58	Rapeseed	Hullar ,M	23
R64	Rapeseed	Bayekolla,M	4	R59	Rapeseed	Hullar ,M	23
R9	Lettuce	Amol ,M	5	R51	Rapeseed	Kordkoi-Kar,Go	24
R18	Rapeseed	Amol ,M	5	R52	Rapeseed	Galugah ,M	25
R23	Rapeseed	Chardangeh,M	5	R53	Rapeseed	Suteh ,M	26
R47	Rapeseed	Behshahr,M	5	R54	Rapeseed	Semeskandeh,M	27
R10	Broad bean	Kiakolla ,M	6	R60	Rapeseed	Galugah ,M	28
R34	Rapeseed	Shirgah , M	6	R61	Rapeseed	Dashtenaz ,M	29
R14	Broad bean	Babol , M	7	R62	Rapeseed	Dashtenaz ,M	30
R56	Rapeseed	Semeskandeh, M	7	R63	Rapeseed	Garakhail ,M	31
R17	Tomatto	Juibar,M	8	R15	Wild sinapis	Juibar ,M	32
R24	Rapeseed	Rezvanshahr,Gi	8	R16	Wild sinapis	Kordkoichardeh3,Go	33
R21	Rapeseed	Babol,M	9	R1	Cucumber	Bahnamir,M	34
R28	Rapeseed	Bandar anzali,Gi	10	R2	Cucumber	Bahnamir ,M	35
R33	Rapeseed	Juibar ,M	10	R3	Cucumber	Juibar ,M	36
R29	Rapeseed	Juibar ,M	11	R7	Lettuce	Juibar ,M	37
R31	Rapeseed	Juibar ,M	12	R11	Broad bean	Kiakolla ,M	38
R32	Rapeseed	Juibar ,M	13	R12	Broad bean	Kiakolla ,M	39
R43	Rapeseed	Bandarturkaman,Go	13				

1-M:Mazandaran province; Gi: Gillan province; Go: Golestan province

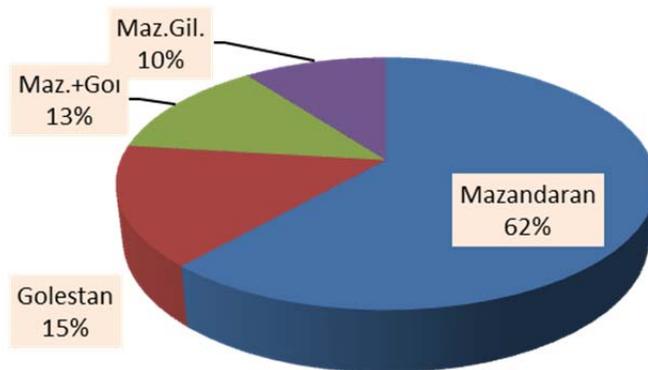


Figure 1. Percentage of total MCGs within the three provinces.

Isolate Virulence within MCGs

Isolate virulence within the three regions analysis of variance (ANOVA) showed that there were significant differences among virulence of isolates (P= 0.01) (Table 3).

In this respect, isolates of Golestan had mean AUDPC values ranging from 0.49 to 56.98. Mazandaran had mean AUDPC ranging from 1.25 to 69.29. Gilan carried mean AUDPC values ranging from 4.7 to 50.02 (Table 2 and 3). The least aggressive isolate (R16) came from Golestan (wild siapis) and responsible for a mean

AUDPC value of 0.49. The most aggressive isolate (R29) came from Mazandaran (rapeseed) and bore the highest mean AUDPC values (Tab. 2 and 3). Four groups of virulences were obtained among all the isolates through UPGMA (Figure 3). Group IV included isolates with low virulence and Group III included isolates with high virulence (Fig.3), that isolates pathogenicity calculated with both methods AUDPC and Regions Analysis of variance matched (Table 2 and Fig. 2).

Table 2. MCG diversity (H₀) of *S. sclerotiorum* populations from different locations.

Location	Sample size	H ₀	H _{avr} /H _{tot}	H _{tot} -H _{avr} /H _{tot}
Gilan	5	0.579		
Mazandaran	47	1.456		
Golestan	13	1.06		
Total	65	1.509	0.68	0.32

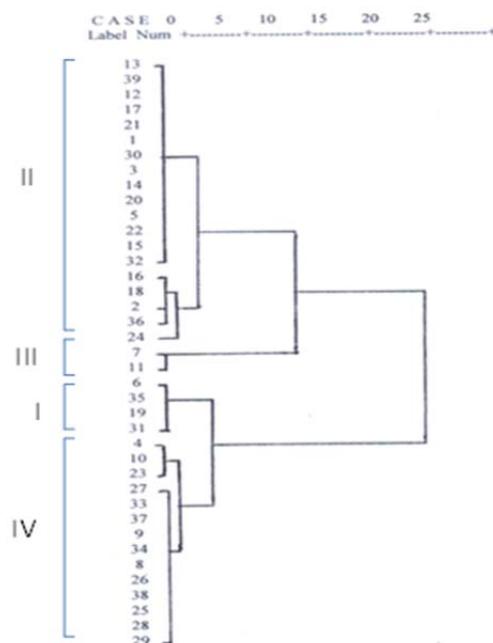


Figure 2. Clustering of 64 isolates of *S. sclerotiorum* as based on AUDPC values.

