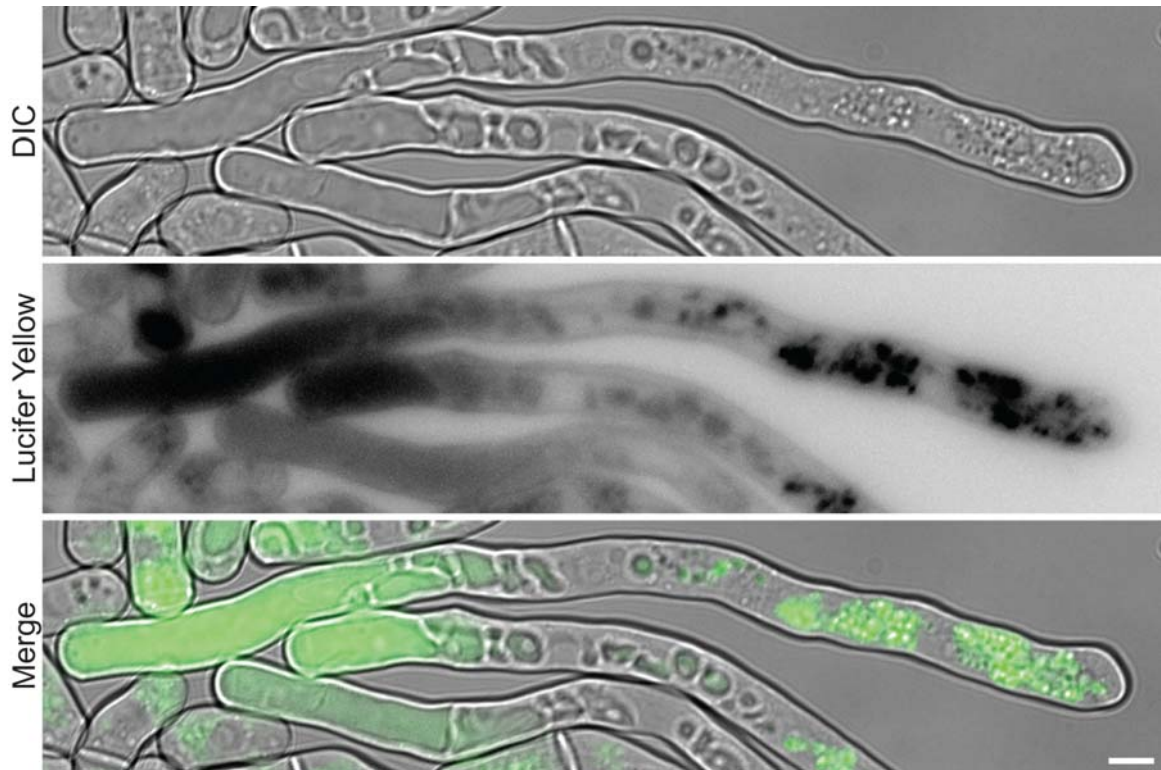


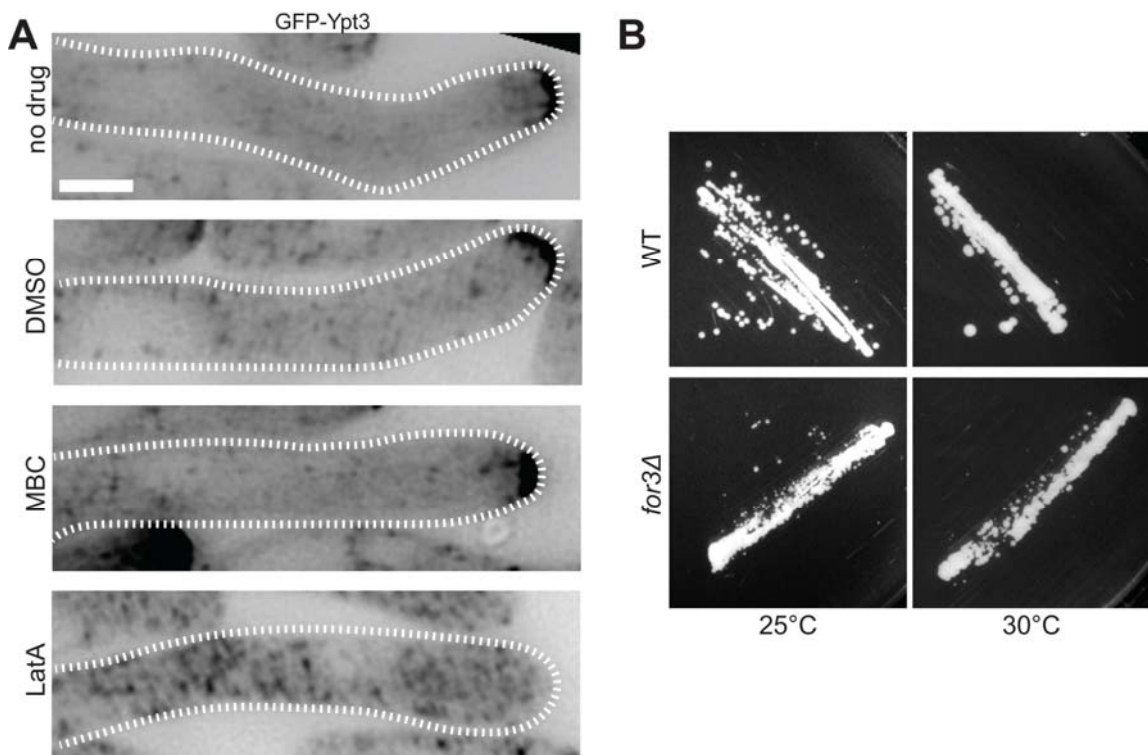
# Supplemental Materials

*Molecular Biology of the Cell*

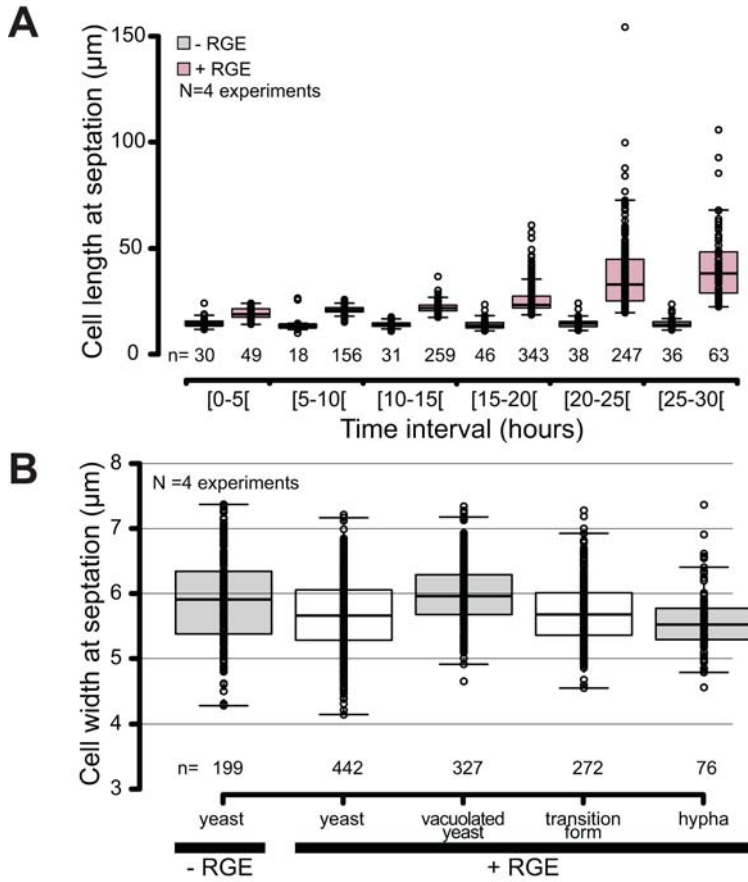
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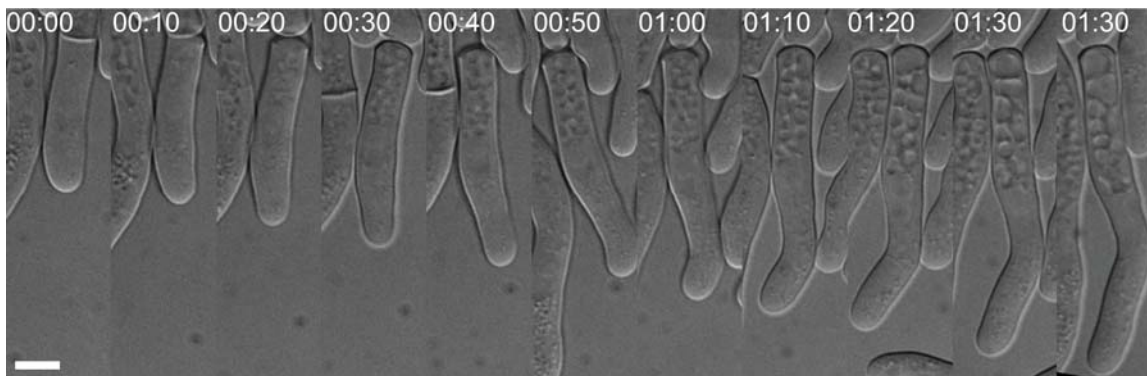
**Figure S1. Vacuole staining with Lucifer Yellow.** Microscopy images showing several hyphae stained with Lucifer Yellow, which accumulates both in the large vacuoles at the back of the cell and in much smaller organelles in the front half. Scale bar 5 $\mu$ m.



