Live Particle Tracking

Introduction

The Prime 95B™ Scientific CMOS camera is great for imaging situations requiring both extreme sensitivity as well as a high acquisition rate, making it ideal for techniques such as super-resolution localization microscopy and single-molecule tracking. The Prime camera platform incorporates a powerful FPGA and performs real-time image processing optimized for specific applications to provide more useful experimental data.

Live Particle Tracking is the latest addition to the real-time processing capabilities of the Prime 95B camera, and is designed to increase the efficacy of single molecule particle tracking experiments.

How It Works

Live Particle Tracking, as its name suggests, is designed to identify individual single molecule particles and track them across the field of view by adapting a published algorithm tuned for two-dimensional tracking. The algorithm processes the images in the following order:

1. Image detection
2. Image restoration
3. Estimation and refinement of point locations
4. Non-particle discrimination
5. Trajectory linking
6. Image output with tracking meta-data

The image detection step requires the camera to determine only the dynamic portions of the image and disqualify anything static from detection. The data is then run through a restoration step which behaves as a bandpass filter, reducing both the high frequency and low-frequency noise, and allows the correction of any noise variation on a pixel-to-pixel basis as well as any background intensity modulations due to uneven illumination.

The points are then processed to determine the local-maxima within the radius of the evaluation kernel, and go through a refinement process to ensure a high efficiency in particle detection based on a threshold to reduce the susceptibility to false

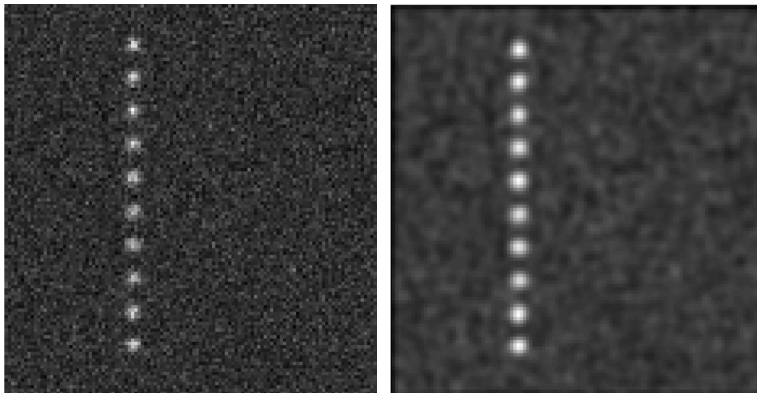

Figure 1. The input image of simulated single-particle data and the output of the image-restoration step to reduce image noise and pixel-to-pixel variation.

positives. Any remaining artifacts are filtered out during the non-particle discrimination step, aimed at hot pixels and cosmic events.

Finally, the particles are tracked and linked through the acquired frame stack. The metadata included with all images is updated to include the particle data within each frame, providing particle IDs as well as the ability to display particle path traces as well as boxes to outline each detected particle.

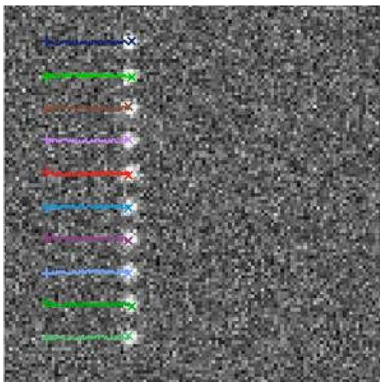

Figure 2: The live particle tracking feature adds meta-data to each image providing the movement information of each particle.

Conclusion

The Live Particle Tracking feature of the Prime 95B Scientific CMOS camera provides the ability to evaluate the behaviour of single-molecules during the acquisition, ensuring that expected behaviour can be determined early in the experimental process. By reducing the number of experiments that do not meet the required or expected rates of movement by the particles, the efficacy of experiments can be increased significantly.

References

I.F. Sbalzarini, P. Koumoutsakos. 2005. Feature point tracking and trajectory analysis for video imaging in cell biology