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Supplemental Information

Candida albicans Morphogenesis Programs Control the Balance between Gut Commensalism and Invasive Infection

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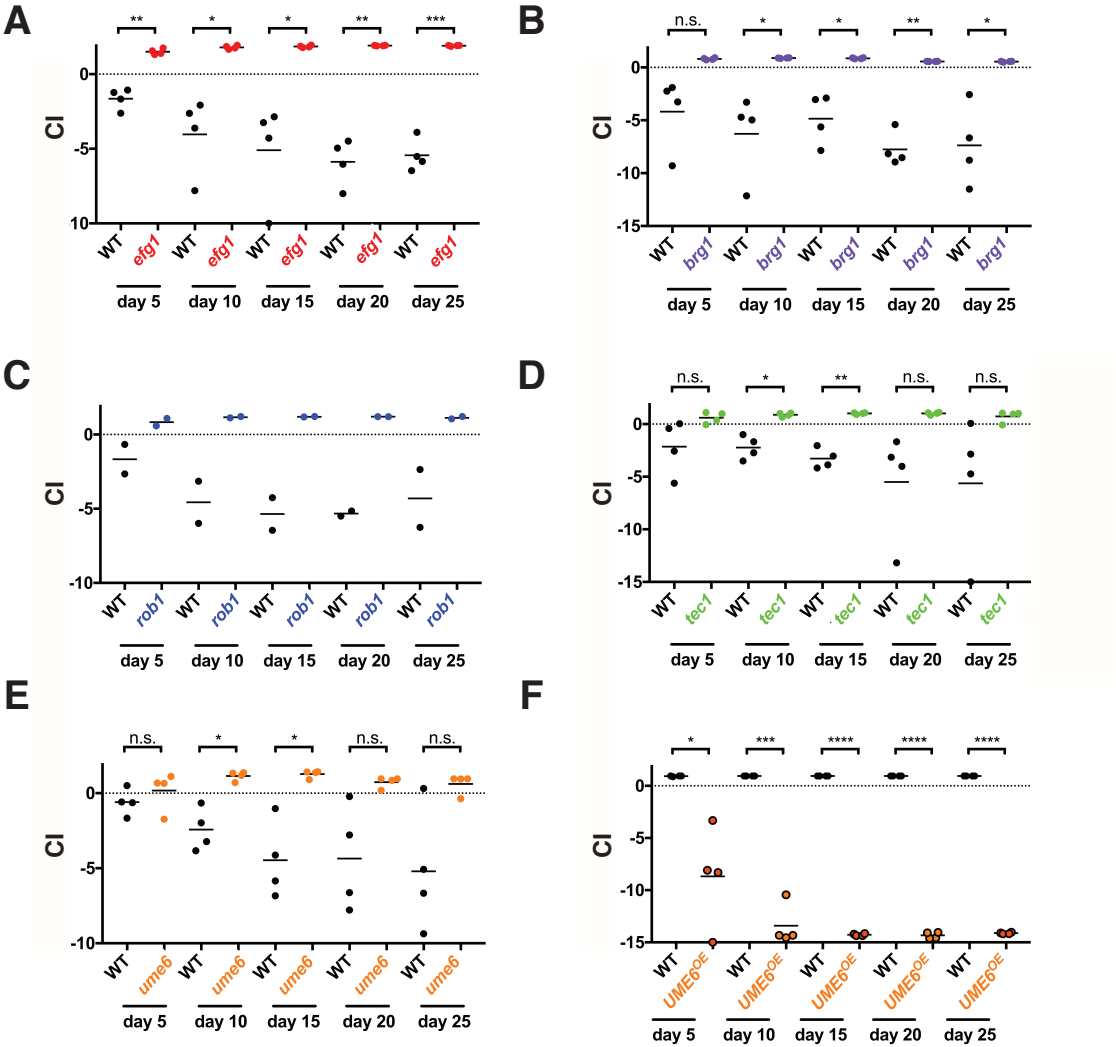

Figure S1. Validation of commensalism phenotypes of transcription factor mutants using independent isolates, Related to Figure 2. One-to-one commensal competitions with wild-type *C. albicans* were performed with independent isolates of each mutant depicted in Figure 2. Bars represent the mean relative abundance. Paired student's t-test: n.s. indicates notsignificant, * p<0.05, ** p<0.01, *** p<0.001, and **** p<0.0001.