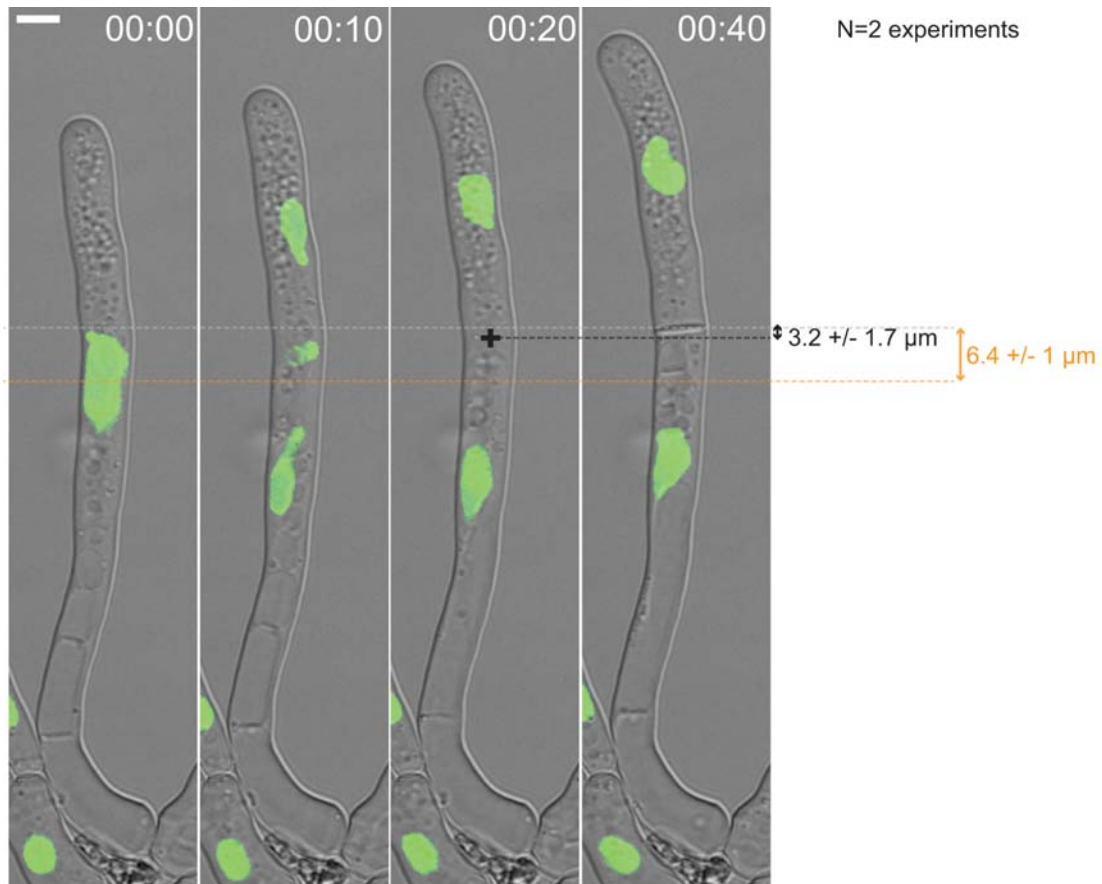
**Figure S2. Importance of formin For3 and F-actin in polarized growth.****A.** Middle plane images of GFP-Ypt3 in hyphae grown in a microfluidics chamber in presence or not of DMSO, MBC or LatA. **B.** *S. japonicus* WT and *for3Δ* strains growing on solid rich media for three days at 25°C or 30°C. Scale bar: 5µm.



**Figure S3. Measurement of cell width and length at septation.****A.** Box plot of septation length of cell populations over 30 hours in microfluidic chambers. Box plots show first and third quartile and median, whiskers extend 1.5 times the interquartile range from the first and third quartile.**B.** Box plot of cell width at septation in the different morphological forms of *S. japonicus*.



**Figure S4. Asymmetric formation and partitioning of vacuoles in the transition form.** DIC microscopy image showing division of a cell growing in RGE highlighting vacuole formation at the back of the front daughter cell. Time in h:min. Scale bar: 5 $\mu$ m.



