

Imaging-based cross-disciplinary research at IIMCB

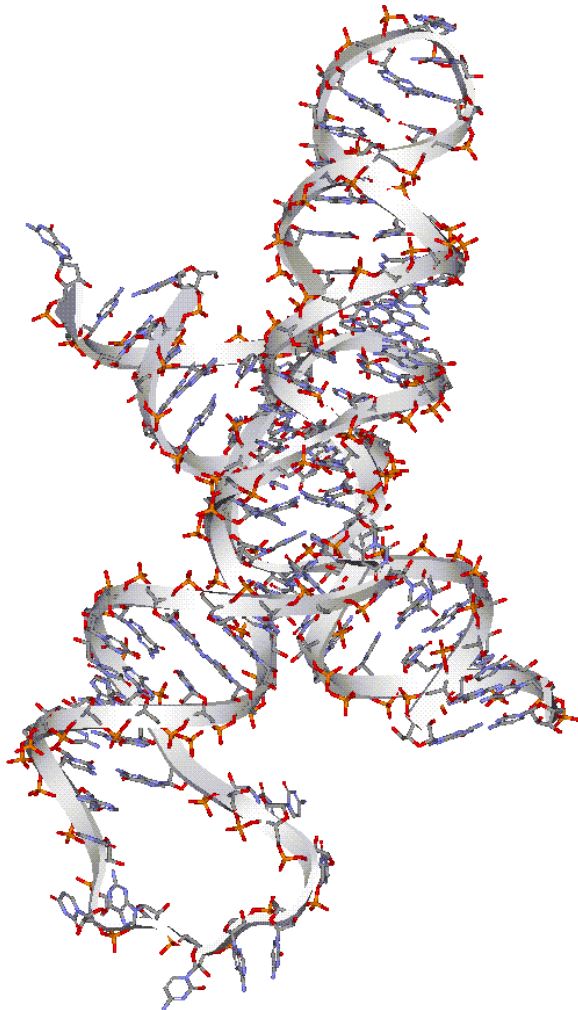
Tomasz Sołtysiński

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in Warsaw**

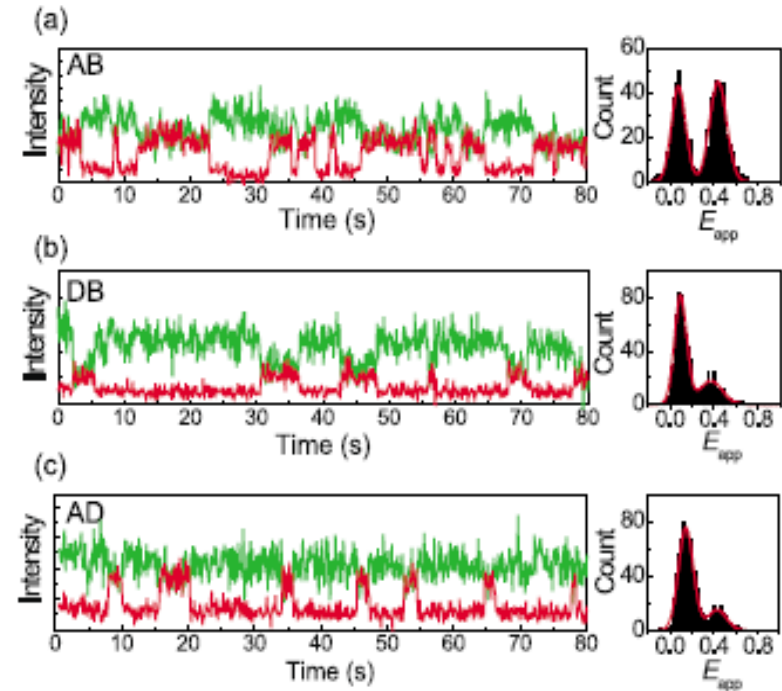


Exemplary problem

RNA 4-way junction of the Hairpin ribozyme



Rupert *et al*, Science, 298, (2002), 1421



smFRET traces show three populations

Hohng *et al*, J.Mol.Biol., 336, (2004), 69

Kalinin, Soltysinski, et al. 2011

Open questions

Number of species in equilibrium

Solution of structures including less populated states (no X-Ray data)

