

Supplemental Information

Title: Method for high frequency tracking and sub-nm sample stabilization in single molecule fluorescence microscopy

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Supplementary Methods

Tracking scheme: For tracking we need to use the real time QAPD values to find the corresponding position values stored in the FPGA memory. We convert real-time QAPD values via the equation:

$$Index = (LR - LRmin) \times LRslope + (BT - BTmin) \times BTslope \times n$$

where LR and BT are the real time difference channel values, $LRmin$ and $BTmin$ are the lowest values from the difference channel maps, n is the number of contours per channel, and $LRslope$ is calculated by:

$$LRslope = \frac{n - 1}{LRmax - LRmin}$$

where $LRmax$ is the highest value from the horizontal difference channel map. $BTslope$ is calculated similarly to $LRslope$. This index holds a 32 bit number which is split to obtain the 16 bit X and Y position.