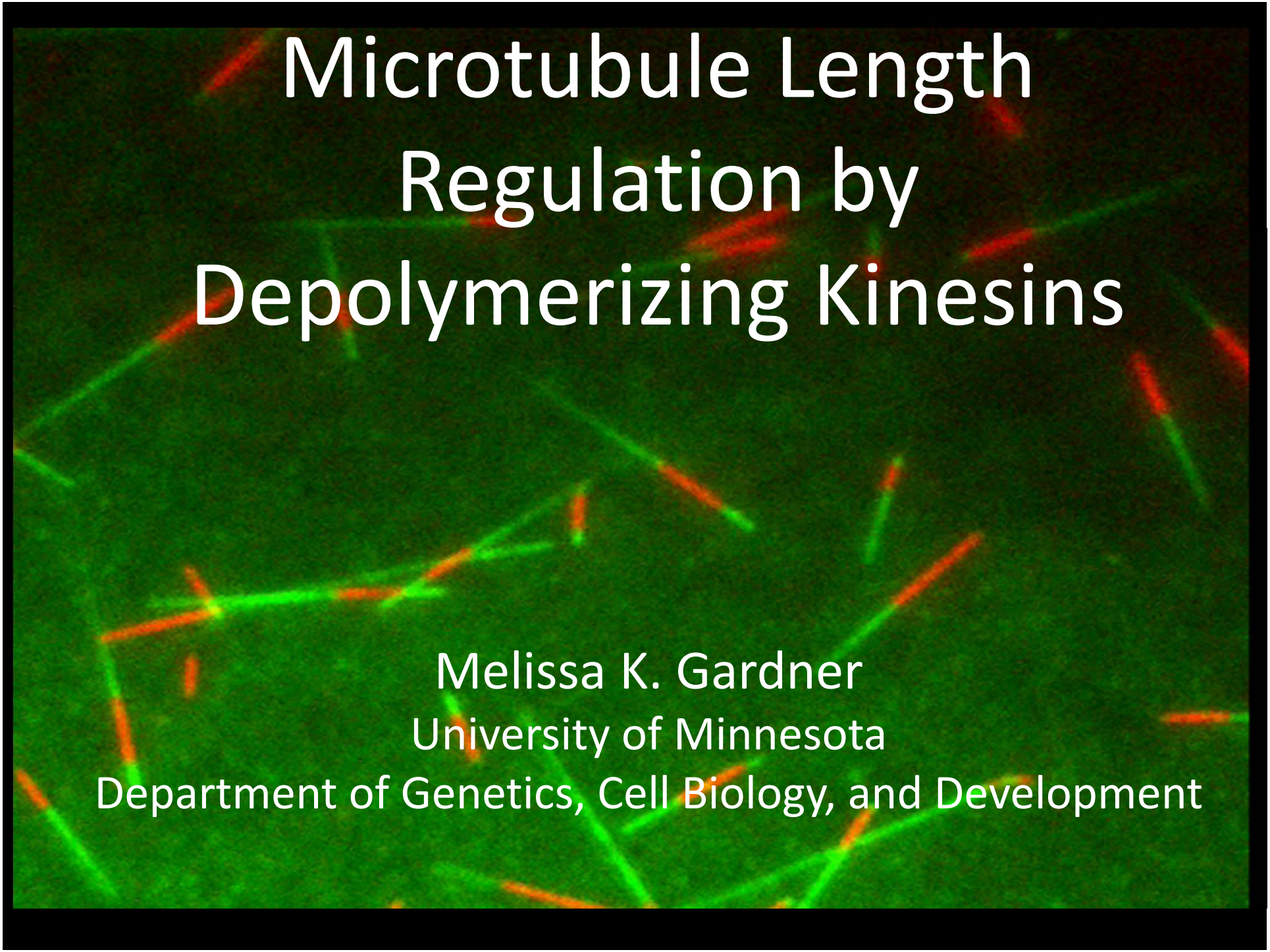
