

## MULTIPLE DISEASE RESISTANCE IN LENTIL GERMPLASM FROM PAKISTAN

SHAUKAT HUSSAIN, ASGHAR ALI, M. ASHRAF ZAHID AND S.M. IQBAL

*Pulses Program, National Agricultural Research Centre,  
Islamabad, Pakistan*

### Abstract

Lentil blight, rust and viral diseases, particularly pea seed-borne mosaic virus (PSbMV) are serious threat to lentil production in Pakistan. Multiple disease resistance in lentil is not available in the country. During 1998-99, 590 local lentil germplasm accessions were evaluated under field conditions to identify multiple sources of resistance against major diseases. A wide range of variation to disease reaction was observed among lentil genotypes. Majority of the accessions was susceptible to rust, whereas maximum number of genotypes showed either resistant or tolerant reaction to *Ascochyta* blight and PSbMV. Only one accession (66013-6) was found with high level of resistance to all the three diseases. Twenty-three accessions were found highly resistant to both *Ascochyta* blight and PSbMV. Two lines (66013-3 and 66013-4) were found resistant to blight and rust. The identified sources of resistance can be utilized in lentil breeding program for the development of disease resistant cultivars for commercial cultivation.

### Introduction

Lentil (*Lens culinaris* Medk.), locally known as Masoor, is an important winter season crop of Pakistan. The crop is mainly grown in Gujranwala and Rawalpindi divisions in Punjab and Swat and Bajaur Agency in North West frontier Province of Pakistan (Tahir *et al.*, 1994). Diseases caused by fungi and viruses are a constant threat and often a limiting factor in the cultivation of lentil crop. *Ascochyta* blight, rust and pea seed-borne mosaic virus (PSbMV) mainly damages lentil crop. Lentil blight caused by *Ascochyta lentis* was first observed in Pakistan during 1982 (Khan *et al.*, 1983) causing heavy damage to lentil crop. During 1982-83, about 30-40% damage to lentil crop by blight was reported by Malik (1983). The pathogen is seed-borne and also perpetuates in infected debris. Rust caused by *Uromyces viciae-fabae*, is another important foliar disease of lentil. Complete crop loss may take place an early infestation (Sepulveda, 1989). Rust is a common disease of lentil in Sialkot district, the major lentil growing area. It infects all the aerial plant parts under severe infection plants dry even before they form seeds. Seed size is greatly reduced. Circular brown patches of dry plants could be seen in the field (Khare & Agrawal, 1978).

Lentil becomes infected with at least 10 viruses under field conditions (Bos *et al.*, 1988; Makkouk *et al.*, 1992), but in Pakistan pea seed-borne mosaic virus (PSbMV) is more common than other viruses. Five viruses are known to be seed-borne in lentil (Makkouk & Azzam, 1986). According to a survey conducted in Pakistan in 1997, about one in five plants of lentil was virus-infected, 15% fields having incidence of higher than 50% (Makkouk *et al.*, 2001). PSbMV is reported to decrease yield about 72% when infection occurs at early stage (Aftab *et al.*, 1992).

The use of genetic resistance may be the most effective strategy to control these lentil pathogens. In some international collections, lentil cultivars and lines with moderate resistance to *A. lentis* has been identified (Singh *et al.*, 1982; Sugha *et al.*, 1991; Nasir & Bretag 1996; Andhraennadi *et al.*, 1996). In case of rust, genetic differences among genotypes and sources of resistance have been reported by many authors (Nene *et al.*, 1975; Khare *et al.*, 1979; Shukla, 1984; Singh & Sandhu, 1988). Use of resistant cultivars and breeding for resistance is currently major control measures to supplement or replace the previously described preventive control, especially when and where hygienic measures are difficult to implement. The present study was undertaken with the objective to screen lentil germplasm to identify multiple sources of resistance to blight, rust and viral diseases.

### **Materials and Methods**

During winter season of 1998-99, 590 lentil germplasm accessions were obtained from Plant Genetic Resources Institute (PGRI), National Agricultural Research Center, Islamabad. Each accession was planted in second week of October, in a single replication with 4m-row length, 30 cm row to row distance. One row of susceptible check was planted after every two-test row. The weather conditions were highly favorable for disease development, particularly for blight and rust. Data regarding blight, rust and viral diseases particularly for PSbMV were recorded by using following 1-9 scoring scale at 15 days interval.

#### **For Ascochyta blight:**

- 1 = No lesions visible (Highly Resistant),
- 3 = Few scattered lesions seen after careful searching (Resistant),
- 5 = Lesions, common and easily observed, but little defoliation. Only in one or two patches in plot (Moderately Resistant),
- 7 = Lesions very common and damaging (Susceptible),
- 9 = Lesions extensive many plants killed (Highly Susceptible),

#### **For rust:**

- 1 = No pustules visible (Highly Resistant),
- 3 = Few scattered pustules, usually seen after careful searching (Resistant),
- 5 = Pustules common on leaves and easily observed but causing no apparent damage (Moderately Resistant),
- 7 = Pustule very common and damaging, few pustules on petioles and stems (Susceptible),
- 9 = Pustule very extensive on all plant parts, some death of leaves and other plant parts (Highly Susceptible),

**For viral diseases:**

- 1 = No infection (Highly Resistant),
- 3 = 1-10% plants affected (Resistant),
- 5 = 10-20% of plants affected (Moderately Resistant),
- 7 = 20-50% of plants affected (Susceptible),
- 9 = More than 50% of plants affected (Highly Susceptible).

