

1 **Infestation of hop seed (*Humulus lupulus*) by chasmothecia of the powdery mildew fungus,**
2 ***Podosphaera macularis***

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10 Powdery mildew of hop (*Humulus lupulus*) is caused by *Podosphaera macularis*. Since, 1997
11 the disease has frequently caused severe economic loss in production regions in the Pacific
12 Northwestern region of the U.S., the primary area where hops are grown in the country (Gent et
13 al. 2008). *Podosphaera macularis* is heterothallic, but to date only the MAT1-1 mating type has
14 been confirmed in the Pacific Northwest (Wolfenbarger et al. 2015) and ascocarps of the fungus
15 have not been observed in this region (Gent et al. 2006). In the autumn of 2015, seed was
16 collected from wild hop plants at 7 locations in Minnesota for grow out and evaluation of various
17 traits. Prior to planting, seeds were examined under low magnification (30-50 \times) and in 9 of the
18 11 seedlots, representing 4 of 7 locations, the seed was found to be externally infested with
19 spherical to flattened, black chasmothecia (syn. cleistothecia). In infested lots, the number of
20 seed bearing chasmothecia averaged 45% (range 5 to 89%; $n = 107$ to 200 seeds per lot).
21 Scanning electron microscopy indicated chasmothecia had a mean diameter of 82 μm and were
22 shriveled with a concaved base. Chasmothecia were easily dislodged from the seed coat despite

23 the appendages being embedded in a mat of pannose mycelium. Conidiophores and conidia
24 were not observed. The morphological characters were consistent with the genus *Podosphaera*
25 (Braun et al. 2002; Wolfenbarger et al. 2015). Chasmothecia were confirmed as *P. macularis* by
26 extracting DNA from 10 to 15 seeds from each of 6 seedlots using a DNeasy PowerSoil Kit
27 (Qiagen, Carlsbad, CA) and amplifying and sequencing the MAT1-1 and MAT1-2 idiomorphs as
28 described by Wolfenbarger et al. (2015). The sequences obtained for MAT1-1 and MAT1-2
29 were identical among the extractions of the 6 seedlots. Standard nucleotide BLAST searches in
30 GenBank indicated that the sequences were 97% similar to MAT1-1 (accession KJ922755.1) and
31 100% similar to MAT1-2 (accession KJ741396.1) sequences of *P. macularis*. To our
32 knowledge, this is the first report of infestation of hop seed by chasmothecia of *P. macularis*.
33 Current quarantine laws that restrict import of planting materials for hop into Idaho, Oregon, and
34 Washington explicitly exempt seed. However, seed infested with chasmothecia may spread the
35 pathogen, potentially introducing novel isolates and mating types of the pathogen. Seed
36 transmission of powdery mildew organisms is scarcely reported (Jarvis et al. 2002), and studies
37 are needed to determine the risk of disseminating *P. macularis* on infested seed. Until such
38 information is available, caution is advised when moving seed from regions where powdery
39 mildew occurs.