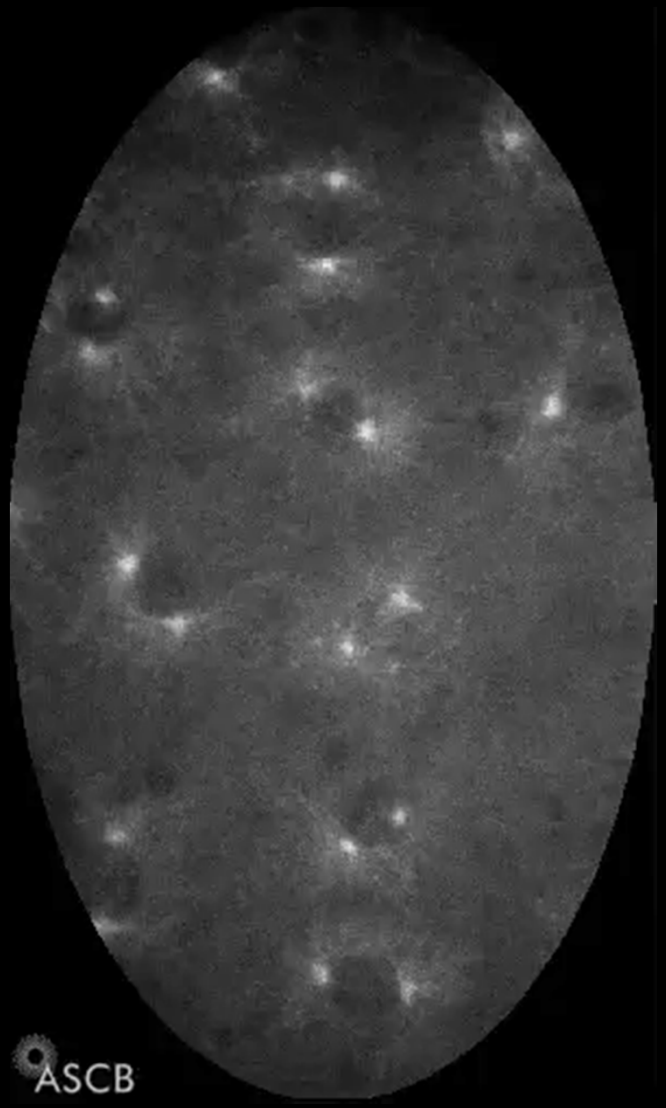


