Supplementary Information

The polyketide synthase PKS15 has a crucial role in cell wall formation in *Beauveria bassiana*

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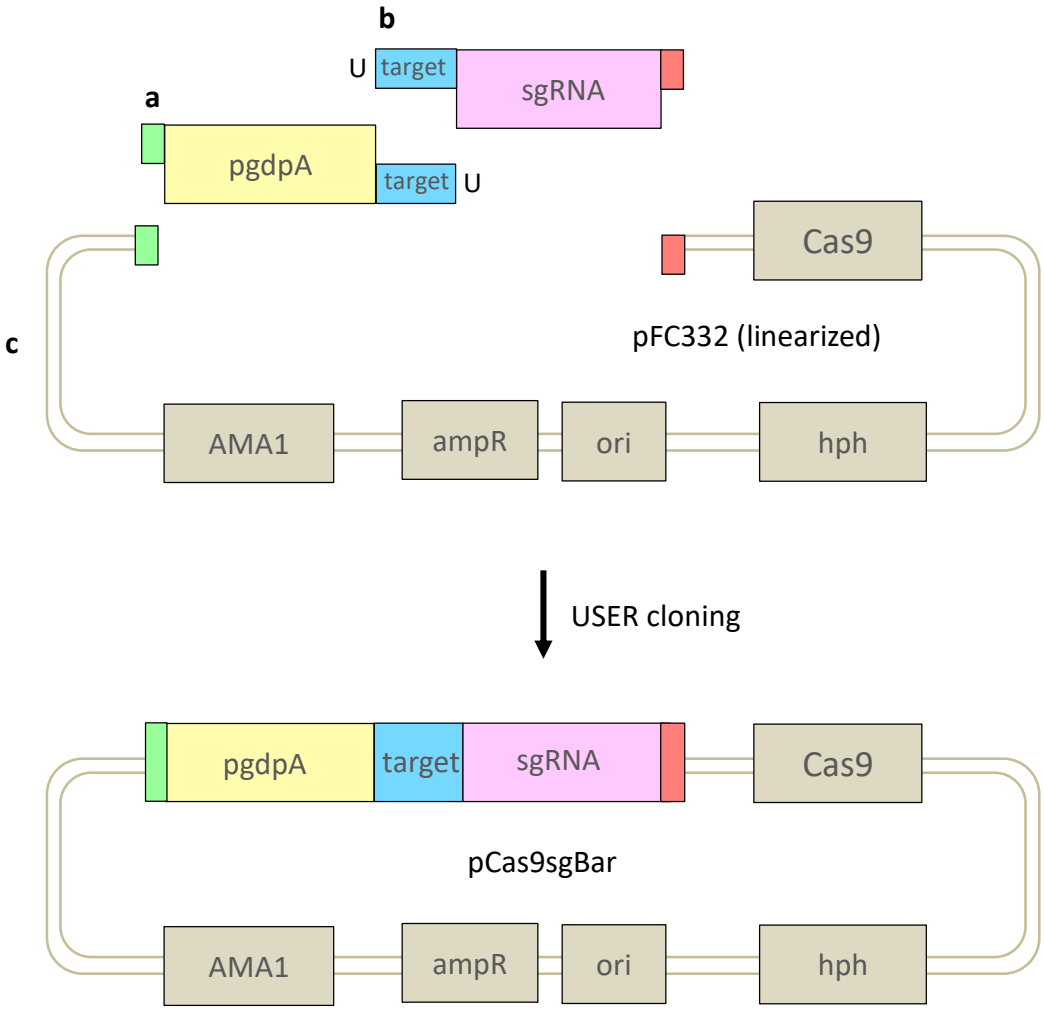

