



Bragantia

ISSN: 0006-8705

editor@iac.sp.gov.br

Secretaria de Agricultura e

Abastecimento do Estado de São Paulo

Brasil

Butt, Haris; Mukhtar, Tariq; Batool, Maria  
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Bragantia, vol. 75, núm. 1, enero-marzo, 2016, pp. 76-78  
Secretaria de Agricultura e Abastecimento do Estado de São Paulo  
Campinas, Brasil

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# *Spilocaea pyracanthae* causing leaf scab on loquat in Pakistan

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**Abstract:** Loquat is attacked by many phytopathogenic fungi. Among these *Spilocaea pyracanthae* is of economic importance. The fungus received no attention in Pakistan and some other parts of the world. The current study is focused on the symptomatology of this disease and the etiology of the fungus. During extensive surveys of loquat orchards in 2013 heavy infestations of this disease were observed. The leaf symptoms were observed as olive brown, opaque necrotic sub-circular spots, with light brown coloration in the center. Two or more spots coalesced in severe infections

covering large leaf areas. The fungus was identified as *Spilocaea pyracanthae* on the basis of conidia and morphological characters. The pathogenicity of the fungus was confirmed on healthy plants under greenhouse conditions by following Koch's postulates. The fungus produced characteristic leaf scab symptoms on young leaves. This is the first report of *Spilocaea pyracanthae* causing leaf scab of loquat in Pakistan.

**Key words:** *Fusicladium eriobotryae*, loquat leaf scab, conidiophores, *Eriobotrya japonica*, pathogenicity.

Loquat (*Eriobotrya japonica* Lindl.) is one of the important sub-tropical fruit trees of Pakistan with an annual production of 10,479 tons (Abbasi et al. 2011). The major loquat producing provinces of the country are Punjab and Khyber Pakhtoon Khwa (KPK) (Hussain et al. 2009) contributing to 98% of the total production (Anonymous 2008). In KPK, it is mainly cultivated in Mardan, Peshawar and Hari Pur districts while, in Punjab, it is mostly grown in the Pothowar region (Hussain et al. 2007). Loquat leaves have great significance from medicinal and nutritional point of view. In China, loquat leaves are being used as folk medicine for the treatment of pulmonary tuberculosis (Zhang et al. 2004). Loquat leaves are also used to cure various skin diseases, pain, inflammation, coughing, diabetes and liver disorders (Nishioka et al. 2002; Hamada et al. 2004; Sakuramata et al. 2004).

There are numerous pathogens that attack loquat. Fire blight caused by *Erwinia amylovora* is a serious bacterial disease of loquat (Seymour 1965). Leaf spot incited by *Diplocarpon mespili* (= *Entomosporium mespili*) is another important disease of loquat (Batool et al. 2014) and several genera of subfamily Pomoideae of the Rosaceae family. The disease has proved itself capable of rendering an entire fruit crop unmarketable (Davidson

1985). Other pathogens reported to infect loquat include: *Pseudomonas eriobotryae* (canker), *Phytophthora* spp. (crown rot), *Lasiodiplodia theobromae* (= *Diplodia natalensis* — collar and root rot), and *Colletotrichum gloeosporioides* (anamorphic *Glomerella*) (anthracnose) (Crane and Caldeira 2006).

Loquat scab is a very common disease in the Southern Italy, especially in Sicily, South Africa, around the Mediterranean basin, and in the eastern regions of North America on different hosts (Gardner and Raabe 1966; Raabe and Gardner 1972; Butt et al. 2015). Very little information is available in literature regarding loquat scab and the associated fungus *Spilocaea pyracanthae*. The fungus was first reported on loquat in 1909 (Smith 1909). The disease resembles pear and apple scabs in all aspects caused by *Venturia pirina* and *V. inaequalis*, respectively (Ohlendorf 1999). Loquat is highly susceptible to leaf and fruit scab especially during wet seasons when control measures are inadequate. Scabbed fruit is rendered unsuitable for market and results in economic loss (Tous and Ferguson 1996; Caballero and Fernández 2004). In Pakistan loquat is a neglected fruit tree and no attention has been paid to fungal pathogens associated with it. The current study is focused on the symptomatology of →

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Received: Jun. 7, 2015 – Accepted: Jul. 21, 2015

this disease and the etiology of the fungus associated with leaf scab of loquat from Pakistan.