Table 3. Mean AUDPC values using Duncan's Multiple Range Test (P= 0.01).

MCG	2d after inoculation	4d after inoculation	6d after inoculation	Cluster	AUDPC mean
1	4.11	9.30	14.87	2	37.58 efg
2	5.85	12.25	19.67	2	50.02 cd
3	4.30	9.10	15.00	2	37.5 efg
4	0.95	2.40	6.67	4	10.42 k
5	3.85	8.65	14.00	2	35.15 fg
6	2.05	4.45	7.17	1	18.12 ij
7	7.36	15.75	26.67	3	65.53 ab
8	0.4	1.15	2.00	4	4.7 lm
9	0.45	1.20	2.00	4	4.85 lm
10	1.05	2.35	4.00	4	9.75 k
11	7.66	17.15	27.33	3	69.29 a
12	5.80	10.55	16.67	2	43.57 de
13	6.15	10.35	16.67	2	43.52 de
14	4.65	9.85	15.17	2	39.52 def
15	3.8	8.35	13.33	2	33.63 gh
16	4.70	11.95	20.67	2	49.27 cd
17	5.46	9.85	16.11	2	41.27 def

MCG	2d after inoculation	4d after inoculation	6d after inoculation	Cluster	AUDPC mean
18	5.65	12.33	20.00	2	50.31 cd
19	2.48	5.48	10.17	1	23.61 i
20	4.70	9.66	15.12	2	39.14 def
21	4.67	10.90	18.08	2	44.55 de
22	3.65	8.80	14.17	2	35.42 gh
23	0.8	2.88	6.00	1	12.56 k
24	6.55	12.88	24.67	2	56.98 c
25	0.35	0.8	2.00	4	3.95 lm
26	0.4	1.05	2.00	4	4.5 lm
27	0.12	0.23	0.67	4	1.25 mo
28	0.65	1.10	3.67	4	6.52 l
29	0.6	0.95	3.00	4	5.5 l
30	3.65	8.15	16.00	2	35.95 fg
31	2.45	4.75	9.00	1	20.95 i
32	3.33	7.80	13.00	2	31.93 gh
33	0.00	0.08	0.33	4	0.49 o
34	0.4	1.20	2.00	4	4.8 lm
35	1.85	3.66	7.33	1	16.5 ij
36	5.75	12.05	19.33	2	49.18 cd
37	0.25	0.45	1.33	4	2.48 lmo
38	0.6	1.15	2.33	4	5.23 l
39	5.65	10.35	16.67	2	43.02 de

DISCUSSION

Belonging to isolates obtained from different hosts in one mycelial compatibility group shows that *S. sclerotiorum* has a wide host range. For example, Broad bean (from Mazandaran province), Rapeseed (from Mazandaran and Golestan provinces) and Lettuce (from Mazandaran province) are placed in group four of MCG. In this study it was found, mycelial compatibility groups common shared between Gillan and west of Mazandaran isolates and also between east of Mazandaran and Golestan isolates. This may show movement of propagules or indicate selection for particular MCG genotypes affected by environmental conditions and competition (Kull *et al.*, 2004)., But isolates

from geographically separated provinces (Golestan and Gilan) were incompatible. This could be a result of geographic distance and failure of sexual recombination. The climatic conditions of Gilan province, is humid with high rainfall and Golestan province is semi-arid climate and low rainfall, that can be an emphasis on the up assumption or the low number of samples to be analyzed, that additional sampling from these regions is required.

A distribution of mycelial compatibility grouping in *S. sclerotiorum* isolates from Iran was for the first time reported in this study. Populations of *S. sclerotiorum* from the studied locations formed a heterogeneous mix of MCGs. This is in agreement with those obtained from

canola and soybean crops in Canada (Kohli *et al.*, 1992; 1995; Kohn *et al.*, 1991; Hambleton *et al.* 2002), cabbage and soybean in USA (Cubeta *et al.*, 1997; Kull *et al.*, 2004).