Supplemental Figure S1. Schematic representation of the pCas9sgBar plasmid construction. Detailed cloning protocols are described previously (Nodvig et al., 2015). Briefly, the guide RNA is assembled by a fusion of two fragments in order to introduce "our gene-specific guide RNA (sgRNA) sequence" in the vector. The gene-specific guide RNA sequence is incorporated in the primers used for the vector construction. The vector pFC334 was used as the template to amplify (a) pgdpA tRNA promoter element and (b) sgRNA expression cassette. Each PCR fragment was amplified by primers containing 25 nucleotides overhang (=target) and followed by a uracil base (U). Uracil DNA glycosylase and DNA glycosylase-lyase Endonuclease VIII (USER Enzyme) was used for elimination of the uracil bases in the PCR fragments. This enzyme digestion generated complementary overhangs at the ends of all three fragments, which facilitated the fusion of the two PCR fragments and the linearlized vector pFC332 by USER cloning in one step. (c) The vector backbone for construction of pCas9sgBar is derived from the pFC332. Sticky ends for USER cloning are achieved by restriction digestion of PacI (green) and Nt.BbvCI (red) sites in pFC332. It is noted that AMA1 is a plasmid replicator, originally reported in *Aspergillus nidulans* (Aleksenko and Clutterbuck, 1997) and recently used in Nodvig et al. (2015). In principle, an AMA1-containing plasmid could be easily lost when there is no selection. However, the autonomous replication of such plasmid remain to be proven in each fungus tested.

Supplemental Figure S2. Sequence alignment of the *pks15* locus of *B. bassiana* wild type (Query) and that of the complemented isolate G6 (Sbjct)

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Query  1     ATGCTCATCGACAAAATGGAGACGCCCACACCTCTTGCCATTGTCGGCATGGCCTGCCGC  60
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Query  361   CAACTTCTCGAATGCACATACGAAGCCCTGGAAAATGCTGGTCTACCCAAGGGTAAGATC  420
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Query  601   TGTTCCTCTAGTTTGCACGCTATGCATGCTGCTGTGCAGAGCATCCGAAGTGGCGACTGC  660
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Sbjct  661   GACTCTGCCATTGTTGCTGCCAGCAATCTATACATGAATCCGGGGCGATCTAATCCAAATG  720