Interconversion dynamics

Solution: experiments in an open environment

Open optoelectronic setups



Components to collect:

Lasers: continuous waves, pulse: femto seconds

Beam steering: prisms, mirrors, (acoustooptical) filters, splitters, Pockel's cell, confocal scanners, resonant scanners, optical fibers, acoustooptical deflectors, objectives

Detectors: Photomultiplier tubes, avalanche photodiodes, fast CCD, wide field CCD, single photons counting cards,

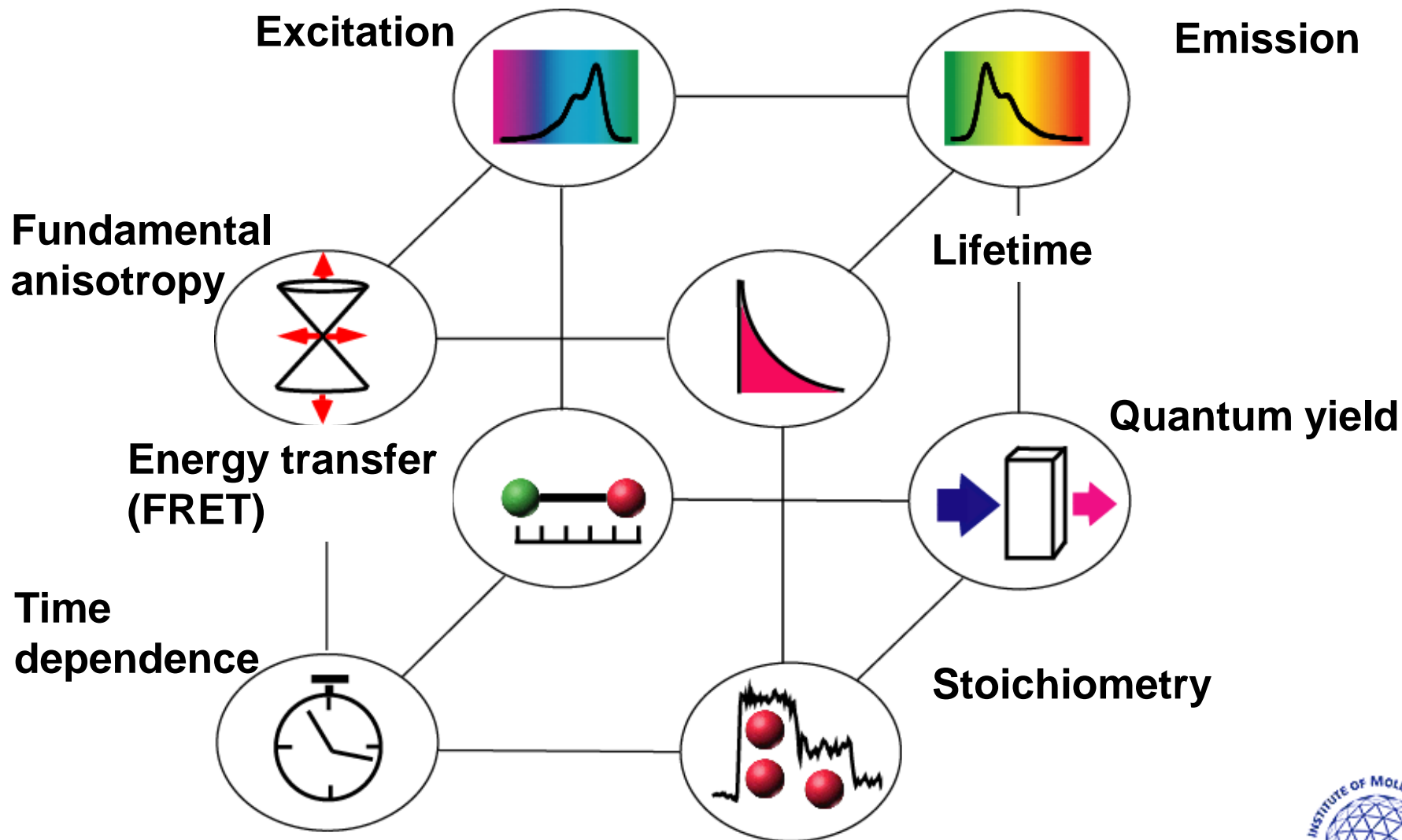
Drivers: PCs: multi CPU/GPU with solid state hard drives, piezoelectronic drivers, FPGA