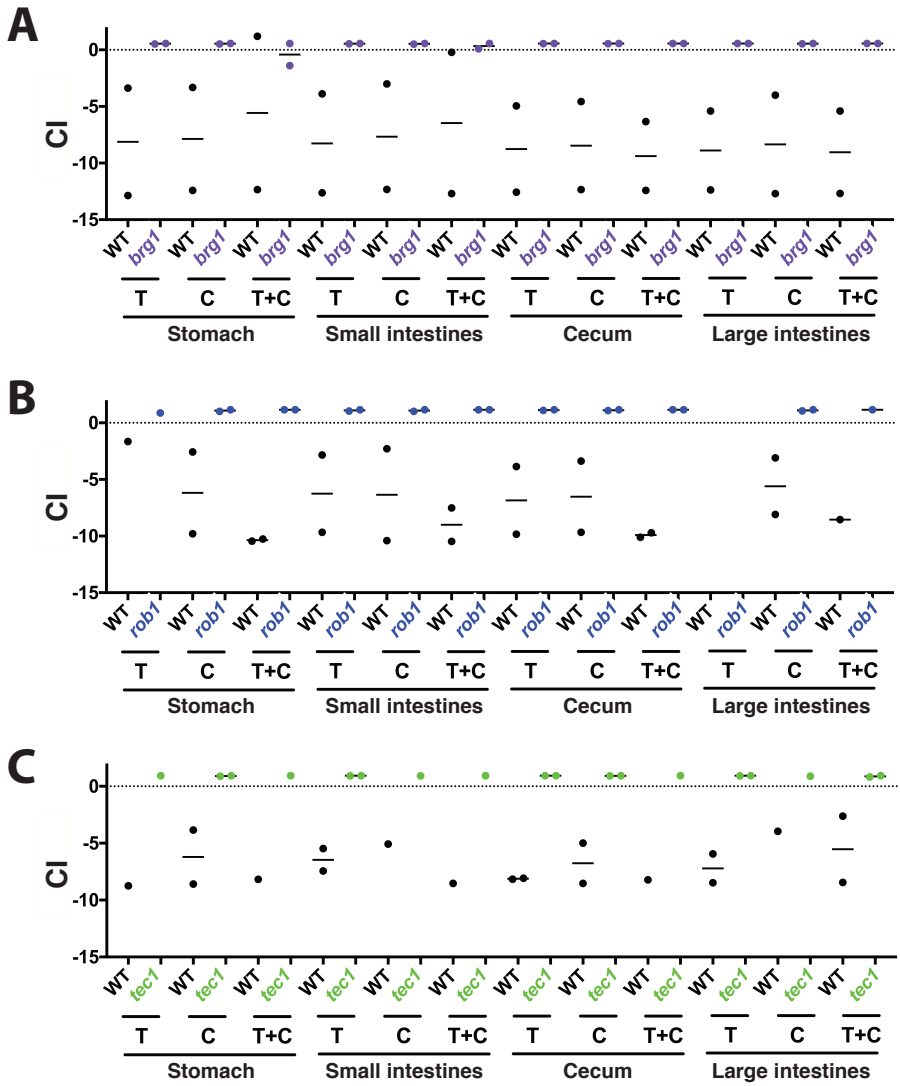

Figure S2. Assessment of competitive fitness of strains recovered directly from the host, Related to Figure 2. Animals used for the experiments presented in Figure 2C, D, and E were euthanized following collection of the Day 25 feces sample. The indicated segments of the digestive tract were recovered, and the relative abundance of *C. albicans* strains associated with tissues, luminal contents, or both was determined by qPCR of genomic DNA. The similarity of results obtained from feces vs. segments of the gut supports the use of feces samples for monitoring of commensal fitness. Bars represent the mean relative abundance.