Query  721   TCTATGCTGGGTCTCTTTACCCCAACAGGTAAGACTTACGCCTTCGATCACAGAGGCCAA  780
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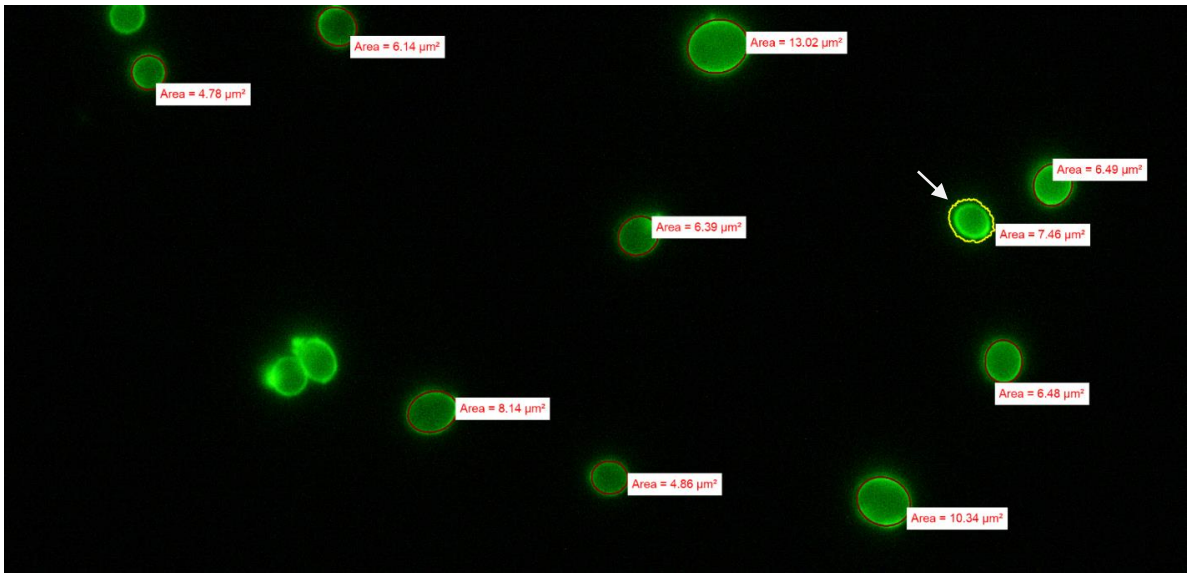
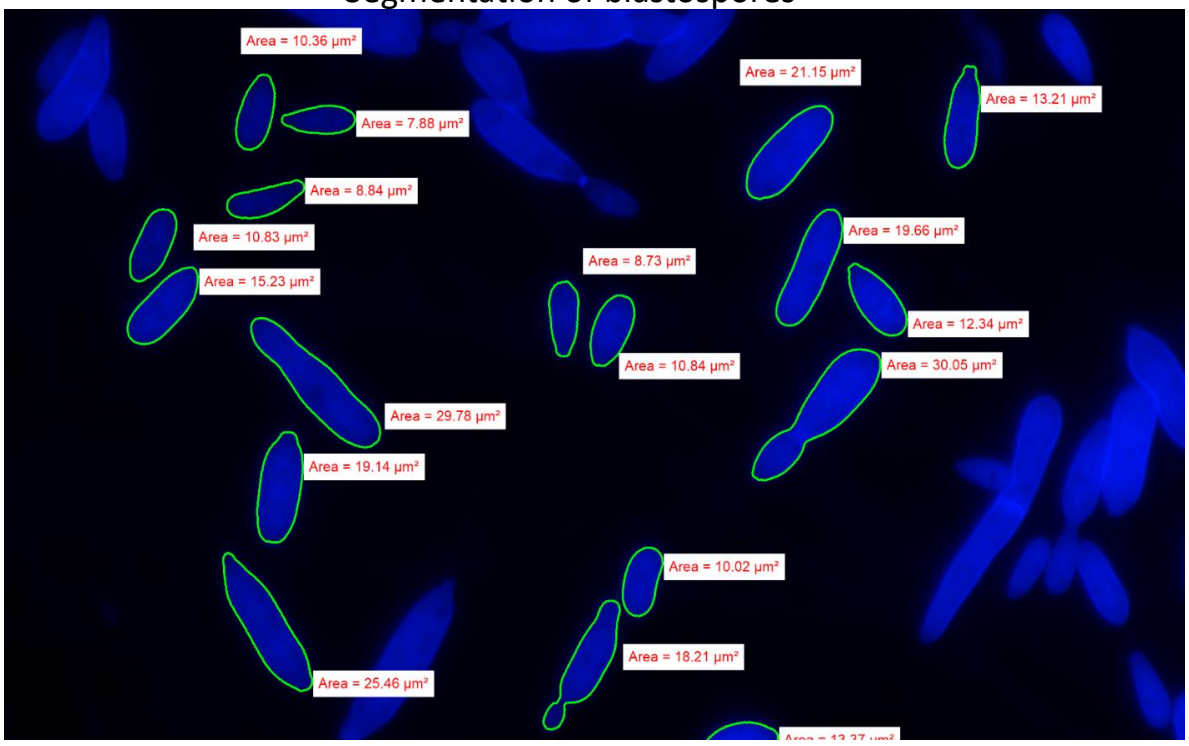
Segmentation of conidia



Segmentation of blastospores



Supplemental Figure S3. Determination of spore size and shape in 2D images. A. Segmentation of conidia, stained by FITC-tagged concanavalin A. The '5 Point Ellipse' feature in the area measurement of the NIS-Elements was manually used to mark the boundary of a conidium. **B.** Segmentation of blastospores, stained by calcofluor white. The 'Auto Detect' feature was used to indicate the boundary of a blastospore. Then, 'size' was automatically determined as 'area' by the software. The budding blastospores were excluded in the analysis. It is noted that if the 'Auto Detect' feature was used to mark the boundary of a conidium (indicated by an arrow), the segmentation was not accurate due to the fluorescence appearing outside the conidium.