
Pathogenic Variability of *Ascochyta rabiei* in Saskatchewan

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Abstract

Forty isolates of *Ascochyta rabiei* collected in Saskatchewan Canada and 18 obtained from other countries were assessed for pathogenicity on eight chickpea differential lines/cultivars under controlled conditions. Each assessment was repeated once. Based on the differential by isolate interactions, 15 distinct pathotypes were identified among the isolates from Saskatchewan. This demonstrates that many races of *A. rabiei* are present in Saskatchewan. Formation of new virulent races through gene recombination is possible. This will have large impact on ascochyta blight development and its control. Plant breeders should anticipate a highly diverse *A. rabiei* population with a high potential for change.

Introduction

Ascochyta rabiei, the cause of ascochyta blight, is a major limiting factor on chickpea (*Cicer arietinum*) production worldwide. Large variations in virulence and the existence of races of *A. rabiei*, have been reported in several countries (Vir and Grewal 1974; Qureshi et al. 1984; Reddy and kabbabeh 1985; Jan and Wiese 1991; Porta-Puglia et al. 1996). The sexual stage which also occurs in Canada (Armstrong et al. 20001), produces wind-borne ascospores which contributes to long-distance dispersal of the pathogen and also helps to create genetically diverse populations through gene recombination. Recent severe blight epidemics in Saskatchewan (Chongo et al. 2000), suggest that ascospores are a major source of inoculum. The objective was to assess pathogenic variability and the existence of races of *A. rabiei* in Canada and to compare them to foreign isolates.

Materials and methods

Several chickpea accessions/breeding lines/cultivars, which included both kabuli and desi types, were assessed to select those that best differentiated amongst *A. rabiei* isolates. Most of the lines were selected from the literature. Twenty-nine kabuli lines were obtained from the International Centre for Agricultural Research in the Dry Areas (ICARDA) in Syria, 20 desi lines from the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) in India, and 11 breeding lines and/or cultivars from the Crop Development Centre, University of Saskatchewan, Canada. Two locally-produced commercial kabuli cultivars; UC27 and Sanford, were included as susceptible and resistant checks, respectively. In total, 62 chickpea lines were inoculated with three aggressive isolates, 14 lines were selected and used to screen a population of 40 Canadian isolates and 18 isolates from other countries. Pots were seeded with three seeds per pot, in a soil-less mix of peat moss, vermiculite and sand (2:2:1 v/v/v) plus slow-release granular fertilizer

(Osmocote, Scotts-Sierra Horticultural Products Co., Marysville, OH). The pots were placed in a greenhouse for 3 weeks at about 20:16°C (day:night) temperature and 16 h photoperiod.

Thirty-nine of the Canadian isolates were collected in Saskatchewan from 1997 to 2000 from 25 different ascochyta blight-infected chickpea fields with typical symptoms (Fig. 1). One isolate (Jan0001) was obtained by crossing isolates May9804 and Jan9702. The group of foreign isolates consisted of five isolates from Australia, three from the USA, six from Syria, two from India and two from Turkey. Seedlings were inoculated with a suspension of 2×10^5 conidia/ml of each single spore isolate, and the inoculated seedlings were incubated in a misting chamber in a growth chamber for 48 h. The chamber was maintained at 20:16°C (day:night) temperature and 16 h photoperiod with $250 \mu\text{E m}^{-2} \text{s}^{-1}$. Plants were rated using a 0-9 scale 14 days after inoculation, and categorized in two infection phenotypes; 0-3 = resistant and 4-9 = susceptible for determining pathotypes.

The disease reactions of the differentials were assessed using analysis of variance (SAS Institute, Carry, NC). Cluster analysis was used to examine the relationship(s) between the Canadian and foreign isolates (NTSYS-pc program, Exeter Software, USA). The isolate by differential data were clustered using the unweighted pair group method arithmetic averages with average distance.

Results and discussion

During the initial screening of 62 chickpea lines/cultivars to select a differential set, several factors were considered. First, in a preliminary assessment of all the chickpea lines with three isolates, only lines that produced consistent differential reactions were selected. Second, when several lines came from similar parental source material, only the one with the most consistent pattern of differential reaction was selected. Third, limited amounts of seed from ICARDA and ICRISAT, required seed multiplication under controlled conditions. Lines that readily set seed with good germination were preferred. In total, 14 lines were selected to screen the *A. rabiei* isolates, but only eight differential lines (Table 1) were included in the final data analyses, based on consistence in results between the two repetitions of the inoculation trial.