Microtubule Length Regulation by Depolymerizing Kinesins

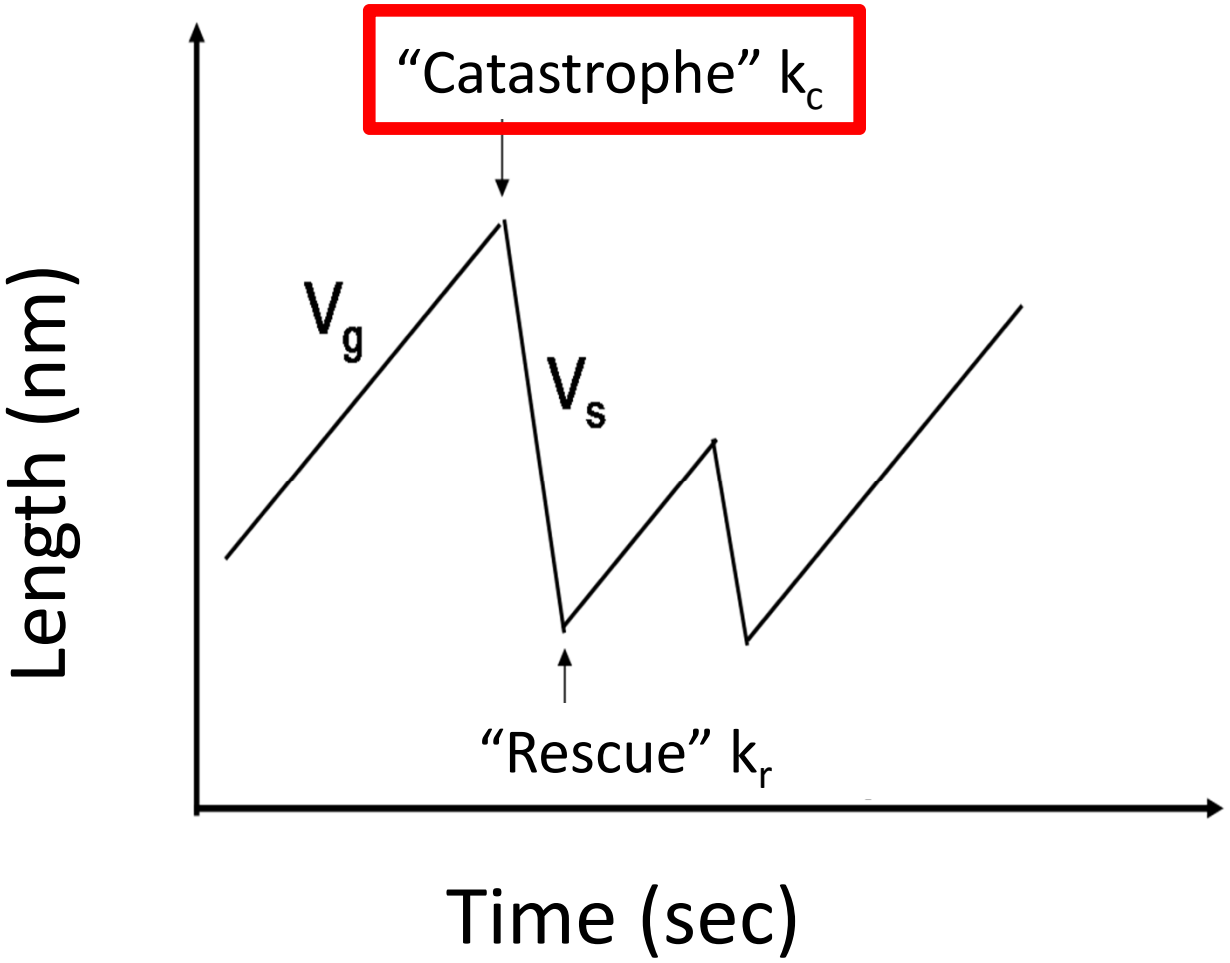
Melissa K. Gardner
University of Minnesota
Department of Genetics, Cell Biology, and Development

