Assessing Ips1 as a potential mRNA transport protein in *Candida albicans*
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Introduction: *Candida albicans* is a commensal fungus and pathogen found in most humans. For immunocompromised patients, *C. albicans* can be life-threatening, causing severe infections (Wisplinghoff et al, 2004). *C. albicans* is polymorphic, forming bud cells and elongated hyphae which increase virulence as they promote host cell adherence and the invasion of host tissues (Sudbery, 2011) . The mechanism by which these cells are infectious has been studied, but much less is known about how proteins and RNA travel to their functional location in the hyphae (Ariyachet et al, 2017).

Intracellular proteins contribute to polarized hyphal cell growth and function by facilitating the transport of mRNA to the growth site (the hyphal tip) (Wang, 2022) (Elson et al, 2009). There mRNAs can undergo translation, a process by which ribosomes in the cell decode mRNA to produce specific proteins. Researching key protein players in these mRNA transport complexes is important to understanding hyphal formation.

Past studies on *Saccharomyces cerevisiae*, or baker's yeast, have identified crucial mRNA transport proteins with potential *C. albicans* homologs. She3 and Myo2 are proteins connected to mRNA localization in the *C. albicans* hyphal tip (Elson et al, 2009). However, other *S. cerevisiae* mRNA transport proteins were not found in *C. albicans*, indicating that this yeast has different proteins contributing to hyphal development within the She3-mediated transport mechanism (Wang, 2022). My research will expand current knowledge of this transport complex by assessing another possible protein player, referred to as the interacting protein of She3, or Ips1. Ips1 and Myo2 are the two predominant proteins that copurify with She3 suggesting that Ips1 is likely a part of the same complex (Pholcharee, 2018). My proposed research project assessed if She3 and Ips1 colocalize in the hyphal tip. Examining this relationship helped us better understand the structure and function of this She-3-mediated mRNA transport, and how it contributes to hyphal growth and *C. albicans* host cell damage.

Methodology: To answer this question we tagged the Ips1 and She3 proteins with different colored fluorescent tags that could be visualized using a confocal miscroscope. We grew our *C. albicans* strains with the inserted flourescent tags and induced hyphal growth before looking at the hyphae under the confocal microscope at different time points. Finally we used image analysis to determine if the proteins traveled to the hyphal tip and localized together.

Results and discussion: Over the past eight weeks we successfully tagged and visualized She3 and Ips1 under the confocal microscope. We tagged She3 with mScarlet, a red fluorescent protein and confirmed that the protein was correctly tagged and expressed using a Western blot. Confocal microscopy indicated that mScarlet tagged She3 was localized at the hyphal tip at 4.5 and 5.5 hour time points in two of our synthesized *C. albicans* strains (Fig. 1). The same strains also expressed GFP (green fluorescent protein) tagged Ips1 at the hyphal tip at these time points (Fig. 1). These results indicate that Ips1 and She3 may be a part of the same She complex localizing at the hyphal tip, although repeat experiments with more time points need to be done to get significant data. The mScarlet-She3 signal was brighter than the GFP-Ips1 signal indicating a higher concentration of She3 at the hyphal tip. These differences in concentration may be the result of overexpression of She3 in the strain used. Future experiments in the fall will focus on reducing the overexpression of She3 in new *C. albicans* strains and repeating these experiments to futher examine if this factor affects Ips1 and She3 colocalization. Futhermore, we will determine if the presence of Ips1 is required for She3 localization to the hyphal tip and vice versa to assess if these proteins are dependent on eachother to function correctly.

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