## Supplementary Material

**Supplementary Table 3.** Oligonucleotides used in this study.

| Oligonucleotide name | Sequence (5' – 3') | Template | Product size (bp) | Purpose |
|---|---|---|---|---|
| Oex-Um11067_1 | [1]*tttt***CCATGG**AGTCACTCGGTCAAAATGGGAGAG | *U. maydis* gDNA | 2299 | To amplify the UMAG_11067 ORF |
| Oex-Um11067_2 | [2]tttgaacgatcGAGCGGAGCATGGTTAGAACC | | | |
| Oex-NOSt_1 | [2]ATGCTCCGCTCgatcgttcaaacatttggcaataaagtttcttaag | Plasmid pUMa2625 | 276 | To amplify NOS terminator for further fusion with UMAG_11067 ORF |
| Oex-NOSt_2 | [2]GGACAGCACCGCgatctagtaacatagatgacaccgcgcgc | | | |
| Oex-IP*locus*_1 | [2]tgttactagatcGCGGTGCTGTCCCG | *U. maydis* gDNA | 964 | To amplify a sequence downstream of the *IP locus* for HR. |
| Oex-IP*locus*_2 | [3]*ttt***GAATTC**GCAACGGATTCTACGATACCTGG | | | |
| [4]oex-Verif_1 | CCTGCTTGACTTGTGACCATGCC | oexUMAG_11067 gDNA | 4086 | To verify insertion of the overexpression cassette in *IP locus* |
| [4]oex-Verif_2 | CGACTTTGCCTGGTGCTGAC | | | |
| [4]oex-Verif_1 | CCTGCTTGACTTGTGACCATGCC | oexUMAG_11067 gDNA | 1760 | To verify insertion of the overexpression cassette in *IP locus* |
| [4]oex-Verif_3 | CTGGAGCAGTTCATGATGGTAAG | | | |
| [4]oex-Verif_4 | CGCTGAACAGATCCTCATTGACC | oexUMAG_11067 gDNA | 1877 | To verify insertion of the overexpression cassette in *IP locus* |
| [4]oex-Verif_5 | CATCAAATCAACGTCAGCCGTCG | | | |
| q-Um11067_1 | CGATGGCTTCCGCAATTA | *U. maydis* cDNA | 146 | To quantify gene expression of UMAG_11067 by qPCR |
| [5]q-Um11067_2 | CGTTGTAGTTGAGACCAAGAG | | | |
| q-Um04869_1 | CTTGCTACGGTCCAACATTTC | *U. maydis* cDNA | 149 | To quantify gene expression of UMAG_11067 by qPCR |
| [5]q-Um04869_2 | TCGCTACTCTCCCTACTCAA | | | |

[1] Thymine in lowercase and italics were added to increase the digestion efficiency after PCR amplification. Bold nucleotides indicate restriction recognition sequence for NcoI enzyme.

[2] The lowercase letters indicate nucleotides to fusion the PCR products by double-joint PCR.

[3] Thymine in lowercase and italics were added to increase the digestion efficiency after PCR amplification. Bold nucleotides indicate restriction recognition sequence for EcoRI enzyme.

[4] The use of this set of oligonucleotides on *U. maydis* SG200 gDNA does not produce any PCR product.

[5] These oligonucleotides were used to synthetize the first strand cDNA from RNA.