Mechanical equipment: optical tables, mounting tools, safety covers, piezomotors/positioners

Software: LabView/Matlab/Python



What can we gain? 8 Dimensions of Fluorescence: MFD Multiparametric fluorescence detection



Research problems to address

Coupling of single molecule experiments to theoretical modeling

Proteins/nucleic acids folding and unfolding

Examination of properties of biological molecules

Interaction of biological molecules

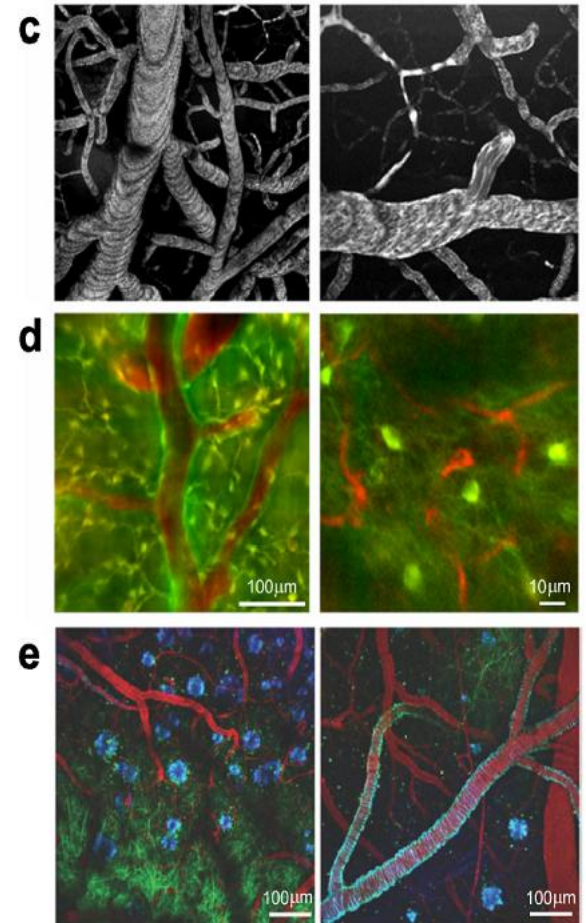
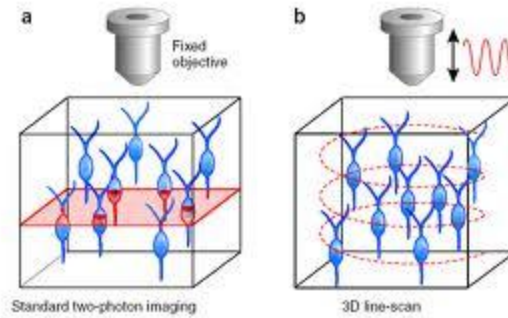
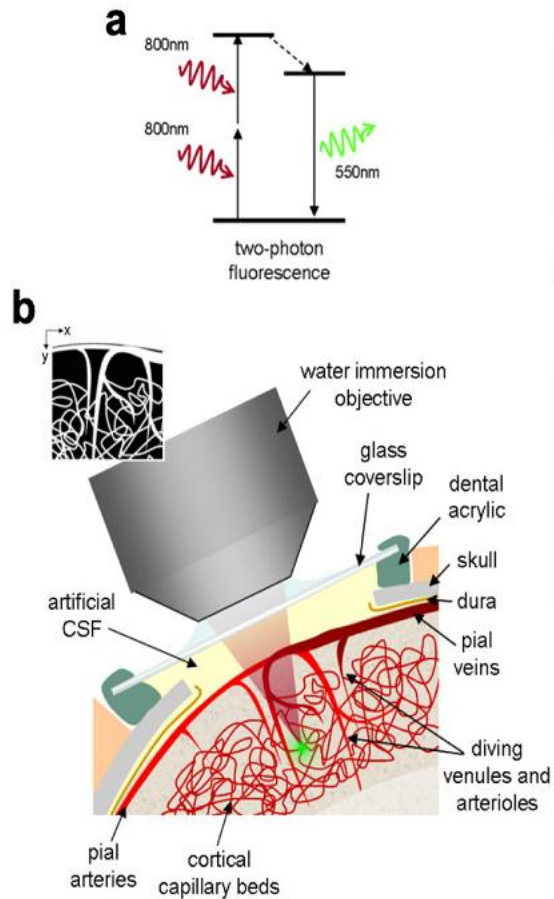
Determination of structure and function of biological molecules