**Results and Discussions**

This study was conducted under natural infection conditions in the field, The climatic conditions during the experiment were favorable for the development of fungal (*Ascochyta* blight and rust) as well as viral diseases (PSbMV). Results of disease reaction of germplasm accessions have been summarized in Table 1. A wide range of variation in disease reaction was observed among lentil genotypes. Majority of the genotypes were susceptible to rust, whereas maximum number of genotypes showed resistant or tolerant reaction against *Ascochyta* blight (*Ascochyta lentis*) and PSbMV. Out of 590 accessions, 70, 65 and 125 accessions were rated as 1 against *Ascochyta* blight, rust and PSbMV respectively. Only one line (66013-6) was observed as resistant to blight, rust and virus. Twenty three 23 lines, 66038-9, 66038-10, 66040-8, 66054-4, 66054-5, 66054-6, 66054-7, 66065-9, 66065-10, 66066-1, 66066-2, 66066-3, 66089-8, 66089-9, 66115-9, 66115-10, 66116-1, 66116-2, 66116-3, 66116-4, 66116-5, 66116-6, 66116-7 were found resistant to both blight and PSbMV, whereas three lines, 66034-8, 66050-9, 66051-1 were found as resistant to rust and PSbMV. Two lines 66013-3 and 66013-4 were found resistant to blight and rust (Table 2).

There are several reports that lentil crop suffers fungal (*Ascochyta* blight, wilt, root rot, rust) and viral diseases. These diseases may causes heavy losses ranging from 25 to 72% depending on the cultivars and pathogen type (Sepulveda, 1985; Singh *et al.*, 1986). Johansen *et al.*, (1994) recently reported the seriousness of lentil blight and rust. The mode of infection in those two diseases is more or less similar. These diseases spread through infected or contaminated seed or through infected plant debris (Richardson, 1979; Morrall & Sheppard, 1981; Morrall & Beauchamp, 1988). In order to control these diseases integrated approaches have been recommended by different authors from time to time which include crop rotation, field sanitation, use of disease free seeds and seed treatment with different fungicides or foliar spray of fungicides and hot water and dry heat treatment. (Morrall & sheppard, 1981; Russell *et al.*, 1987; Seid & Beniwal, 1991). Use of the resistant cultivars is the most economical and environmentally safe and sound control measure for disease management. Some sources of resistance against rust disease have been identified in different part of the world, BARI Masur 2 in Bangladesh (Anon., 1995); Centinela-INIA (ILL 5540) (Bascur & Sepulveda, 1989) in Chile; INIAP-406 (ILL 5764) in Ecuador; Gudo (ILL 5748) and Ada'a (ILL 6027) in Eithiopia (Bejiga & Anbessa, 1994); Precoz (ILL 4605) in Morocco and Manserha-89 (ILL4605) in Pakistan (ICARDA, 1994). NEL 358 (ILL 5680) (Million & Beniwal, 1988). Similarly, resistance to *Ascochyta* blight has been reported from Canada (Tay, 1989), India (Singh *et al.*, 1982), Newzealand (Cromey *et al.*, 1987) and Pakistan (Hussain *et al.*, 1998; Iqbal *et al.*, 1990).