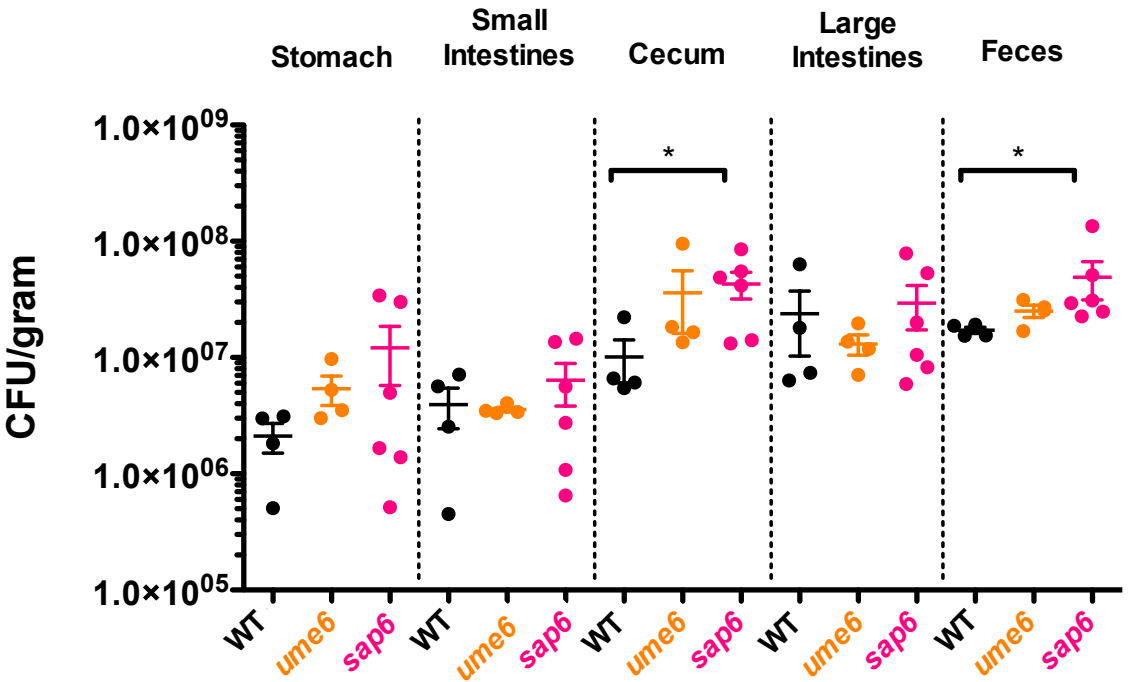

Figure S3. *ume6* and *sap6* mutants colonize the mammalian GI tract to similar levels as WT, Related to Figures 2 and 6.
Mice were gavaged with either WT (SN250), *ume6* (SN1479) or *sap6* (SN1664 or m886), as described in Star Methods. After 10 days, whole GI compartments were collected, weighed, homogenized, and plated onto Sabouraud agar plates containing antibiotics. CFUs were quantified one day after plating. Bars represent means + SEM. * $p<0.05$, unpaired Student's t-test.

