



First report of *Macrophomina phaseolina* causing charcoal rot of strawberry in Tunisia

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Received: 10 Jul 2015. **Published:** 11 Oct 2015. **Keywords:** crown rot, emerging pathogen

Strawberry is an annual crop along the coastal areas of northern Tunisia grown in plastic tunnels and in open fields. Charcoal rot in strawberry usually occurs after the plants are well established and begin to produce fruit: the roots become necrotic, initially older leaves wilt, turn greyish-green, and dry up (Fig. 1a), but in time all the foliage collapses and dries up (Fig. 1b). During the growing season September 2014 to June 2015 all these symptoms were observed in strawberry plants (cv. Camarosa) in more than 20 fields in the Cap Bon peninsula.

Cross sections of the crowns of strawberry plants with these symptoms showed necrotic tissue and brown-red to dark brown areas of the vascular and cortical tissues (Fig. 2). Pieces of necrotic crowns and roots disinfected with 0.5% hypochlorite for 30 seconds were placed on potato dextrose agar (PDA). After five days of incubation at 25°C, dark cultures with numerous dark oblong sclerotia immersed in the isolation medium were observed (Fig. 3). DNA isolated from five isolates was amplified using primers MpKF1 and MpKR1 that had previously been shown to be specific for *Macrophomina phaseolina* (Babu *et al.*, 2007). The resulting 350-bp product together with the morphological observations of the colonies confirmed that the isolates corresponded to *M. phaseolina*.

Pathogenicity tests were performed on propagated runner plants grown in sterile potting soil under greenhouse conditions for three months. Three isolates, each derived from a single sclerotium, were used for pathogenicity tests. Each isolate was used to inoculate six strawberry plants (cv. Camarosa). Plants were inoculated by inserting a colonised toothpick into each crown (Mertely *et al.*, 2005), while as a control, sterile toothpicks were inserted into the crown of control plants. All inoculated plants collapsed or died within two weeks of inoculation, while the control plants remained healthy during the observation period. The pathogen was re-

isolated from inoculated plants and PCR confirmed the presence of *M. phaseolina* DNA in infected plant tissues. Charcoal rot disease caused by *M. phaseolina* has been reported on strawberry in USA, Spain and Chile (Koike, 2008; Avilés *et al.*, 2008; Sánchez *et al.*, 2013). To our knowledge, this is the first report of *M. phaseolina*, an emerging and devastating fungal pathogen, on strawberry in Tunisia. This disease is a potential threat for Cap Bon strawberry production, which is a major source of employment and income in this region.

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Figure 1



Figure 2

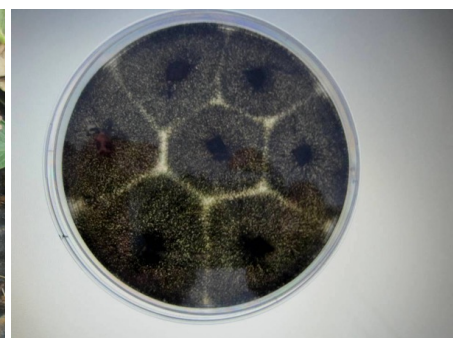


Figure 3

To cite this report: Hajlaoui MR, Mnari-Hattab M, Sayeh M, Zarrouk I, Jemmali A, Koike ST, 2015. First report of *Macrophomina phaseolina* causing charcoal rot of strawberry in Tunisia. *New Disease Reports* **32**, 14. <http://dx.doi.org/10.5197/j.2044-0588.2015.032.014>
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