How to significantly improve drift / stability performance in your existing SMLM setup

Michael Schwertner, Arnold Kamp, Martijn van Nugteren, Hildo Vader
Linkam Scientific Instruments, 8 Epsom Downs, Tadworth, Surrey, KT20 5LR, United Kingdom. Email: MichaelSchwertner@linkam.co.uk

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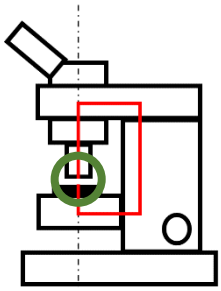
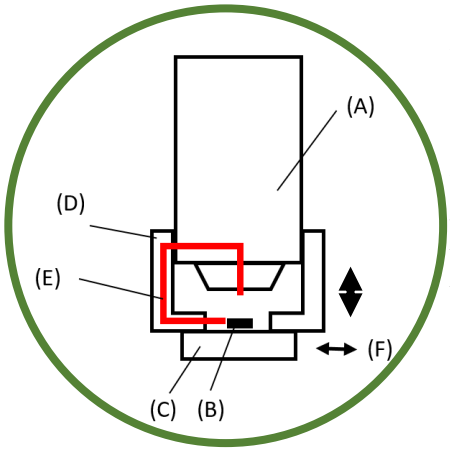
Summary We demonstrate a novel design enabling high-stability imaging largely independent of temperature drift by the host microscope frame, improving data acquisition for SMLM setups. This is achieved by miniaturizing the mechanical paths contributing to sample drift and mounting the sample directly to the tip of the objective lens.

To start we define the mechanical loop as the shortest path along the setup from the sample to the tip of the objective lens. The figure on the right shows a typical upright microscope with the mechanical loop highlighted in red. Here the mechanical loop is in the order of 300 mm. All components and materials along the mechanical loop are susceptible to temperature drift, may flex against each other or pick up vibrations. When a stability in the order of tens of nanometers or better is required, it is difficult to keep such a relatively large system stable over time. Therefore a good strategy is to reduce drift is to reduce the size of the mechanical loop.





As shown in the figure on the left we can mount the sample directly to the tip of the objective lens (A), where the cosmetic cover of the lens has been removed for direct interfacing to the lens barrel. The sample (B) on the sample holder (C) is clipped magnetically to the focus mount (D), where coarse focus is available by means of a fine thread between (D) and the lens barrel of (A) or more convenient by a piezo actuator for the same task. Again the mechanical loop (E) is marked in red. Overall the mechanical loop of the system has been reduced down to roughly 25 mm while the Z-components of the mechanical loop relevant for focus drift is reduced down below 10 mm. The sample holder (C) is magnetically coupled and acts as a magnetic stick-slip mechanism for XY sample navigation (F) by means of a sample manipulator (not shown). The lens sub-assembly* with sample is then mounted to a conventional microscope frame (upright or inverted), upgrading drift performance. Outstanding low-drift performance can be achieved without active tracking designs. Further reduced sensitivity to vibrations is achieved by decoupling the XY system during imaging, engaging it only for sample navigation. In many cases optical tables or active vibration insulation is not required anymore due to the intrinsic properties of the design. Quantitative data for the XY drift components will also be shared in this presentation.

*patent pending