# Methods

## Image Processing

Each set of collagen gel and Lifeact images were processed individually to identify the lifetimes of the invadopodia actin puncta. The bright puncta in each Lifeact image were indentified using a high pass filter and a threshold determined by finding the high-pass filtered pixels five standard deviations from the mean. The identified puncta regions were tracked and the lifetimes of each puncta found. To determine which puncta constituted invadopodia, the local difference in the collagen gel intensities around each puncta was found. The local difference was quantified by finding the difference between the average collagen gel intensity immediately below the puncta and the average gel intensity within a five pixel border around each puncta. If the mean local gel difference value was below zero with 95% confidence (T-test, 2 tailed) for the life of a puncta, that puncta was marked as an invadopodia.