During extensive surveys of loquat orchards in 2013, spots were observed on leaves. The spotted leaves were brought to the laboratory for identification of disease symptoms and the associated pathogen. The color and shape of the spots were observed. For isolation of the pathogen, the spots were cut, and the surface was sterilized in ethanol and plated on autoclaved Potato Dextrose Agar (PDA) in 90 cm Petri plates. The plates were then incubated at 25 °C for the growth of the fungus. The fungus was purified from a single spore and identified on the basis of morphological characters (Schubert et al. 2003; Schubert and Braun 2005).

The pathogenicity of the fungus was confirmed by the following Koch's postulates. Leaves of one-year-old loquat plants were inoculated with conidial suspension of the purified fungus. A concentration of  $10^5$  conidia  $\text{mL}^{-1}$  of the fungus determined by haemocytometer was applied for pathogenicity test. Distilled water was used in control plants. The plants were kept in the glasshouse for one month at 25 °C with about 85% relative humidity. After production of symptoms and lesions, the fungus was re-isolated and cultured on PDA. One-year-old plants were inoculated for a second time with re-isolated culture of the fungus and were observed for production of symptoms. The fungus was again isolated from the infected plants and was cultured.

Leaf symptoms were observed as olive brown to dark colored, opaque necrotic sub-circular to circular spots, with light brown coloration in the center. In severe infections two or more spots coalesced and covered larger leaf areas. Under microscope, fungal mycelium appeared to be hyaline and immersed with rare superficial growth on media. Later, olive coloration of colonies without aerial mycelia was observed in maturation. Conidia had ovoid shape with no septation and round from the base while pointed at apex as previously described in the literature (Sánchez-Torres et al. 2007, 2009). Conidia was usually a solitary type with size varying from 5 – 8  $\mu\text{m}$  to 9 – 20  $\mu\text{m}$ . Conidiophores and conidia were pigmented and pale to dark brown colored with conidiophores usually bearing

single conidia. Conidiophores varied in size according to age. Conidiogenous percurrent proliferations were usually seen with a few annulations. The reproduction stage of the fungus was not observed in culture as the cultures were kept isolated at 20 °C for 3 months. In the present study we come across with the conidial stage of the fungus only and did not observe the reproduction stage. The fungus has been reported to reproduce asexually by means of conidia (Raabe and Gardner 1972). There is no evidence in the available literature that the fungus has teleomorph, although it has been reported once by D'Oliviera and D'Oliviera (1946). In the present study variations in size of conidia were observed from the average size of  $6.3 \times 16.3 \mu\text{m}$  reported by Ogawa and English (1991) and Sánchez-Torres et al. (2009). These variations might be due to different climatic conditions which can affect the morphology of the fungus.

The pathogenicity test confirmed the association of the fungus with the disease. The fungus produced the characteristic symptoms of leaf spots which have been observed during identification. The manifestation of symptoms was observed after ten days and became well-developed after three weeks. The findings are in accord with those of Raabe and Gardner (1972), who also inoculated loquat plants in green house and observed appearance of symptoms within two to three weeks. They also reported that only young leaves and twigs were susceptible to fungus; however, incubation period of the fungus showed variations which are attributable to temperature.

Based on symptoms, morphological characters and pathogenicity tests, the fungus was identified and confirmed as *Spilocaea pyracanthae*. To the best of our knowledge, this is the first report of *S. pyracanthae* causing leaf scab of loquat in Pakistan.

## ACKNOWLEDGEMENTS

The authors acknowledge Dr. Ather Rafi and Dr. Sayed Ahmed Zia, Department of Plant and Environmental Protection, National Insect Museum, National Agricultural Research Centre, Islamabad for providing lab facilities.

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