In the analysis of variance, the significant differential by isolate interaction for both the Canadian isolates only and in the combined analysis of both Canadian and foreign isolates demonstrated that the disease reaction of some isolates was specific to certain differentials. Based on these differential reactions, 15 pathotypes were identified in the Canadian population (Table 1). These ranged from those that were only weakly pathogenic even on the susceptible cv. UC27, to those that were virulent on as many as 6 of 8 lines. Pathotype 2, was the most common and was represented by 20% of the population of isolates from Canada. The next most frequent pathotypes were 1 and 4 which represented 10 and 13% of the isolates, respectively. Substantial pathogenic variation was observed even within single fields. At least two to four different pathotypes were present in six of seven fields that contained multiple samples of isolates.

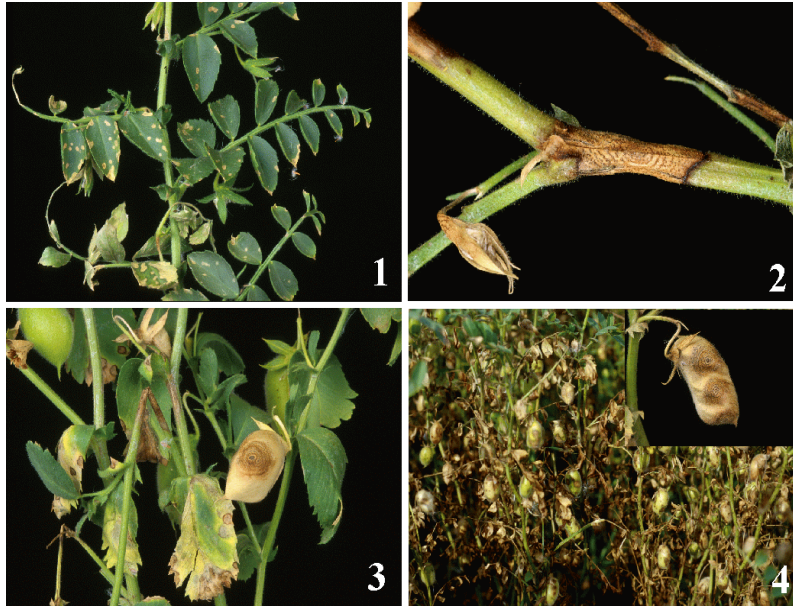


Figure 1. Ascochyta blight symptoms: 1 = early and 2-4 = advanced.

Seedlings of cv. Sanford were susceptible to pathotypes 11, 12, 14 and 15 (Table 1), which represented 33% of the Canadian isolates. Sanford is one of the very few chickpea cultivars that are adapted to the short growing season of the Canadian prairies and also carries a useful level of resistance to ascochyta blight. As a result, it is currently one of the most widely-grown cultivar in this region. These results suggest that its useful life as a resistant cultivar in the region may be quite short. No isolates were pathogenic on line ILC 4421, 5% were pathogenic on Flip 48-83 and ILC 3856, 20% on ICC 6328, 50% on ICC 4200 and 55% on ICC 4475. In general, the desi lines were susceptible to more isolates than the four kabuli's. Other studies have also reported better resistance in kabuli than desi types (Reddy and Singh 1984; Reddy et al. 1992). Although line ILC 4421 was resistant to all isolates, it has also been reported to be susceptible in certain locations (Singh and Reddy 1990; Reddy et al. 1992). This difference in disease reaction among sites is almost certainly due to differences in virulence in local pathogen populations. Lines ILC 4421 and Flip 83-48 represent potential sources of resistance for use in western Canada, but their reaction needs to be assessed against a larger population of isolates, and in the field before a major effort is made to incorporate this resistance into adapted cultivars.

Table 1. Disease reaction[†] of eight chickpea differential lines/cultivars to 40 isolates of *Ascochyta rabiei* from Saskatchewan, Canada.

