

28th May 2018

FINAL REPORT

Tracking protein biogenesis in fungal networks using chimeric mRNA-protein reporters.

My research visit to St. Andrews from 16th-20th May 2018 has been a considerable success for several reasons.

First of all, the newly developed chimeric mRNA-protein live-cell imaging reporter works with 80 % confidence as the imaging data acquired at the University of St. Andrews shows. Due to the unpublished nature of the technique I cannot disclose any details here, but can say that we found meaningful changes in subcellular localization of the mRNA-protein reporter in the presence of its selected target mRNA and labelled protein, respectively (Fig.1).

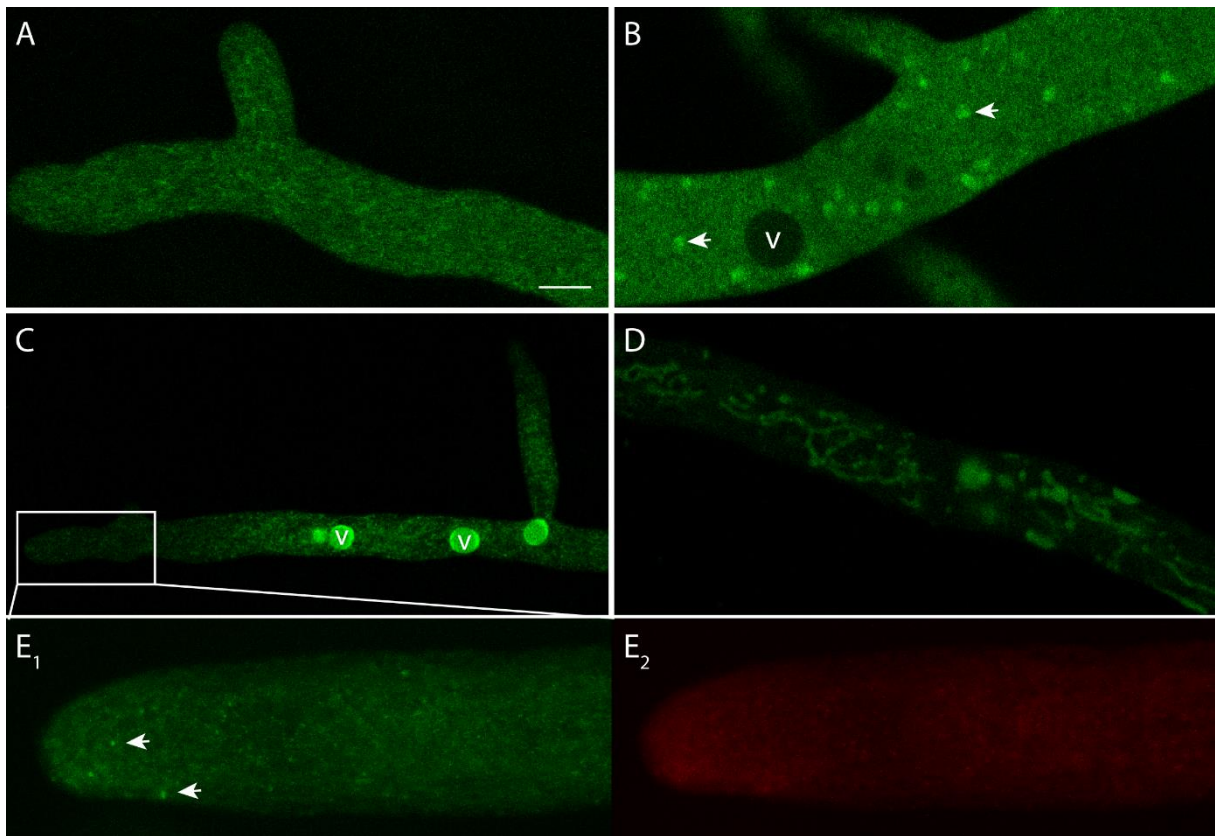


Figure 1. Changes in the subcellular localization of the chimeric mRNA-protein reporter when presented with its target mRNA indicate its functionality. (A) When expressed alone, i.e. in the absence of its target mRNA, the reporter localises diffusely throughout the cytoplasm in peripheral hyphal compartments. Scale bar, 5 μ m. **(B)** In subperipheral hyphal compartments, nuclei become positively labelled (arrowheads), whereas vacuoles (V) become negatively labelled. **(C)** In case a target mRNA is co-expressed, these default subcellular localisations change significantly: the diffuse cytoplasmic localisation diminishes and small mRNA granules appear (arrowheads in E₁) that

concentrate in the tip apex and co-localise with the accumulation of the target protein in an apical cloud (red signal in E₂). Furthermore, vacuoles become strongly positively stained. (D) In subperipheral hyphal compartments the target mRNA-reporter complex localises to tubular structures; presumably the ER which is a known biogenesis site of the selected target protein.

Further controls are now underway to confirm these initial, very promising results and to improve the detection of the target protein. The comprehensive data analyses and personal discussion with my collaboration partner Dr. Jens Tilsner provided clear strategies for improvement and modification of suitable controls. I am confident that within the next couple of months the envisioned reporter system will be fully operational, and will become part of a research grant proposal to investigate protein biogenesis from transcription to translation inside fungal networks.

Secondly, I had the opportunity to present my recent research to an expert audience and received very good feedback, including some ideas how to develop this topic further and expand it from fungus-fungus interactions to fungus-fungus-plant interactions, and thus come much closer to the real-life situation in the field.

This, thirdly, led to a new collaboration with Dr. Lionel Dupuy from The James Hutton Institute in Dundee who provided me with a sample of novel artificial soil that allows the co-cultivation of plant seedlings and filamentous fungi in a micro-format. This technique will likely make mycoparasite-host fungus-plant interactions accessible to live-cell imaging analyses for us for the first time, and thus promises to be a big step forward in our research.

I am very grateful to the **BritInn Network** not only for providing the financial support for this trip, but also – and even more crucially – by doing so, giving me the reason and motivation for initiating this project. An idea Dr. Tilsner and I had in our minds for the last six years finally is becoming reality and we are planning to apply for BritInn funding again for a reciprocal visit of Dr. Tilsner to Innsbruck once the project is further developed.

Thank you very much.



Dr. Alexander Lichius