**Figure S5. Septum position is better predicted by the middle of the anaphase spindle than the position of the pre-divisional nucleus.** Timelapse imaging of a hypha where the positions of the pre-divisional nucleus (orange dotted line), the inferred middle of the anaphase spindle (black cross) and the septum (grey dotted line) are marked. The arrows show the distance between the position of the septum and that of either the pre-divisional nucleus or the middle of the anaphase spindle with their respective average and standard deviations over 20 cells. Time in h:min. Scale bar: 5 $\mu$ m.

**Table S1.** Strains used in this study.

Figure	Name	Genotype	Species	Source
1. A-D, 2. A-E, 3. A, 6. A, E-J, 7. A, S2. A-B, S3	JSM020	<i>h+mat-2017wcs1::natMX6wcs2::kanMX6</i>	<i>S. japonicus</i>	(Okamoto et al., 2013)
4. H, 5. D-E, 7. F, S1. B	JSM023	<i>h+ ade6sj-domE ura4sj-D3</i>	<i>S. japonicus</i>	(Furuya and Niki, 2009)
3. A, 4. A-E, S1. A	JCK060	<i>h+ GFP-ypt3-ura4+ ade6sj-domE</i>	<i>S. japonicus</i>	This study
6. B-D, 7. B-D, S4	JCK090	<i>h+ pAtb2 NLS-GFP-NLS-ura4+ wcs1::natMX6</i>	<i>S. japonicus</i>	This study
5. B-C, 7. E	JCK093	<i>h- pAtb2-mCh-Atb2-ura4+ GFP-NLS-GFP-ura4+</i>	<i>S. japonicus</i>	This study
5. F-H	JSM003	<i>h- GFP-Atb2::ura4 ade6sj-domE</i>	<i>S. japonicus</i>	(Yam et al., 2011)
4. F, 5. A	JCK027	<i>h- Lifeact-GFP-ura4+ mCherry-Atb2-ura4 ade6sj-domE</i>	<i>S. japonicus</i>	This study
3. B, C	JCK024	<i>h+ spa2-GFP-ura4+ ade6sj-domE</i>	<i>S. japonicus</i>	This study
3. B, C	JCK0026	<i>h+ bud6-GFP-ura4+ ade6sj-domE</i>	<i>S. japonicus</i>	This study
3. B, C	JCK033	<i>h+ tea1-GFP-ura4 ade6sj-domE</i>	<i>S. japonicus</i>	This study
3. B, C	JCK056	<i>h+ exo70-GFP-ura4+ ade6sj-domE</i>	<i>S. japonicus</i>	This study
4. G	JCK049	<i>h+ for3::ura4 Lifeact-GFP-ura4+ ade6sj-domE</i>	<i>S. japonicus</i>	This study
4. H, S1. B	JCK031	<i>h+ for3::ura4 ade6sj-domE</i>	<i>S. japonicus</i>	This study
5. D-E	JCK061	<i>h+ tip1::ura4 ade6sj-domE</i>	<i>S. japonicus</i>	This study
7. F	JSM046	<i>h+ mid1::ura4+ ade6sj-domE urasj-D3</i>	<i>S. japonicus</i>	(Gu et al., 2015)
7. F	JCK003	<i>h+ pom1::ura4 ade6sj-domE</i>	<i>S. japonicus</i>	This study
7. G	JSM018	<i>h- pom1-GFP::KanMX6</i>	<i>S. japonicus</i>	This study
3. C	YSM735	<i>h+ bud6-3GFP-kanMX ade6-M216 leu1-32 ura4-D18</i>	<i>S. pombe</i>	(Martin and Chang, 2006)
3. C	YSM1023	<i>h- spa2-GFP ade6- leu1- ura4-</i>	<i>S. pombe</i>	Lab strain
3. C	YSM1253	<i>h+ tea1-GFP-kanMX ade6- leu1- ura4-</i>	<i>S. pombe</i>	(Martin et al., 2005)
3. C	YSM2075	<i>h- exo70-GFP-kanMX ade6-M210 leu1-32 ura4-D18</i>	<i>S. pombe</i>	(Bendezu et al., 2012)
1. C	YSM1371	<i>h+ WT (975) ade6+ leu1+ ura4+ his7+</i>	<i>S. pombe</i>	Lab strain
1. C	YSM2336	<i>h90 WT</i>	<i>S. octosporus</i>	(Rhind et al., 2011)
1. C	W303	<i>α leu2-3 trp1-1 can1-100 ura3-1 ade2-1 his3-11</i>	<i>S. cerevisiae</i>	(Ralser et al., 2012)