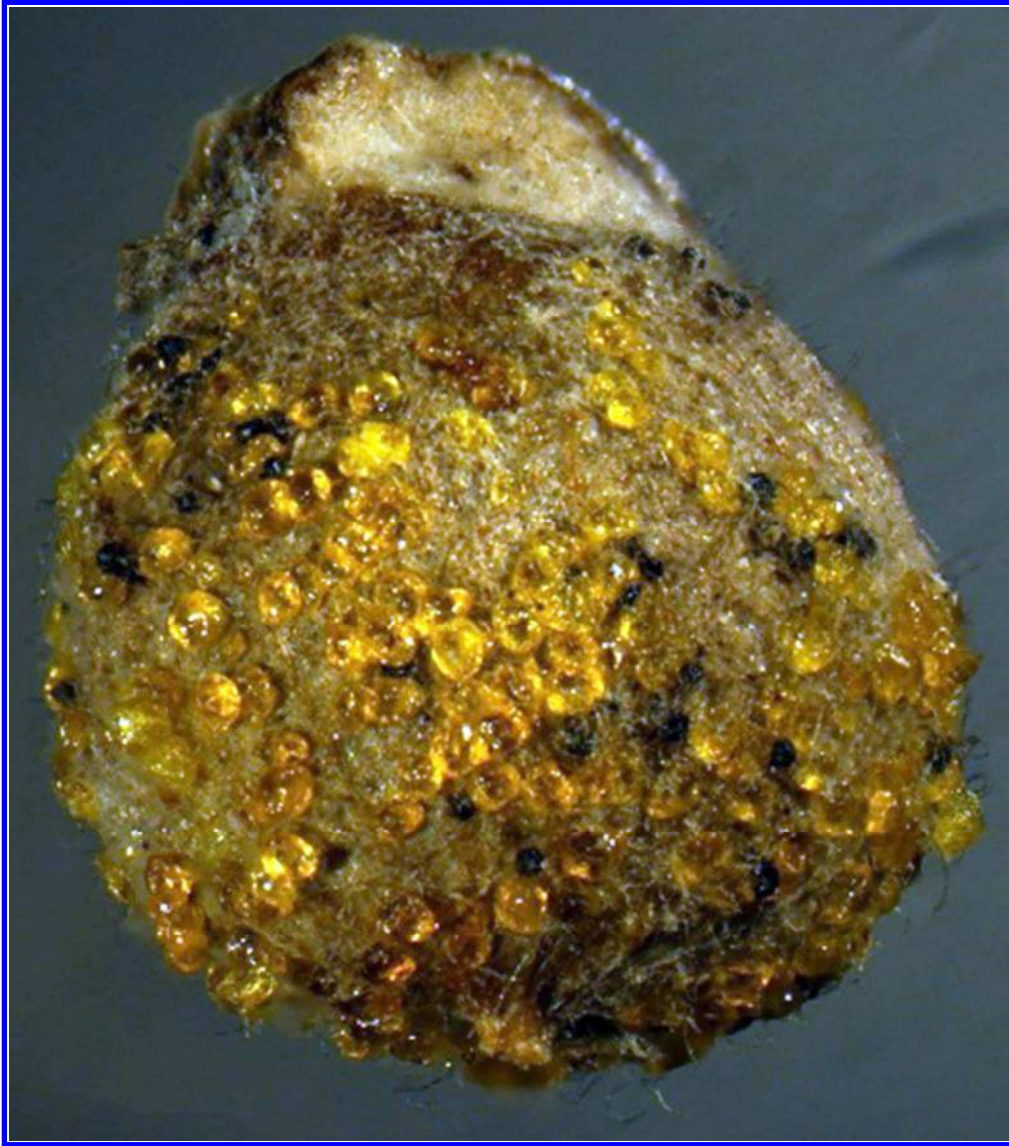
40

41 **References:**42 **Braun et al.** 2002. Pages 13 in: The Powdery Mildews: A Comprehensive Treatise, APS Press,
43 St. Paul.44 **Gent, D. H.,** et al. Plant Health Progress doi:10.1094/PHP-2008-0314-01-RV.

- 45 **Gent, D. H.**, et al. Plant Dis. 90:14333, 2006.
- 46 **Jarvis et al.** 2002. Pages 169 in: The Powdery Mildews: A Comprehensive Treatise, APS Press,
47 St. Paul.
- 48 **Wolfenbarger, S. N.**, et al. Plant Pathol. 64:1094, 2015.

