## The Annotation of Novel Datasets for the Training of Cellori (Cell Origin) Spots, a Deep Learning Algorithm for RNA FISH Spot Detection

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Developments in microscopy imaging have allowed for in depth study of biological systems at a level never previously reached. One such method is RNA fluorescence in situ hybridization (FISH), which is commonly used to visualize the spatial and temporal expression of specific genes within cells. Analysis of large datasets can be cumbersome and time consuming, especially when assessing fluorescent localization manually. Attempts to automate this process through formulas and filters include the Laplacian of Gaussian (LoG) operator made publically available by programs such as TrackMate and big-fish. Although LoG increases annotation efficiency, its performance and efficiency are limited by a researcher's ability to set an accurate threshold.

Developments in deep learning have led to automatic algorithms for RNA FISH spot detection. However, current deep learning algorithms fail to perform on datasets without uniform spots and low background noise. In this research, I used NimbusImage to annotate RNA FISH images to develop a dataset to train Cellori (cell origin) Spots, a deep learning algorithm developed by Raj et al. I demonstrated that, using my dataset, Cellori Spots can outperform LoG and current deep learning approaches to RNA FISH spot detection on images with high background variance and irregular spots. It can be concluded that the diversity of training images is reflected in the accuracy of Cellori Spots. Maintaining accuracy on more difficult images is vital for spot detection as it allows researchers to draw definite conclusions from RNA FISH images without high quality staining.