Microtubules: Length Regulation at the Nanoscale



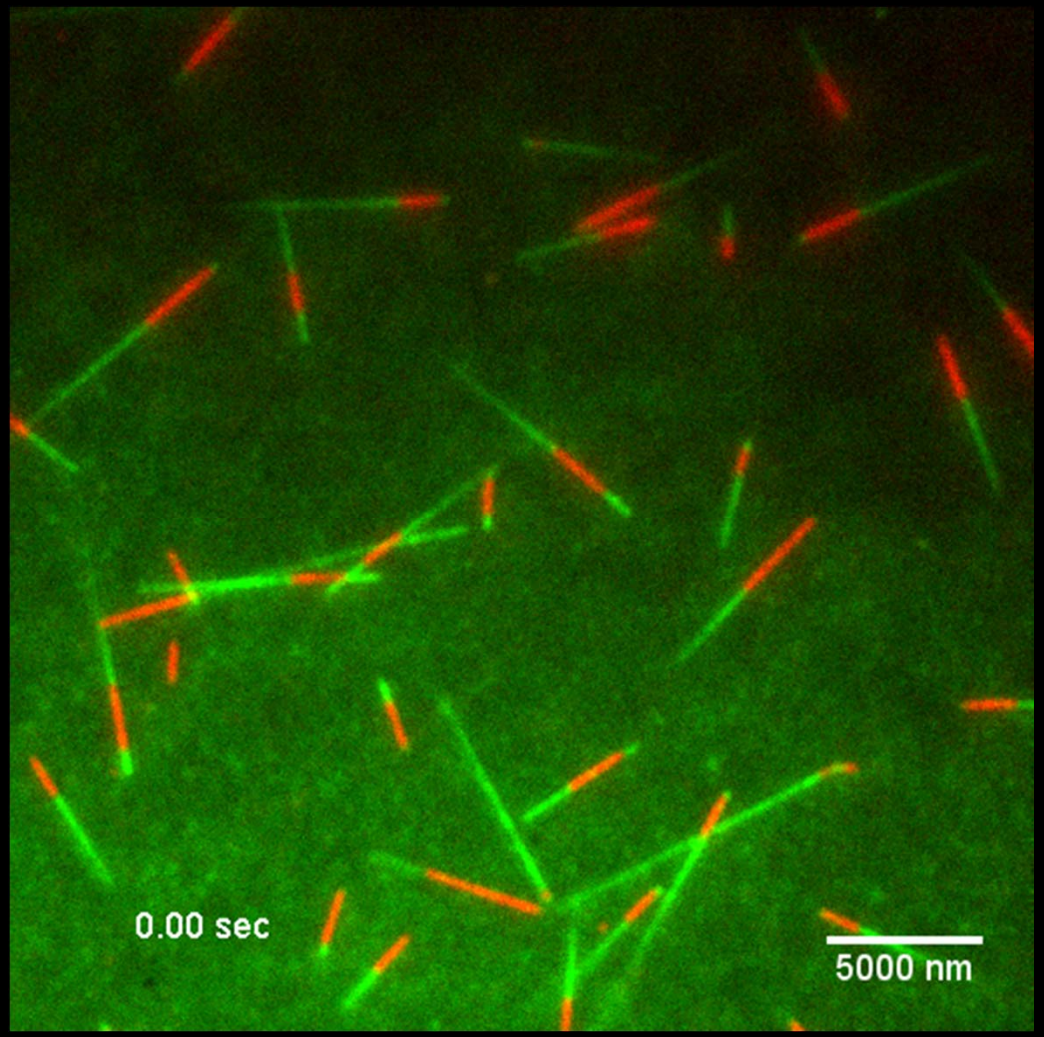
Silverman-Gavrila, R. In perfect synchrony - tubulin. ASCB Image & Video Library. August 2006:VID-5. Available at: http://cellimages.ascb.org (Drosophila melanogaster line expressing a fusion protein of GFP and alpha-tubulin (Grieder et al., 2000)