Monitoring target molecules in a cell natural environment

Monitoring of cell functions



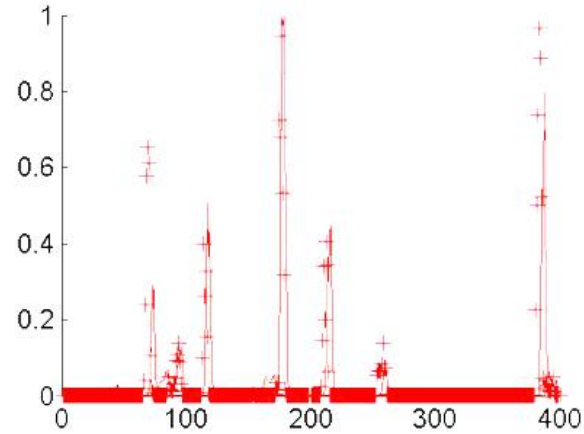
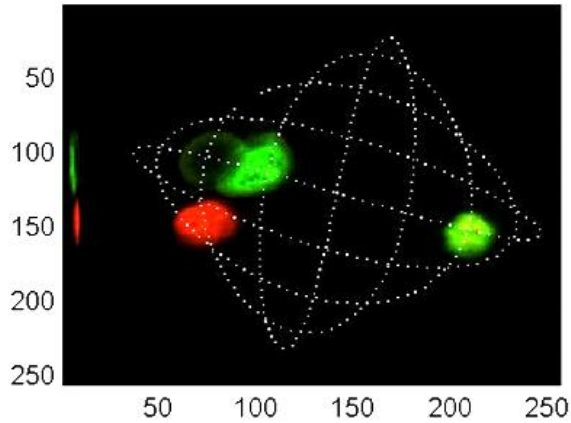
MFD *in vivo*? Multiphoton imaging



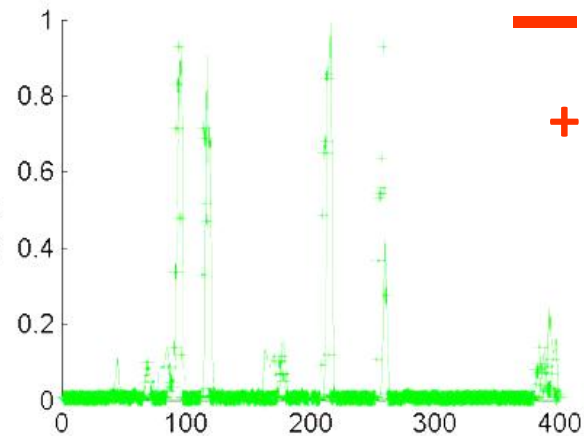
Kherlopian *et al.* *BMC Systems Biology* 2008 2:74



