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Three-dimensional flow measurements with a four-focus microscope JAMES A. GERMANN, BRIAN K. CANFIELD, JASON K. KING, ALEXANDER TEREKHOV, LLOYD M. DAVIS, Center for Laser Applications University of Tennessee Space Institute — The measurement of a one-dimensional flow using a confocal fluorescence microscope with two excitation volumes has been well documented. This technique can be extended to measure flow in all three dimensions simultaneously through a four-focus, two-photon microscope. To this end, an apparatus has been constructed in which the beam from a modelocked Ti-Sapphire laser is passed through a double interferometer configuration to create four displaced focal volumes. Fluorescence is gathered onto a single photon avalanche diode and time-gated by a TimeHarp 200 timer card. Calibration of one-dimensional flow through a square bore capillary has been performed. Flow of adjustable speed and direction in three dimensions is created using a cross-channel microfluidic device. To evaluate flow measurements, Monte Carlo simulations of fluorescence cross-correlation spectroscopy between the four foci were conducted and a LabView program was created to discern the flow parameters from the 16 cross-correlation functions. For simplicity, the model for the correlation functions assumes each focal volume is a three-dimensional Gaussian, but a Gaussian-Lorentzian model may improve fitting.

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