| Pathotype [‡] | Isolate | Host differential | | | | | | | | |
|------------------------|---------|-------------------|---------|---------|---------|---------|---------|------------|---------|---|
| | | UC27 | ICC4200 | ICC4475 | ICC6328 | Sanford | ILC3856 | FLIP 83-48 | ILC4421 | |
| 1 | Jul9815 | MR | R | R | R | R | R | R | R | |
| | Jul9816 | MR | MR | MR | MR | R | R | R | R | |
| | Nov9803 | MR | MR | MR | R | R | R | R | R | |
| | Nov9806 | R | MR | R | R | R | R | R | R | |
| 2 | Oct9808 | HS | MR | MR | MR | MR | R | R | MR | |
| | Nov9805 | HS | MR | MR | MR | MR | MR | MR | R | |
| | Dec9804 | HS | R | MR | R | R | R | R | R | |
| | Feb9914 | HS | MR | MR | R | MR | R | R | R | |
| | Jan9907 | HS | R | R | R | R | R | R | R | |
| | Mar9703 | HS | MR | R | MR | R | R | R | R | |
| | Nov9807 | HS | MR | MR | MR | MR | R | R | R | |
| | Sep9806 | HS | MR | MR | MR | MR | R | R | R | |
| | 3 | Oct9805 | HS | S | MR | MR | R | MR | R | R |
| | | Feb9803 | HS | MS | MR | R | MR | MR | R | R |
| Jan2001 | | HS | MS | MR | MR | MR | MR | R | R | |
| 4 | Aug9803 | HS | S | S | MR | MR | MR | MR | MR | |
| | Jan9702 | HS | MS | MS | MR | MR | MR | MR | R | |
| | Jul9812 | HS | MS | S | MR | MR | MR | MR | MR | |
| | Jul9806 | HS | S | MS | MR | R | R | R | R | |
| | Jul9807 | HS | S | MS | MR | MR | R | MR | MR | |
| 5 | Jul9808 | HS | MS | MS | MS | MR | MR | R | MR | |
| | Jul9810 | HS | MS | S | MS | MR | MR | R | MR | |
| | May9804 | HS | MS | MS | MS | MR | MR | MR | R | |
| 6 | Jan9805 | HS | MS | S | MS | MS | MR | R | MR | |
| | Jul9811 | HS | MS | MS | MS | MS | MR | MR | MR | |
| | Sep9804 | HS | S | S | S | MS | MR | MR | MR | |
| 7 | Jul9809 | HS | MS | S | MS | S | MS | MR | MR | |
| 8 | Oct9806 | HS | MR | MS | R | MR | R | MR | MR | |
| | Jan9804 | HS | MR | MS | R | MR | R | R | R | |
| 9 | Oct9809 | HS | MR | MR | MR | MS | MR | MR | R | |
| 10 | Jul9803 | S | MR | R | R | R | S | R | MR | |
| 11 | Oct9804 | HS | MS | R | MR | MS | MR | MR | R | |
| 12 | Jul9813 | HS | MR | S | MR | S | MR | MR | MR | |
| | Jul9818 | HS | MR | MS | MR | MS | MR | MR | MR | |
| | Jul9814 | HS | MR | MS | MR | S | MR | MR | R | |
| 13 | Jul9805 | HS | MR | S | MS | MR | MR | MR | R | |
| 14 | Jul9804 | HS | MS | MS | MR | MS | MR | R | R | |
| | Feb9710 | HS | MS | MS | MR | MS | R | MR | MR | |
| 15 | Feb9923 | HS | MS | MS | MR | MS | R | MS | R | |
| | Nov9808 | HS | MS | MS | R | MS | MR | MS | R | |

[†] Based on a 0-9 scale where, 0,1 = resistant (R), 2,3 = moderately resistant (MR), 4,5 = moderately susceptible (MS), 6,7 = susceptible (S), and 8,9 = highly susceptible (HS). Pathotypes: 0-3 = R and 4-9 = S.

[‡] The foreign isolates were represented by seven pathotypes including pathotypes 1 (7 isolates), 2 (3), 3 (1), 5 (1), 9 (3), 11 (1) and 14 (1 isolate). One isolate from Australia was not represented by any pathotype.

In cluster analysis, isolates in one major cluster were weakly pathogenic or nonpathogenic and contained only pathotype 1 (Fig. 2). In the second major cluster isolates were pathogenic on one

or two differentials and contained isolates from pathotypes 9 and 12. In the third major cluster, which contained the rest of the pathotypes, all of the isolates were pathogenic, although their disease reaction on the differential lines was highly variable, resulting in many sub-clusters. The foreign isolates were pathogenic, so they occurred in two of the three major clusters (Fig.2). Two of the six previously described races from Syria (race 2 and 3) clustered together and were nonpathogenic. Four of the five isolates from Australia were in one sub-cluster, both isolates from Turkey clustered together, and so did two of three isolates from the USA. This suggested some grouping based on locality. However, in each of the sub-clusters containing foreign isolates, there was at least one Canadian isolate.

This study demonstrated that there is a broad range of pathogenic variation in the population of *A. rabiei* in western Canada; 15 races were identified using eight differential chickpea lines. As reported above, several studies have examined the pathogenic variability of *A. rabiei* in various parts of the world. Each study used different chickpea lines as differentials and different rating scales and categories to assess disease reaction. This permits researchers to include locally-adapted resistant cultivars, in order to identify races that are most important for their region. However, the lack of a standard system makes race comparisons among regions very difficult. Since there is no standard race characterization system for chickpea, we decided not to designate races identified in the present study (e.g. by numbers 1 to 15), to avoid confusion with the races that have already been described (Reddy and Kabbabeh 1985).

