

Stem rust (*Puccinia graminis*) identified on spring barley in the UK adjacent to infected *Berberis vulgaris*

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Wheat stem rust caused by the fungus *Puccinia graminis* f.sp. *tritici* is a notorious disease of wheat and barley (Leonard & Szabo, 2005). In western Europe, following many decades of absence the disease started to re-emerge in 2013 with sporadic wheat stem rust outbreaks reported in Germany, Sweden and Denmark (Hovmøller, 2019). In the same year a single wheat plant infected with stem rust was discovered in the UK, which marked the first record of the disease in over 60 years (Lewis et al., 2018). These initial outbreaks were seen as an early warning of the potential resurgence of a forgotten foe (Saunders et al., 2019). Accordingly, a much larger outbreak was recorded in Sicily in 2016 (Hovmøller, 2019).

The wheat stem rust pathogen is heteroecious undertaking asexual reproduction on cereals and grasses and completing its sexual cycle through infection of an alternate host, common barberry (*Berberis vulgaris*) and many other species in the *Berberis* genus (Leonard & Szabo, 2005). The importance of the alternate host as a source of genetically diverse inoculum historically led to legislation and exclusion campaigns to limit its presence particularly in western Europe, USA and Canada. However, due to the absence of the disease for many decades in western Europe, *B. vulgaris* has been increasing in prevalence. Accordingly, a wheat stem rust outbreak in Sweden in 2017 occurred in an area where *B. vulgaris* was located (Berlin, 2017).

In early-August 2019, we identified symptoms typical of stem rust on approximately 20 late-sown spring barley plants in Suffolk in the UK (Figures 1 and 2). These infected barley plants were recorded on the edge of a field within metres of an established *B. vulgaris* hedgerow that was also heavily infected in the spring with orange, tube like aecial structures typical of stem rust (Figures 3 and 4). DNA was extracted from infected barley stems and aecia identified on *B. vulgaris* leaves using the DNeasy Plant mini kit (Qiagen, UK) and the internal transcribed spacer (ITS) region amplified using primers 5ITS (5'-GGAAGTAAAGTCGTAACAAGGT-3') and 3ITS (5'-ACTCCTTGGTCCGTGTTTCA-3'). PCR products were sequenced (Genewiz, USA) and sequences deposited in Genbank (accessions to be added). A sequence alignment was then performed including 27 additional ITS sequences from various *P. graminis* formae speciales. Phylogenetic analysis was performed using a neighbour-joining approach with 1,000 bootstrap replicates (Figure 5). This analysis confirmed (i) the presence of *P. graminis*, and (ii) that the *P. graminis* ITS sequences from barley and *B. vulgaris* infections clustered in a clade with other *P. graminis* formae speciales that predominantly infect wheat (*Triticum aestivum*) and barley (*Hordeum vulgare*), wild rye (*Secale strictum*), and couch grass (*Elymus* spp.), which are too similar to differentiate using gene sequence analysis.

This report illustrates that stem rust can be identified late in the season on barley in the UK, most likely caused by inoculum originating on neighboring *B. vulgaris* plants. The early maturation of UK wheat and barley varieties may currently act as an effective control mechanism for stem rust infection. However, as the climate continues to shift, earlier summer temperatures could encourage wheat stem rust re-emergence, as was seen during the wheat stem rust outbreak that occurred in 2013 in Germany (Olivera Firpo et al., 2017). This supports the need for enhanced vigilance and monitoring across cereals and grasses in the UK.

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Figure Legends

Figure 1. Wheat stem rust identified on late sown spring barley in the UK. Black telia (left) and orange uredinia (right) were identified on the stem of a barley plant in early August 2019.

Figure 2. Erumpent black telia full of teliospores visible on the stem of a late sown barley plant in the UK. The infected barley plant was identified and collected in early August 2019.

Figure 3. Orange structures typical of *P. graminis* infection were identified on *Berberis vulgaris* plants.

Figure 4. Orange tube-like cup-shaped structures typical of *P. graminis* infection identified on *Berberis vulgaris* plants.

Figure 5. Gene sequence analysis confirms stem rust in the UK on barley and the alternate host *B. vulgaris*. Stem rust identified on barley and *B. vulgaris* grouped with formae speciales known to infect cereals (bottom clade) rather than wild and cultivated grasses (top clade). Phylogenetic analysis was performed using the sequence of the internal transcribed spacer (ITS) region amplified from barley and *B. vulgaris* (light blue circles), with 27 isolates of different *P. graminis* formae speciales, using a Neighbor-joining method. A total of 403 positions were included in the final dataset. Names are accession numbers from NCBI. Adapted from Lewis *et al.*, 2018.



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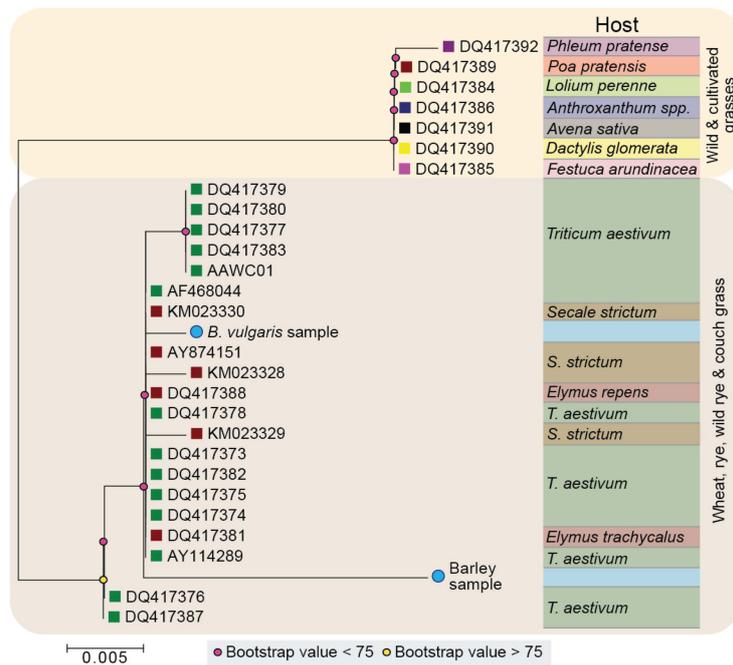


Figure 5. Gene sequence analysis confirms stem rust in the UK on barley and the alternate host *B. vulgaris*. Stem rust identified on barley and *B. vulgaris* grouped with formae speciales known to infect cereals (bottom clade) rather than wild and cultivated grasses (top clade). Phylogenetic analysis was performed using the sequence of the internal transcribed spacer (ITS) region amplified from barley and *B. vulgaris* (light blue circles), with 27 isolates of different *P. graminis* formae speciales, using a Neighbor-joining method. A total of 403 positions were included in the final dataset. Names are accession numbers from NCBI. Adapted from Lewis *et al.*, 2018.