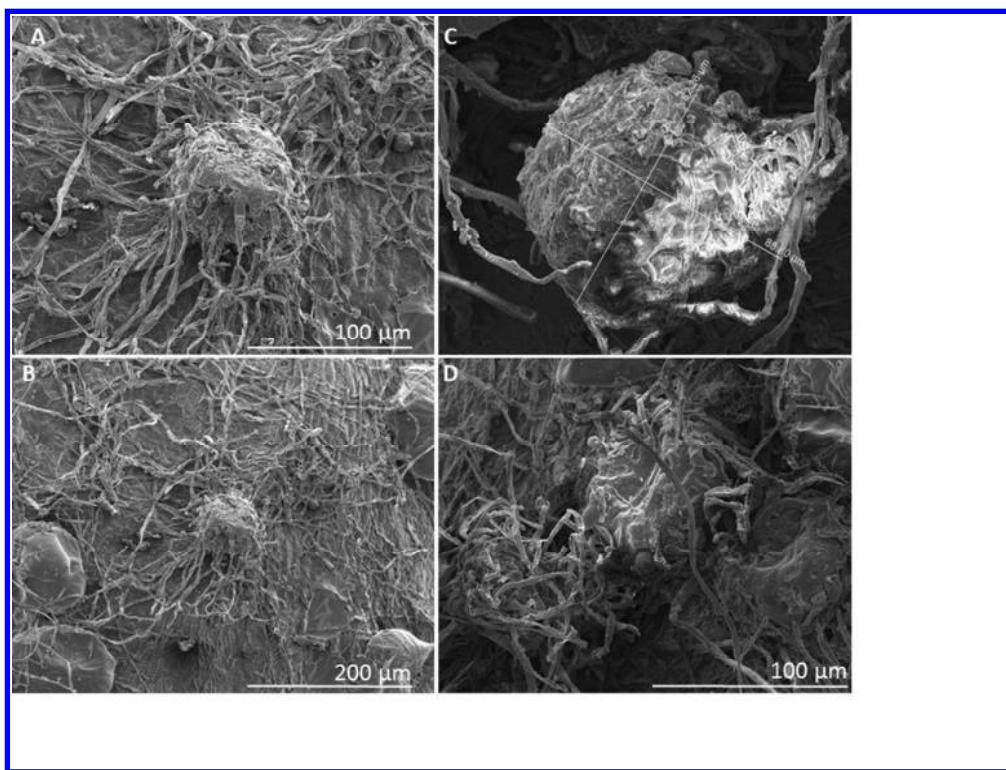
49 **Supplemental Figure 1.** Seed from a wild hop plant (*Humulus lupulus*) with numerous, black,
50 spherical chasmothecia of *Podosphaera macularis* and extensive mycelial colonization on the
51 seed coat. The larger yellow structures are lupulin glands. Note the prominent cluster of
52 chasmothecia on the bottom of the seed. Mycelia is most conspicuous on the top half of the
53 seed.

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55 **Supplemental Figure 2.** Scanning electron micrographs of chasmothecia of *Podosphaera*
56 *macularis* on hop seed. **A**, Chasmothecium with myceloid appendages on seed coat. **B**,
57 Chasmothecium on seed coat with appendages embedded in pannose mycelium. Larger, non-
58 descript structures are lupulin glands. **C**, Close-up with measurements of the diameter of an
59 ascocarp. **D**, Shriveled ascocarps with concaved basal portions.



Supplemental Figure 1. Seed from a wild hop plant (*Humulus lupulus*) with numerous, black, spherical chasmothecia of *Podosphaera macularis* and extensive mycelial colonization on the seed coat. The larger yellow structures are lupulin glands. Note the prominent cluster of chasmothecia on the bottom of the seed. Mycelia is most conspicuous on the top half of the seed.

68x77mm (220 x 220 DPI)



Supplemental Figure 2. Scanning electron micrographs of chasmothecia of *Podosphaera macularis* on hop seed. A, Chasmothecium with myceloid appendages on seed coat. B, Chasmothecium on seed coat with appendages embedded in pannose mycelium. Larger, non-descript structures are lupulin glands. C, Close-up with measurements of the diameter of an ascocarp. D, Shriveled ascocarps with concaved basal portions.

254x190mm (96 x 96 DPI)