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First Report On Neodeightonia Phoenicum As A Potential Contaminant Of Date Palm Tissue Culture In Iraq

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Abstract: In this work, the fungal species Neodeightonia phoenicum was isolated from date palm tissue cultures and characterised morphologically and molecularly by using internal transcribed spacer primers. Notably, N. phoenicum has not been previously reported as a causal agent of fungal contamination in date palm tissue cultures in Iraq.

Key words: date palm, fungal contamination, morphology, Neodeightonia phoenicum, phylogenetic analysis

Date palm (Phoenix dactylifera L.) is a good food and economic source because it produces fruits with high nutritional value. Its fruits contain sugars, vitamins, mineral elements and energy. Therefore, this species has an important role in the food industry (Amal et al., 2015). The propagation of date palm via traditional methods is slow given the low number of shoots. Moreover, the offshoots must remain with the mother tree for periods of 2–3 years before separation to form a strong root system. Separation requires extensive experience, and the separation area must be maintained free of contaminants. Additionally, offshoot cultivation has a low success rate. Therefore, the world turned to using the in vitro technique (tissue culture) involving callus stimulation and somatic embryogenesis induction as a modern method for date palm propagation. This method has been used for commercial propagation by several international companies specialising in this field, as well as by researchers (Al-Khalifah and Shanavaskhan, 2012). This technique enables the rapid propagation of healthy and true-to-type date palms (Al-Samir et al., 2015). Microbial contamination with fungi or bacteria is one of the most important barriers to date palm tissue culture (Odutayo et al., 2007; Abass, 2013). Microbial contaminants are introduced from the excised plant materials and from the environment surrounding the laboratory or during the culture process or the performance of workers and sterilisation processes. The contaminants remain in well-sterilised cultured plants, leading to a reduction in the growth rate and an increase in the death rate of plants (Abahmane, 2017). The fungal contaminants present in relevant farms have been comprehensively examined to detect the fungal pathogens in date palm tissue cultures.

This research specifically focused on the identification of certain Neodeightonia sp. (Neodeightonia phoenicum) causing the fungal contamination of embryonic callus in date palm tissue culture. N. phoenicum is one of the causes of fungal contamination in date palm tissue cultures. Primarily, the phenotypic identity of the isolates was determined on PDA medium and under standardised conditions (Fig. 1 A to C) because the isolates had been found to match the fungi mentioned in a number of relevant reports (Phillips et al. 2008; Ligoxigakis et al. 2013; Konta et al. 2016; Resna and Talaat.2020; Fonseca et al., 2020).

A number of studies have found that the examined fungus is a pathogenic causative agent in date palms. Phillips et al. (2008) recorded the fungus N. phoenicum in Phoenix sp. in Spain, and Ligoxigakis et al. (2013) mentioned that this fungus causes Phoenix sp. rot in Greece. Konta et al. (2016) reported that this fungus causes the disease of Calamus palms in Thailand. N. phoenicum has was also been recorded as a cause of date palm root rot disease in Qatar (Resna and Talaat, 2020).

Colonies were in the form of fungal mycelia spreading ring-like with pink centres surrounded by white grains and grey-black rings. The edges of the colony were in the form of a white to grey fluffy growth. Conidia were ovoid to elongate or spherical in shape, divided by one septate and brown to dark brown in colour (24–31.2 × 7.2–12 µm). Conidiophores appeared as short formations that were distinct from fungal hyphae. They were wide and yellow in colour with short divisions and swelling at the site of their attachment to the conidia. The mycelia formed chlamydorospores, which were brown to dark brown.

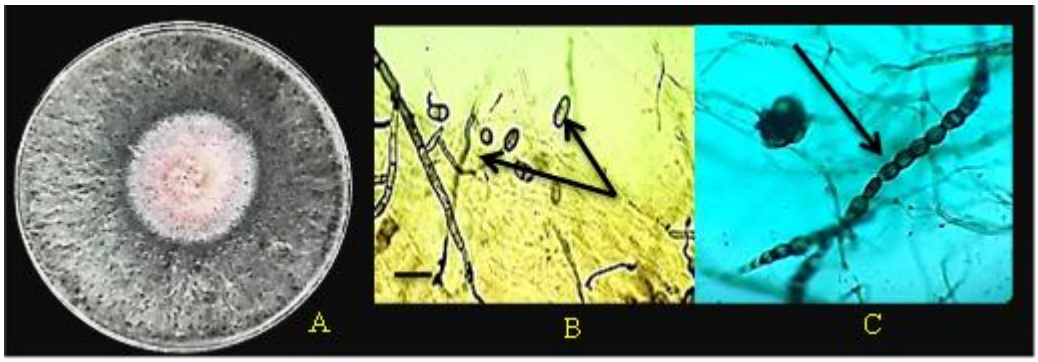

Fig. (1) Morphological traits of N. phoenicum.

(A) colony morphology on PDA after 7 days, (B) Conidiophores and Conidia (C) Chlamydorospore with a magnification of 40 x. Scale bar = 10 µm.

Notably, reference studies have not reliably discriminated phenotypic traits; moreover, the phylogenetic description of some Neodeightonia spp. remains confusing (Phillips et al., 2008; Fonseca et al., 2020) . In addition, the internal transcribed spacer (ITS) region from relevant isolates was amplified with the primers ITS1 and ITS4 and sequenced (GenBank Accession No. OL589157.1). The 583-bp amplicons shared 99.48% identity with N. phoenicum (GenBank Accession No. KF766198.1). Phylogenetic analysis identified the isolated fungus as N. phoenicum, N. phoenicum inhibits the growth of embryonic calli and covers them completely. Many studies have confirmed that the most common contaminants of date palm tissue culture are fungi, which can inhibit the growth and cause the death of

embryonic calli (Abass, 2017; Abeer and Abdel, 2017; Ahmed and Abass, 2022). This study is the first to report the fungus N. phoenicum as one of the causal agents of fungal contamination in date palm tissue cultures in Iraq.

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