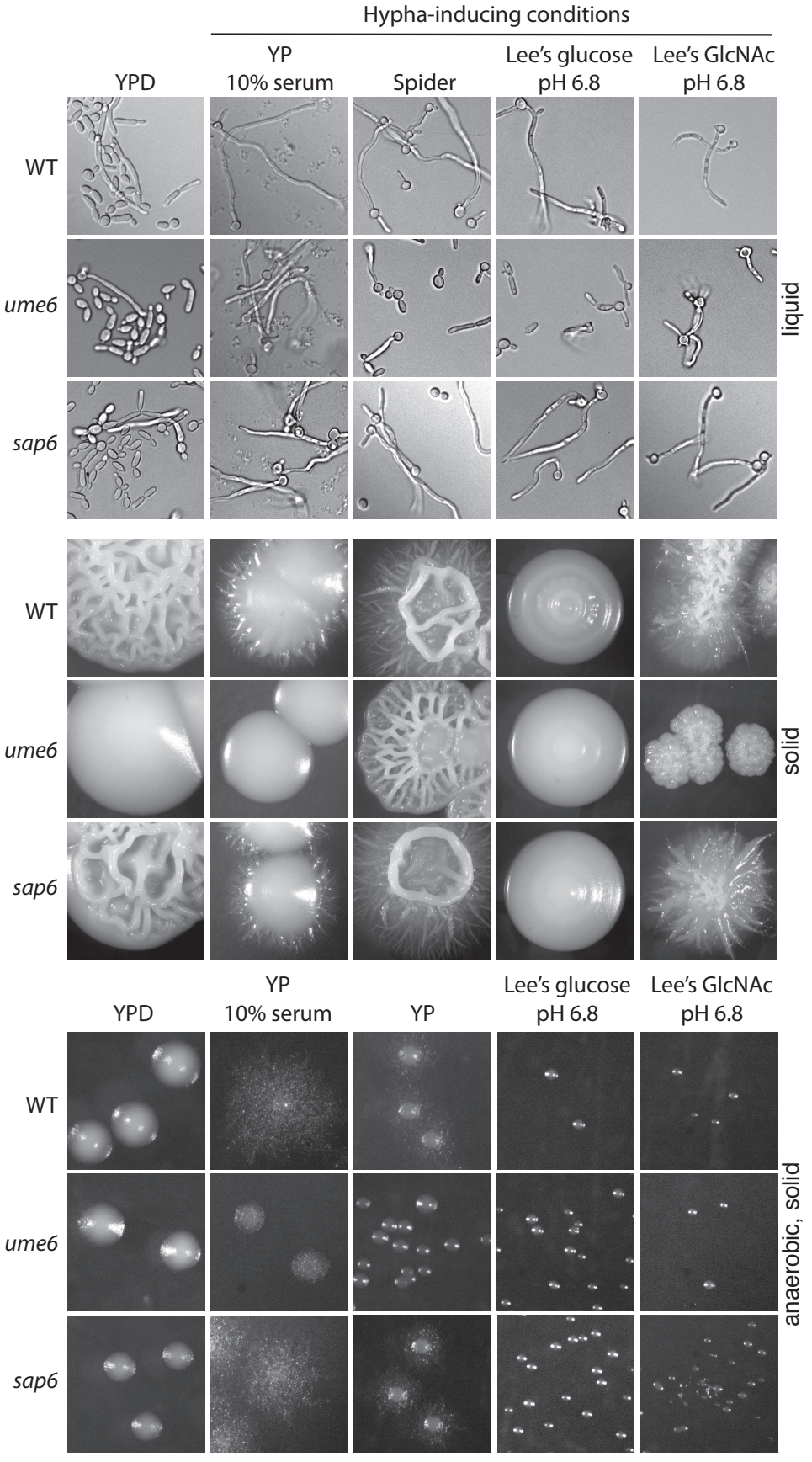

Figure S4. ume6 is defective for filamentation in vitro but sap6 displays normal filamentation, Related to Figure 3. Images depict representative cellular (upper) and colony (lower) morphology of wild-type *C. albicans* (SN250), *ume6* (SN1478), and *sap6* (m886). From saturated overnight cultures, cells for colony imaging were propagated on solid media (YPD/2% agar, YP+10% serum/2% agar, Spider/2% agar, Lee's glucose pH 6.8/2% agar, or Lee's GlcNAc pH 6.8/2% agar) for 5 days at 37°C, and those for imaging of cell morphology were diluted to A600 0.1 and propagated in liquid media (YPD, YP+10% serum, Spider, Lee's glucose pH 6.8, or Lee's GlcNAc pH 6.8) for 4 hours at 37°C. For assessment of colony morphology under anaerobic conditions, plated cells were propagated overnight in air at 37°C prior to being transferred to an airtight container containing a BD Anaerobe Gas Generator Pouch with Indicator for an additional 4 days at 37°C.