Unique trajectories



Red channel



Solid line: data from frame image

+ data from trajectory

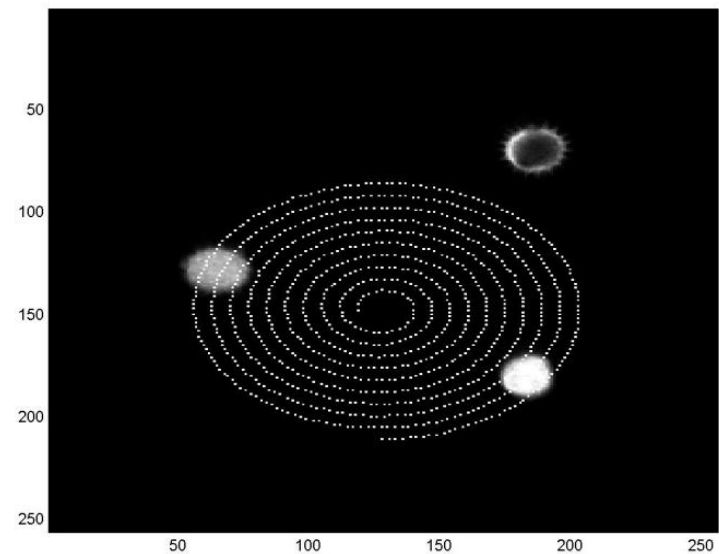
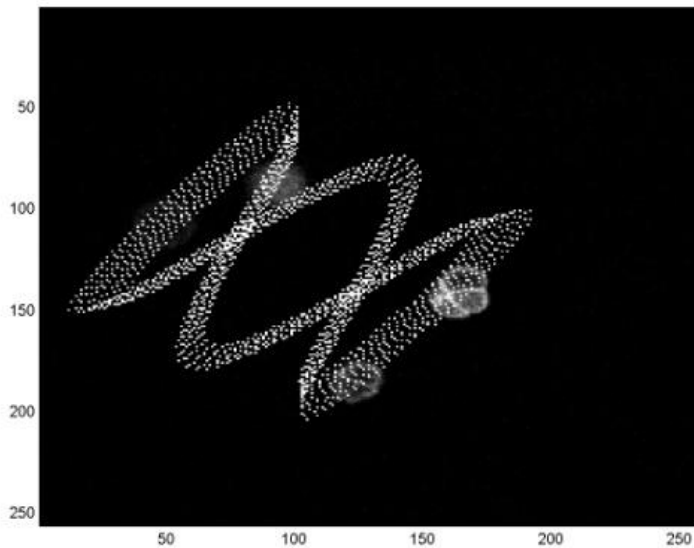
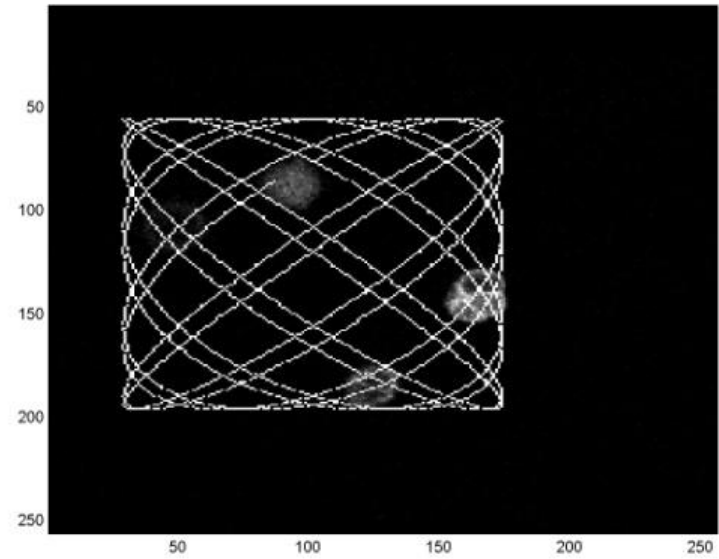
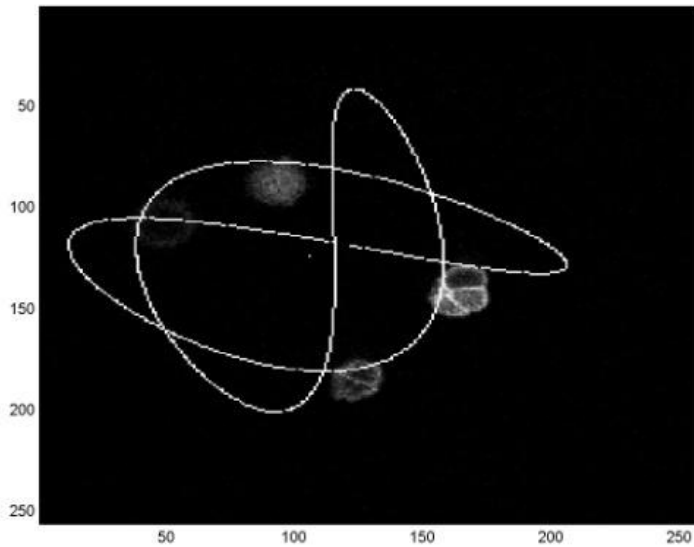
Green channel