Catastrophe Events Limit Microtubule Lengths

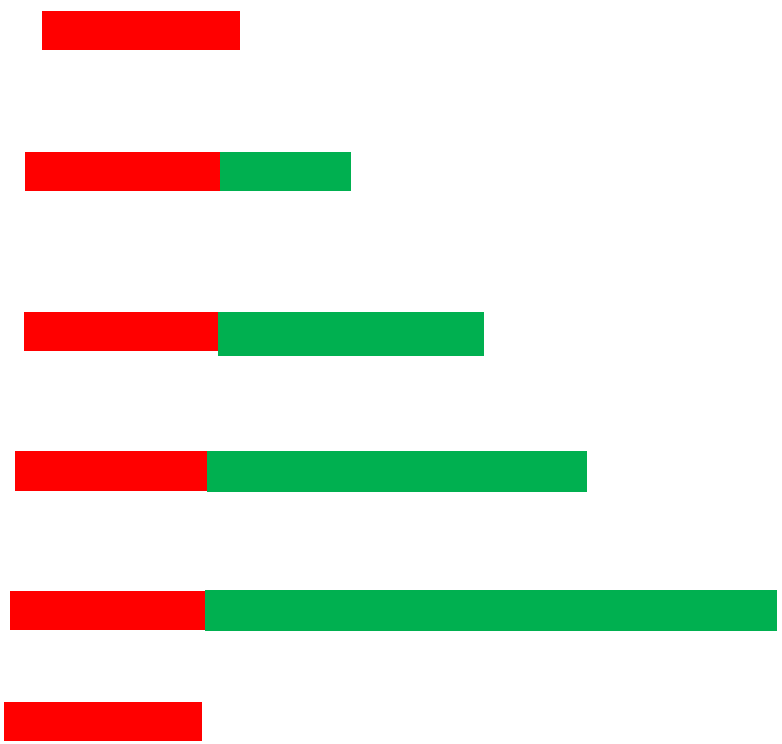


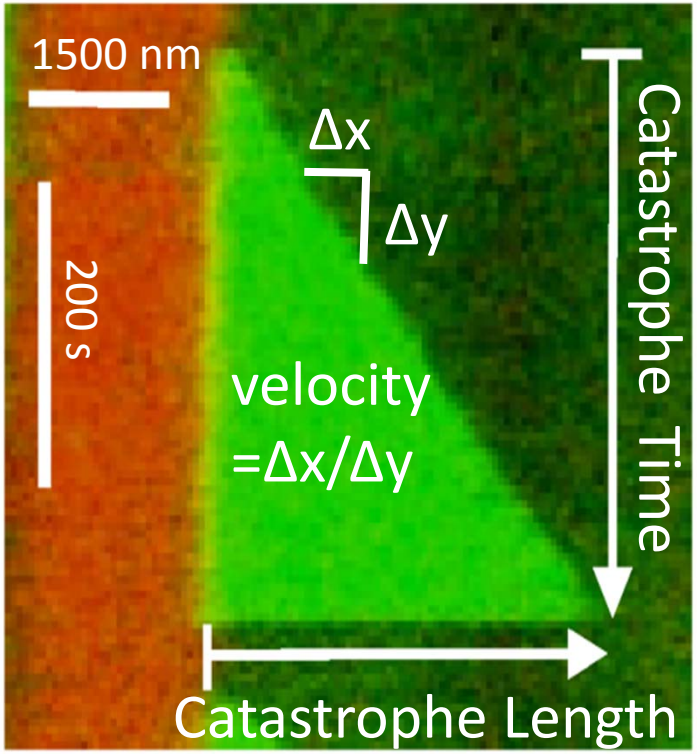
Catastrophe

Control (12 µM GTP-Tubulin)

