Supplementary information

Supplementary tables:

Table S1: Fungal isolates used for whole genome sequencing. Previous genetic characterization of S1 and S2 isolates is presented in Stukenbrock et al, 2007a.

Table S2: Summary of de novo assemblies of Illumina paired end reads. Assemblies were done using the SOAP assembly software (Li et al, 2008).

Table S3: Summary of LastZ alignment of the 12 re-sequenced genomes to the reference genome of *M. graminicola*. Pairwise alignments were subsequently joined into a 12x multiple alignment.

Table S4: Lengths and numbers of dispensable chromosomes as assessed by pulsed field gel electrophoresis. Only chromosomes below 1Mb in size are included.

Table S5: Summary of within and between species variation and counts of shared polymorphisms between *M. graminicola* and S1 and *M. graminicola* and S2. Shared polymorphisms were divided as being either the same nucleotide polymorphisms (type 1) or different nucleotide polymorphisms (type 2).

Table S6: Summary of definitions for parameters used in the population genomic analyses.

Table S7: Summary of analyses of α estimated among genes with signal peptides and without. The dataset was correct for sequence composition to account for differences in GC content. Genes with zero counts were removed from the MK tables.

Table S8: α and f computed for Gene Ontology (GO) categories as defined by the JGI. The *S. passerinii* genome was used as outgroup for the analyses. As the number of fully aligned genes in the *S. passerinii* genome is less than between *M. graminicola*, S1 and S2 only 1406 genes were included in the analyses.

Table S9: Summary of mating type sequence analyses. K_a and K_s were estimated according to the method described in Nei and Gojobori, 1986.

Table S10: Summary of coalescence parameters from chromosome wise analyses as described in Dutheil et al. 2009.

Table S11: Primers for amplification of mating type loci

Supplementary figures

Figure S1: Detached leaf assay experiment where leaves of *T. aestivum*, *D. glomerata*, *E. repens*, *L. perenne* and *L. multilflorum* were inoculated with the 12 Mycosphaerella isolates studied here including *M. graminicola*, S1, S2 and *S. passerinii*. The virulence of each strain on each host was quantified by measuring the leaf area covered by pycnidia.

Figure S2: Mean values of divergence (Dxy) and within species polymorphisms (Pi) between *M. graminicola* (Mg) and S1 and Mg and S2. Shared polymorphisms were divided into mutations of the same nucleotide (type 1) or of different nucleotides (type 2). The distribution of type 1 and type 2 polymorphisms shared between Mg and S2 is shown across chromosomes. See Figure 2 for the same distribution in Mg and S1

Figure S3: Sliding window across chromosome 5 and 8 illustrating examples of heterogeneous distributions of shared polymorphisms type 1 (same nucleotide mutation in both species) across chromosomes. The proportions of shared polymorphisms were assessed in windows of 10kb aligned sequence between *M. graminicola* and S1.

Figure S4: Box plot of average K_a and K_s in calculated for all genes and for the subset of genes encoding signal peptides. The rates were obtained from the inter-specific comparisons of *M. graminicola* and S1 (MgS1) and *M. graminicola* and S2 (MgS2)

Figure S5: Illustration of the four coalescence models used in the coalhmm approach. Modified from Dutheil et al. 2009 (Dutheil JY, Ganapathy G, Hobolth A, Mailund T, Uyenoyama MK, Schierup MH. Ancestral population genomics: The coalescent hidden Markov model approach. *Genetics* **183:** 259–274) with permission from the Genetics Society of America © 2009.