Scanning frequency 1 kHz

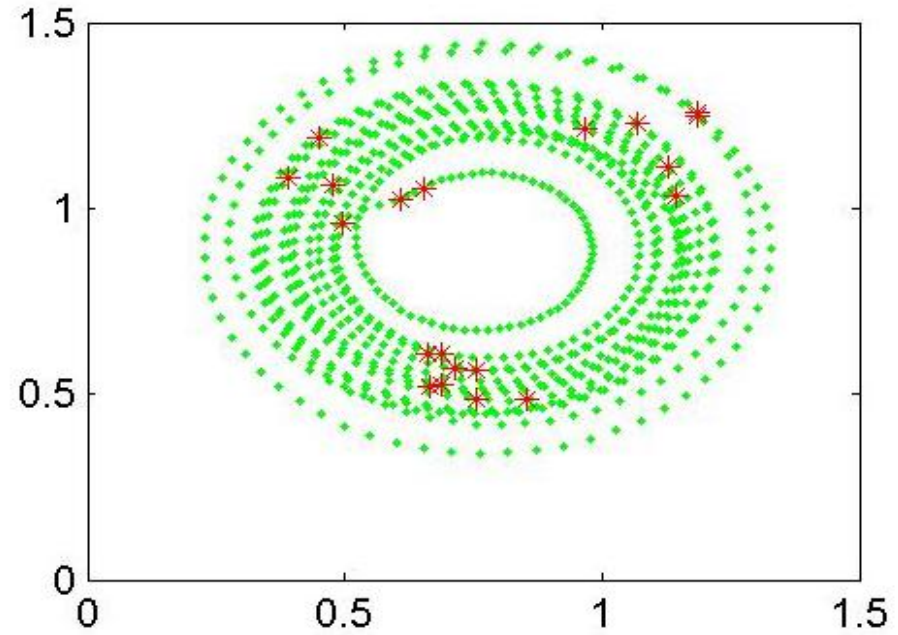
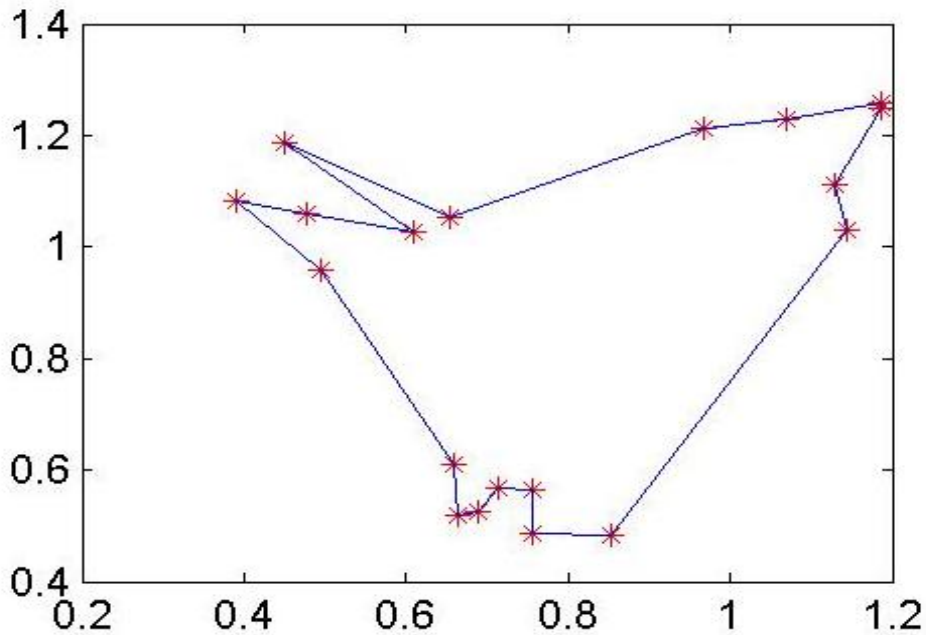
Sołtysiński, in prep.



Novel algorithms for rapid scanning



Low inertial scan modes for rapid scanning



Sołtyśński, Proceedings of WMIC 2010



Research problems to adress (*in vivo*)

Tracing the hemodynamic response to a stimulus

Calcium-based imaging in neocortex

Blood flow monitoring at capillary level

Glial and neuronal networks coupling

Disfunction of capillaries in Alzheimer's disease

Dendrites and synapses growth

Gene expression in neuronal cells and optogenetics

Trafficking of tumor cells



**Thank you for
your attention!**



Early microscopes