Colocalization of potential RNA-transport proteins in *Candida albicans*

Lia Scharnau 2026

Purpose:

Candida albicans is a found in most of the human population but can become pathogenic to individuals with compromised immune systems. *C. albicans* exists in two shapes: a budding yeast form, which appears as an oval-shaped cell, and a hyphal form, which has a long, thin growth protruding from the mother cell. The hyphal form is crucial for *C. albicans* to adhere to host cells and subsequently release enzymes that facilitate invasion and infection of the host (de Groot 2013).

While previous research has confirmed that the transition between budding yeast and hyphal form is key to infection, how mRNA transport plays a role in this mechanism is not fully understood in *C. albicans* (Elson 2009).

To investigate this, we turned to our understanding of the mRNA transport system of *Saccharomyces cerevisiae* (yeast) as the framework for investigating the structure and function of *C. albicans* mRNA transport. In both organisms, the She protein transport system includes the She3 protein and a motor protein; in *C. albicans*, She3 is designated CaShe3 (Elson 2009).

CaShe3 has been confirmed to play an important role in mRNA transport to the hyphal tip. The interacting protein of She3 (Ips1) copurifies with She3, suggesting that Ips1 may assist She3 in mRNA transport in *C. albicans* (Pholcharee 2018). Ips1 has also been identified at the hyphal tip through microscopy, reinforcing its potential role in the mRNA transport complex (Wang
2022).

The goal of the study was to continue characterizing the proteins important to mRNA transport within *C. albicans* by determining whether Ips1 and She3 would localize at the hyphal tip together.

Methods

The McBride Lab already had two *C. albicans* strains: one with Ips1 tagged with a green fluorescent protein (GFP) tag and another with She3-GFP. To visualize both proteins simultaneously, we introduced a She3 gene tagged with mScarlet (which appears red under the microscope) into the Ips1-GFP strain. After constructing the needed strain, we checked that the proteins we were studying were present in the cells through a Western blot. The project concluded with a series of microscopy experiments to determine where these proteins appeared in the cells.

Results and Discussion

An immunoblot confirmed that an mScarlet-tagged She3 protein was expressed in live *C. albicans*. Microscopy also supported these results with the red fluorescence detected in the cells.

After confirming that the mScarlet-tagged She3 protein could be seen in the cells, we tested whether She3 and Ips1 proteins colocalized at the hyphal tip. Microscopy examined strains LSC$ (containing both proteins) and AMC238 (Ips1 only, used as a control for background fluorescence) using different laser channels. In the composite images, the overlap between where these two proteins are found can be seen by the orange and yellow. She3 and Ips1 were observed at the same location at 4.5 and 5.5 hours (Figure 1).

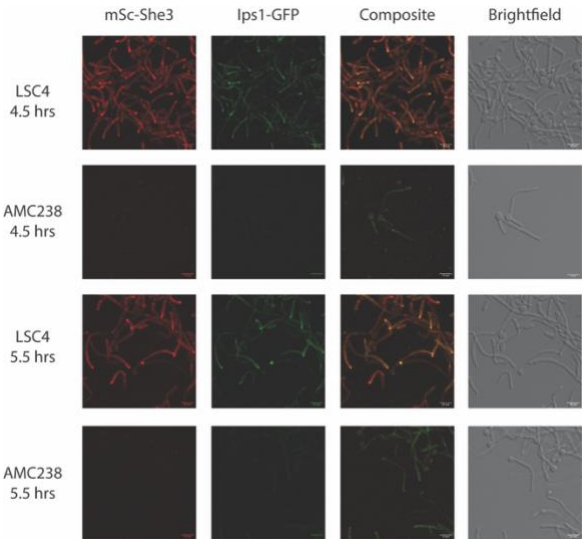

Figure 1. She3 and Ips1 proteins are found at the hyphal tip together.

These preliminary results suggests that the two colocalize at the hyphal tip. Future experiments could investigate how these proteins move throughout the cell to the hyphal tip over time and whether the proteins move together.

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Sources

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