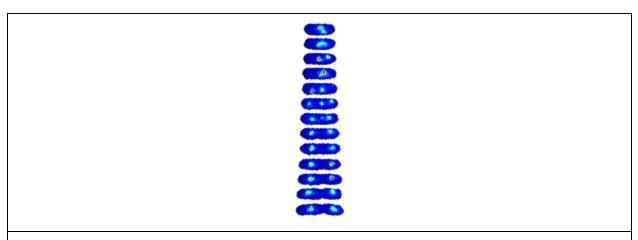
## **Protocol: Single-Cell Localization Dynamics.**

**Summary:** This protocol describes the generation of single-cell image towers.



**Figure 1:** Image tower showing the single-cell localization of SeqA-gfp in false color. Th localization in a newborn cell is shown in the first frame. The final frame shows the cell jus prior to division.

- 1. Segment and link data to generate cell stacks. (Protocol)
- 2. Background subtract fluorescence images by subtracting the background level defined as the mean fluorescence, throughout the frame, in regions outside of cells.
- 3. Rotate cell image and mask in each frame to align the major axis of the mask with the x-axis and placing the old pole (the new pole is created in the last division) on the left hand side.
- 4. Center each image on the y axis
- 5. Stack masked fluorescence images of cells with frame number increasing downwards.
- 6. Normalize false color fluorescence to the brightest pixel in the stack.