Only one line was resistant to all 40 Canadian isolates. Faced with a genetically diverse, sexually reproducing pathogen population, plant breeders need to assess the potential for developing cultivars with durable forms of resistance. Also, pathologists need to monitor changes in the race structure of the pathogen population to predict the breakdown of resistance in existing chickpea cultivars.

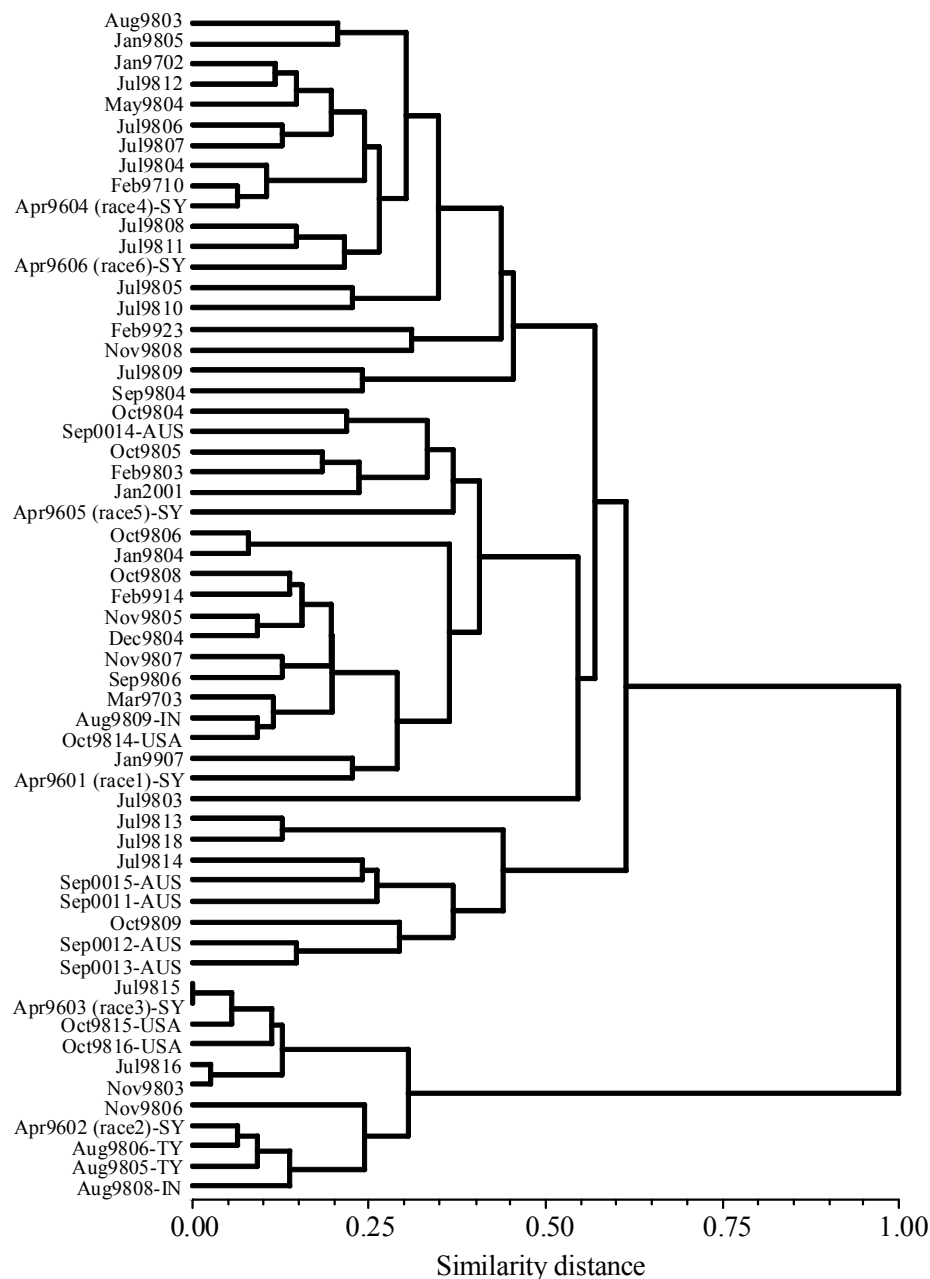


Figure 2. Cluster analysis of *A. rabiei* isolates in 3 major groups from Canada, Australia (AUS), Syria (SY), India (IN) and Turkey (TY).

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