**Table 1. Reaction of *Ascochyta* blight, rust and virus diseases on lentil accessions.**

<b>Disease reactions</b>	<b>Accessions</b>
<b>Resistant to blight</b>	66013-3, 66013-4, 66013-6, 66038-9, 66038-10, 66040-8, 66040-9, 66041-1, 66044-6, 66044-7, 66045-1, 66045-2, 66045-3, 66053-5, 66053-8, 66053-10, 66054-1, 66054-2, 66054-3, 66054-4, 66054-5, 66054-6, 66054-7, 66054-8, 66054-9, 66055-1, 66055-3, 66055-4, 66055-5, 66055-7, 66055-9, 66056-2, 66056-3, 66056-4, 66056-5, 66056-8, 66056-9, 66056-10, 66057-3, 66057-4, 66057-5, 66057-6, 66057-7, 66057-8, 66057-9, 66059-3, 66059-9, 66059-10, 66060-1, 66060-2, 66060-3, 66060-4, 66065-9, 66065-10, 66066-1, 66066-2, 66066-3, 66073-1, 66073-2, 66089-8, 66089-9, 66115-9, 66115-10, 66116-1, 66116-2, 66116-3, 66116-4, 66116-5, 66116-6, 66116-7.
<b>Resistant to rust</b>	66005-9, 66007-1, 66007-5, 66007-6, 66007-7, 66007-9, 66008-3, 66008-4, 66009-1, 66009-2, 66009-3, 66009-4, 66009-5, 66009-6, 66009-7, 66009-8, 66009-9, 66010-1, 66010-2, 66010-4, 66010-7, 66012-10, 66013-2, 66013-3, 66013-4, 66013-6, 66013-7, 66013-9, 66017-2, 66017-3, 66017-4, 66017-5, 66017-6, 66017-7, 66017-9, 66018-5, 66018-6, 66018-7, 66018-10, 66019-1, 66019-3, 66019-6, 66020-10, 66023-8, 66034-1, 66034-2, 66034-3, 66034-4, 66034-5, 66034-6, 66034-7, 66034-8, 66037-10, 66038-1, 66038-6, 66038-7, 66038-8, 66040-2, 66040-3, 66040-4, 66040-5, 66040-6, 66040-7, 66050-9, 66051-1.
<b>Resistant to virus</b>	66013-6, 66020-8, 66021-4, 66021-7, 66024-4, 66024-5, 66024-6, 66024-7, 66024-9, 66025-1, 66025-2, 66026-2, 66026-5, 66026-6, 66026-8, 66026-9, 66026-10, 66027-1, 66027-2, 66027-3, 66027-4, 66027-8, 66027-9, 66031-1, 66031-3, 66031-4, 66031-9, 66031-10, 66032-3, 66034-8, 66036-4, 66036-7, 66036-9, 66036-10, 66037-1, 66037-2, 66038-9, 66038-10, 66039-1, 66039-2, 66039-5, 66039-6, 66039-8, 66039-9, 66040-8, 66041-7, 66041-9, 66043-3, 66043-4, 66043-5, 66043-6, 66043-7, 66043-8, 66043-9, 66043-10, 66044-1, 66044-2, 66044-3, 66044-4, 66046-2, 66046-3, 66046-6, 66046-9, 66047-2, 66048-9, 66048-10, 66049-5, 66049-6, 66050-9, 66051-1, 66054-4, 66054-5, 66054-6, 66054-7, 66060-2, 66060-3, 66060-4, 66060-5, 66062-1, 66062-2, 66062-3, 66062-4, 66062-5, 66062-8, 66064-2, 66064-3, 66065-9, 66065-10, 66066-1, 66066-2, 66066-3, 66066-4, 66066-5, 66066-6, 66067-7, 66067-8, 66089-5, 66089-6, 66089-8, 66089-9, 66091-4, 66091-5, 66091-6, 66093-3, 66093-5, 66093-6, 66093-8, 66093-9, 66103-1, 66103-7, 66103-8, 66104-5, 66104-6, 66104-7, 66104-8, 66104-9, 66115-9, 66115-10, 66116-1, 66116-2, 66116-3, 66116-4, 66116-5, 66116-6, 66116-7,

**Table 2. Reaction of multiple resistant in lentil accessions.**

<b>Disease reactions</b>	<b>Accessions</b>
Resistant to blight, rust and viral diseases complex	66013-6
Resistant to blight and rust diseases	66013-3, 66013-4
Resistant to blight and virus diseases	66038-9, 66038-10, 66040-8, 66054-4, 66054-5, 66054-6, 66054-7, 66065-9, 66065-10, 66066-1, 66066-2, 66066-3, 66089-8, 66089-9, 66115-9, 66115-10, 66116-1, 66116-2, 66116-3, 66116-4, 66116-5, 66116-6, 66116-7
Resistant to rust and virus diseases	66034-8, 66050-9, 66051-1

According to the survey conducted in 1997 two viruses such as PSbMV and Cucumber mosaic viruses (CMV) are common in lentil growing areas of Pakistan. Both viruses are seed-borne in lentil and spread by several species of aphids under field conditions. The infected seed becomes major sources of infection under field condition at initial stage. In the present study, most of the lentil accessions were found free of virus symptoms, which indicate that the accessions were free of seed-borne infection. However, a few accessions were infected and the infection may be from infected seed. PSbMV as well as CMV were reported in lentil germplasm when evaluated under field as well as greenhouse conditions (Bashir *et al.*, 1995a; 1995b). In order to obtain virus free seed of lentil, the evaluation of lentil germplasm to produce virus free seed is essential, otherwise the infected seed may spread the pathogen from one locality to other. PSbMV spread by several species of aphids in the field. In the present study, we did not find abundant population of aphids, therefore, the overall viral disease incidence was low, and more accessions were found free of viral symptoms. The accessions found free of viral infection will be tested under greenhouse conditions to verify their resistance.

In present studies, multiple resistances have been found against blight rust and viral diseases in local germplasm collections. Mansera-89 was evaluated from exotic breeding material and released for multiple resistant to *Ascochyta* blight and rust in Pakistan. However, the local germplasm was never found for such unique multiple resistances against these major diseases in the country before. The local germplasm has the adaptability genes and carries the advantage over exotic genetic material for using in the breeding program to develop varieties for high yield potential with wider adaptability and disease resistance against a complex of disease pathogens.

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