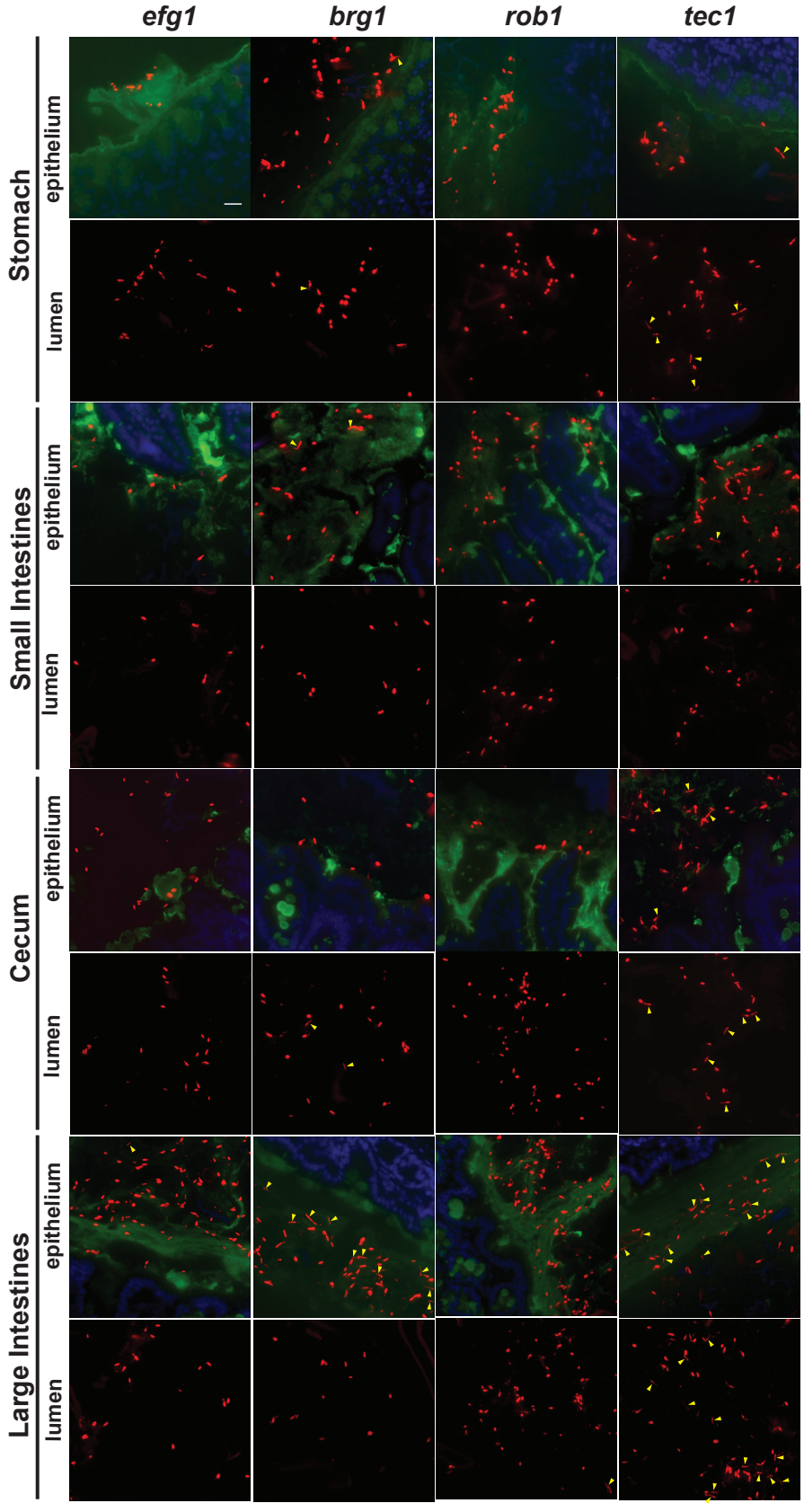

Figure S5. Propagation within the host GI tract suppresses the filamentation defect of *tec1* but not *efg1, brg1*, or *rob1*, Related to Figure 3. Groups of three mice were individually colonized with *efg1* (ySN1011), *rob1* (SN1439), *tec1* (SN1441), or *brg1* (SN1106) for 10 days prior to visualization of *C. albicans* morphology using FISH. Red: *C. albicans* (Cy3-coupled fungal-specific oligonucleotide), green: mucus (FITC-coupled lectin UEA-1 +/- WGA-1), blue: host epithelium (DAPI). Scale bar is 20 μm. Images are representative 18 fields of view assessed for each strain.

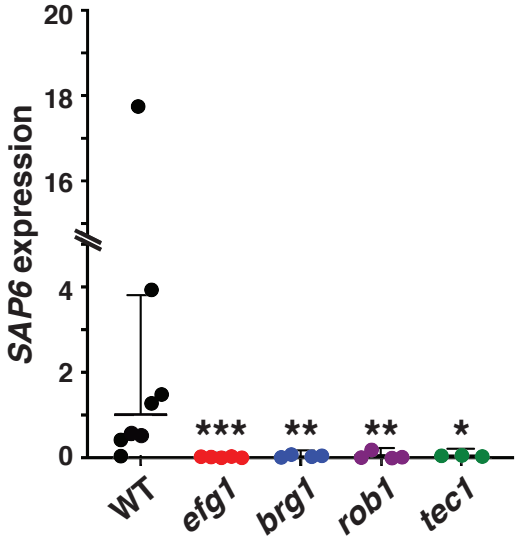

Figure S6. *SAP6* expression is downregulated in commensally propagated *efg1, brg1, rob1*, and *tec1* mutants, Related to Figure 6. Animals were individually colonized with WT (SN250, n=8 mice), *efg1* (SN1011, n=5 mice), *brg1* (SN1106, n=4 mice), *rob1* (SN1439, n=4 mice), or *tec1* (SN1441, n=3 mice). RNA was extracted from the contents of the large intestines (efg1, brg1, rob1, tec1) or feces (WT), and RT-qPCR was performed to quantify SAP6 levels; levels of the housekeeping gene *PMA1* were quantified as an internal control. Three technical replicates were performed for each reaction, and each data point represents the mean result for *SAP6* expression (normalized to *PMA1*) in one animal. Also shown are the mean and standard deviation of the mean for each set of biological replicates. The significance of differences between WT and each mutant was determined using a one-tailed Mann-Whitney U test, *$p<0.05$, **$p<0.01$, ***$p<0.001$.