Use of calcium indicator proteins in spike counting mode

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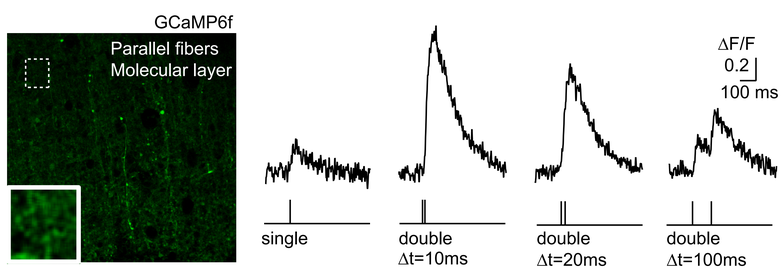
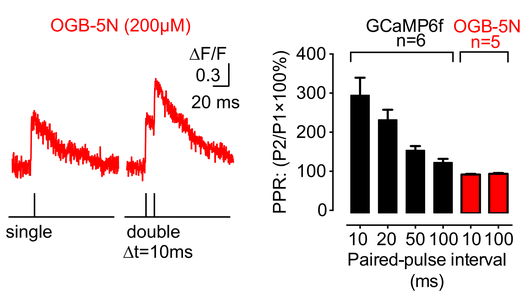
THE CHALLENGE: SLUGGISH RESPONSES

Although calcium sensor proteins such as GCaMP have emerged as a powerful technology for monitoring neural circuitry, they give slow signals when expressed in cell bodies. This sluggishness arises from two factors: intramolecular kinetic bottlenecks, and slow calcium dynamics in large compartments.

IN BRAIN SLICES, AXONAL BOUTONS MAKE LARGE SIGNALS



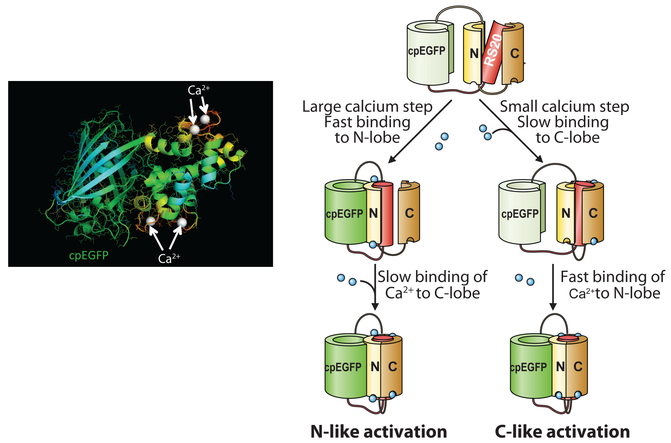
GCaMP6f transients last for 0.3 second...



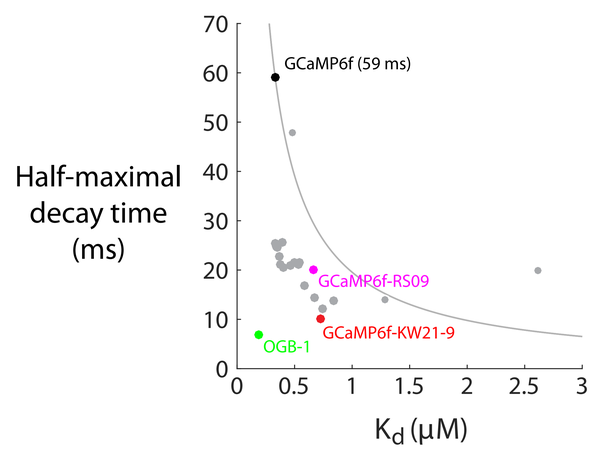
...but true calcium signals decay in 50 ms.

FAST-GCAMPS RELIEVE KINETIC BOTTLENECKS

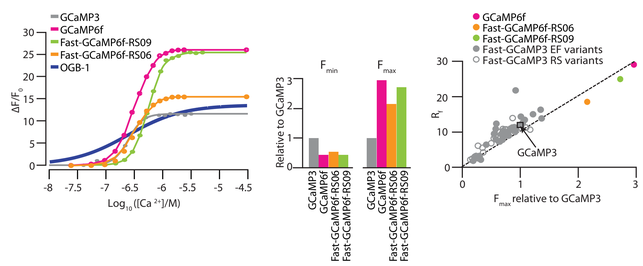
Kinetic scheme



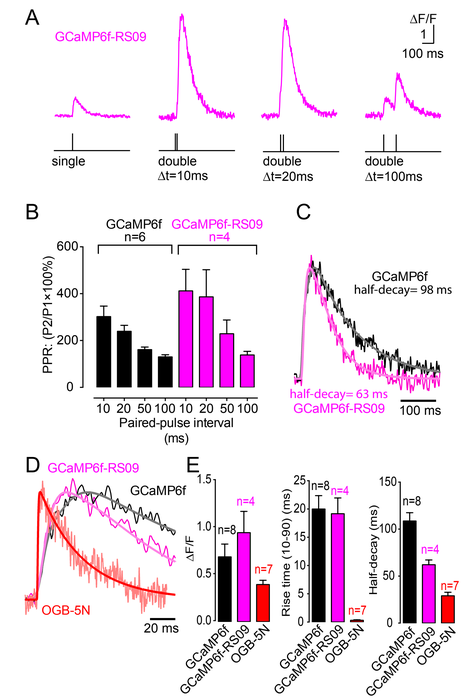
In stopped-flow fluorimetry, off-responses approach the unbinding limit



Fast-GCaMPs retain their brightness

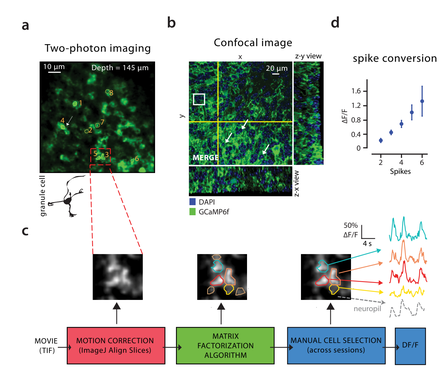


PROOF OF PRINCIPLE



We seek to:
1) Accelerate Fast-GCaMPs further,
2) Target them to axon terminals,
3) Reduce multispike nonlinearity, and
4) Resolve spikes in vivo at ~50 Hz.

NONNEGATIVE MATRIX FACTORIZATION METHODS CAN RESOLVE SINGLE-NEURON SIGNALS



Collaboration with Andrea Giovannucci, Liam Paninski, and Eftychios Pnevmatikakis