Furthermore, the results indicated a high level of MCG diversity. Genetic exchange, meiotic recombination (Carbone *et al.*, 1999), mitotic recombination, transitory selection, selective neutrality, diversifying selection (Kohli *et al.*, 1992), infrequent outbreeding in *S. sclerotiorum*, immigration of strains from other sites (Glass and Kuldau, 1992), could account for the possibilities of MCG diversity.

The data indicated that 66.6% all of MCGs were unique. Localization of unique MCGs was observed in winter canola in Harrison, Ontario (Kohn *et al.*, 1991). The existence of unique MCGs in a sampling area suggests that new MCGs and so new genotypes are evolving. The emergence of new genotypes could indicate that MCGs could be adapted to specific field microclimates. Also, evolving of new genotypes may be associated with increased cropping of canola in such areas (Hambleton *et al.*, 2002), leading to movement of *S. sclerotiorum* onto this crop from several other host plants, or due to ascogenous system in sexual reproduction, indicating a sexual population in contrast to the clonal population structure as indicated by other studies (Kohli *et al.*, 1992; Cubeta *et al.*, 1997).

AUDPC score was assessed after 2, 4, and 6 days past inoculation. Thus, this approach would distinguish differences in disease progress caused by different isolates.

Different levels of virulence of *S. sclerotiorum* have been previously examined. A study report (Barari, *et al.*, 2012 ; Kull *et al.*, 2004) through use of a limited-term, plug inoculation technique, showed that isolate aggressiveness varied in some studied locations. The present study revealed a considerable range in AUDPC values within MCGs constituted from individuals from a single location or even from a single field. These pathogenic variations in

isolates and in MCGs do not seem to be related with their geographic origins. Moreover, it may indicate that MCG is not a suitable marker for virulence. Additionally, upon comparison, the isolates from different sources, notable variation in cultural morphology and virulence was found among protoplast regenerated isolates of *S. sclerotiorum* (Barari *et al.*, 2010a, 2011, 2012; Boland and Smith, 1991).

Few reports regarding variation in *S. sclerotiorum* aggressiveness on rapeseed are available. A 1975 report (Price and Calhoun, 1975) comparing pathogenicity of 14 *S. sclerotiorum* isolates on 11 different hosts showed a variation in degree of pathogenicity dependent on host species. On the basis of a detached celery petiole assay 50 MCGs identified from 160 Argentinean isolates (Durman *et al.*, 2003) were found not to differ in aggressiveness among MCGs. By using a limited-term, plug inoculation technique, Kull *et al.* (2004) reported that aggressiveness varied between isolates and MCGs from different locations, but not in MCGs produced from isolates originating from infections in single fields.

This genetic association of low aggressiveness and high pervasiveness supports the popular idea held by evolutionary biologists that pathogens can evolve to become harmless, more deadly, or anything in between depending on the forces guiding natural selection; such forces can pull the pathogen in opposite directions at the same time, creating an evolutionary tradeoff between fecundity and infectivity factors (Kohn, 1995).

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مجاميع التوافق الميسليومي والأمراضية لفطر *Sclerotinia sclerotiorum* على عدة عوائل في شمال إيران

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ملخص

تمت دراسة التركيب الوراثي والقدرة الامراضية لفطر *Sclerotinia sclerotiorum* المسبب لمرض تعفن الساق القطني الأبيض من خلال اعداد مجاميع التوافق الميسليومي ومقارنة الأمراضية للعزلات الفطرية. تم وضع خمسة وستين عزلة جمعت من ستة عوائل من ثلاث محافظات في شمالي ايران (محافظة جولستان، مازنداران و جيلان) في 39 مجموعة ميسليومية توافقية. كانت هناك فروق واضحة بين المحافظات الثلاث، بحيث كان %61.53 و %15.38 من التوافق للعزلات من مازنداران و جولستان، و 10.25 و 12.82 مشترك مابين مازنداران و جيلان من جهة و مازنداران و جولستان من جهة اخرى على التوالي. أظهرت العزلات جميعها 1.509 نقطة على مقياس شانون للتنوع (Htot). كان %68 من التنوع الكلي يعود الى التنوع في مجاميع الفطر *Sclerotinia sclerotiorum*. درست القدرة الأمراضية بطريقة عدوى الساق تحت ظروف البيوت الزجاجية. قدرت الامراضية بقياس طول بقعة الساق والمساحة تحت منحنى تطور المرض وظهرت فروق معنوية بين العزلات في معظم المقارنات. اظهرت النتائج ان العزلات الفطرية اشتملت على تنوع وراثي في المجاميع الميسليومية التوافقية، مما يتوجب الأخذ بعين الاعتبار تنوع القدرة الامراضية، وشكل هذه المجاميع عند عمل برامج مكافحة المرض في المحافظات المختلفة.

الكلمات الدالة: *Sclerotinia sclerotiorum*، مقياس شانون، المساحة تحت منحنى تطور المرض (AUDPC).

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