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First Report of Dollar Spot caused by *Clariireedia jacksonii* and Brown Ring Patch caused by *Waitea circinata* var. *circinata*, on *Agrostis stolonifera* in Spain.

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In June 2015, small, straw-coloured, circular spots with abundant white aerial mycelium, which one month later turned into irregular patches, were observed on a large area of creeping bentgrass (*Agrostis stolonifera* L.) putting green at the Manises Royal Golf Course, in Valencia, Spain. Lesions covered more than 80% of leaf tissue. Symptomatic creeping bentgrass samples were taken from the putting green and laboratory processed. Portions of diseased leaves and root tissues were surface disinfected for 1 min in 1.5% NaOCl, washed twice with sterile distilled water and plated onto potato dextrose agar (PDA) amended with streptomycin sulphate (0.5 g L⁻¹) (PDAS). Plates were incubated at 25°C in the dark and all colonies were transferred to PDA. Two different fungal colony types were obtained. One culture had white floccose mycelium, which turned cinnamon brown after 3-wk. The other had white cottony growth, that turned orange to brown as the culture aged. The ITS region of representative isolates CE-1 and CE-2 was amplified and sequenced with primers ITS1F (Gardes and Burns, 1993) and ITS4 (White et al. 1990). These isolates were identified as *Clariireedia jacksonii* C. Salgado, L.A. Beirn, B.B. Clarke, & J.A. Crouch and *Waitea circinata* var. *circinata* Warcup & P.H.B. Talbot, respectively (Accession Nos. MK418798 and MK418799), by sequence comparison in GenBank database. Sequences showed 100% identity and 100% query coverage with sequences of *C. jacksonii* CBS136618 (NR158355) and *W. circinata* var. *circinata* WRCC 0.1 (FJ755884), respectively. Pathogenicity tests were conducted with inoculum produced on red fescue (*Festuca rubra*) seeds that were soaked for 12 h in flasks filled with distilled water. Each flask contained 300 mL of seeds that were subsequently autoclaved three times after excess water was drained. Two fungal disks of a 2-week-old culture of isolate CE-1 (*C. jacksonii*) grown on PDA were placed aseptically in each flask, incubated at 25°C for 2 wk, and shaken once a week to avoid clustering of inoculum. A 4 g sample of this inoculum was spread over a creeping bentgrass (cv. 'L-93') turf cultivated in a polyethylene pot (40 cm height and 16 cm of diameter) filled with a sand (0.6 mm particle size)/peat mixture (90/10 w/w) and replicated 4 times. The same process

was carried out to confirm pathogenicity of isolate CE-2 (*W. circinata* var. *circinata*). Controls were inoculated with sterile uninoculated seeds. Pots were irrigated with 100 ml of water, individually covered with plastic bags during 72 h in order to reach optimum infection conditions, and placed in a controlled environmental chamber at 25°C during 12-h daylight. Symptoms of Dollar spot (circular straw colored spots) were observed 3 days after inoculation and brown ring patch symptomatology was observed after 13 days, whilst controls remained asymptomatic. Both, *C. jacksonii* and *W. circinata* var. *circinata* were reisolated from inoculated turf. To our knowledge, this is the first report of *C. jacksonii* and *W. circinata* var. *circinata* on *A. stolonifera* in Spain, providing useful information for a better management of soilborne pathogens on turfgrass in this country.

Gardes, M., and Bruns, T. D. 1993. ITS primers with enhanced specificity for basidiomycetes-applications to the identification of mycorrhizae and rusts. *Mol. Ecol.* 2:113-118.

White et al., 1990. Amplification and direct sequencing of fungi ribosomal RNA genes for phylogenetics. In: *PCR Protocols. A Guide to Methods and Applications*. Academic Press, San Diego, CA. pp 315–322.