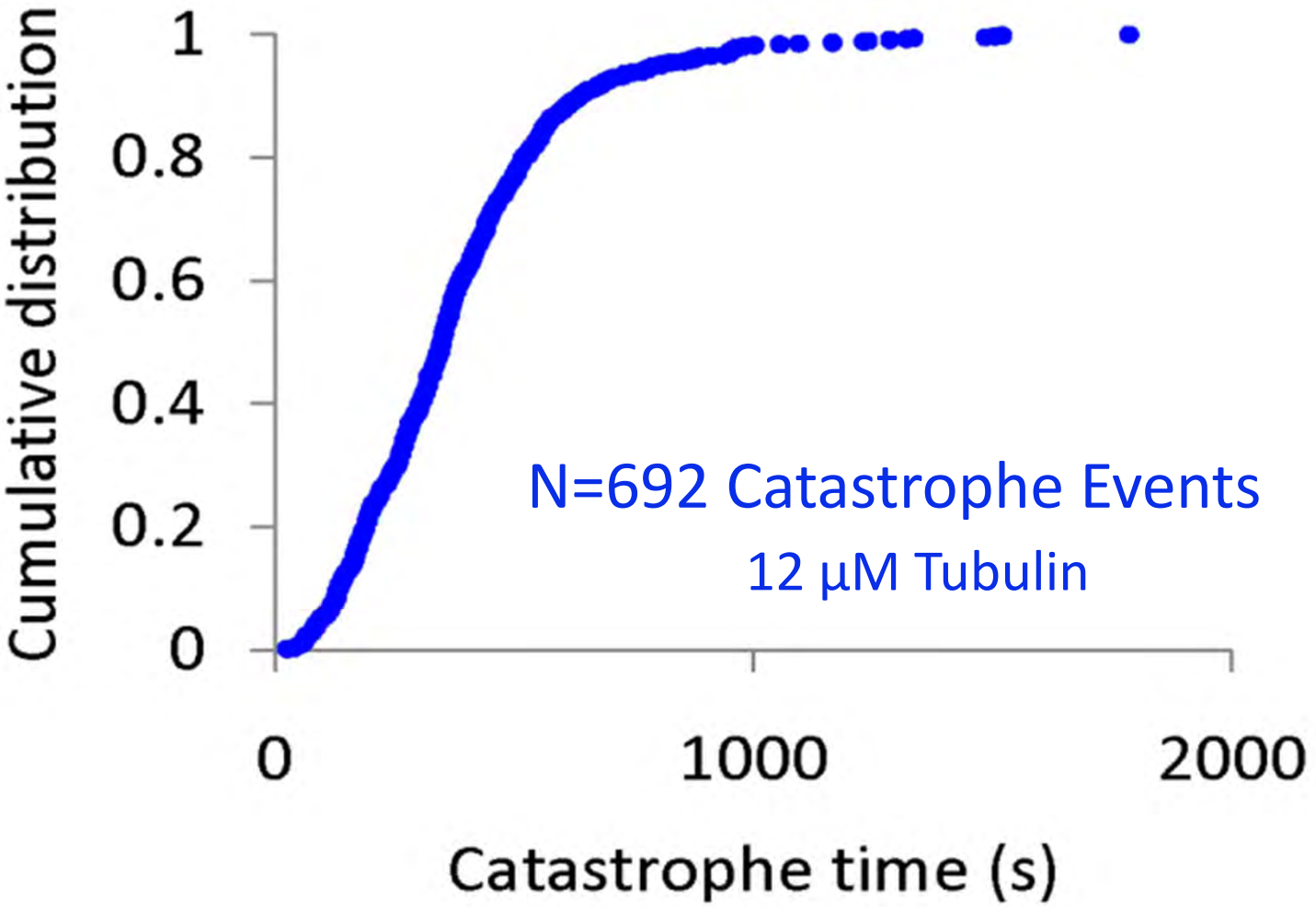


Catastrophe Length and Catastrophe Time Quantitatively Describe Catastrophe Events

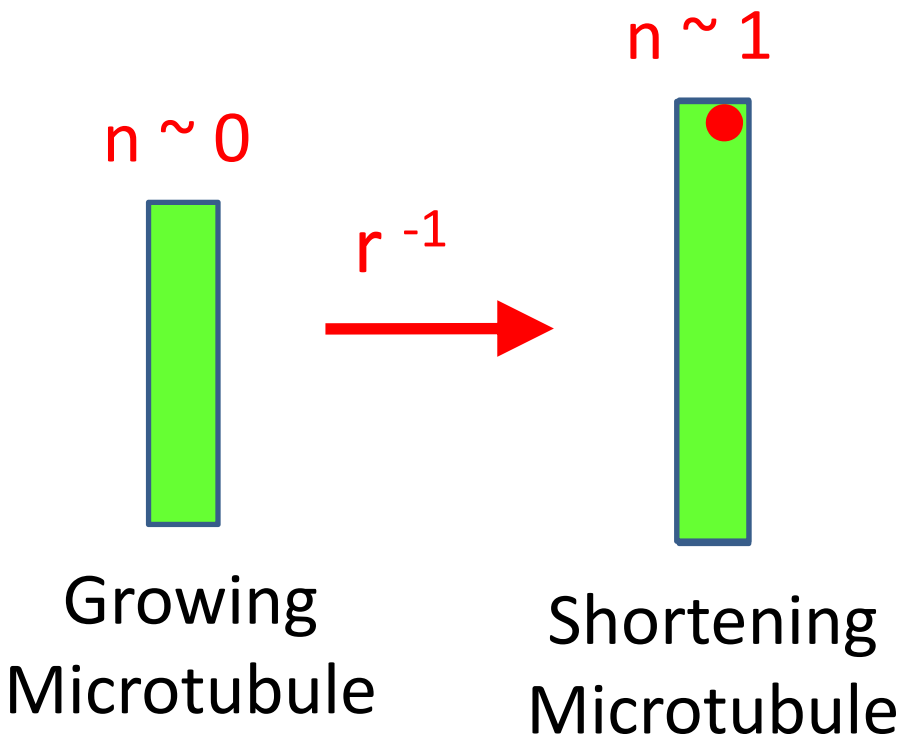




Cumulative Distribution of Catastrophe Times

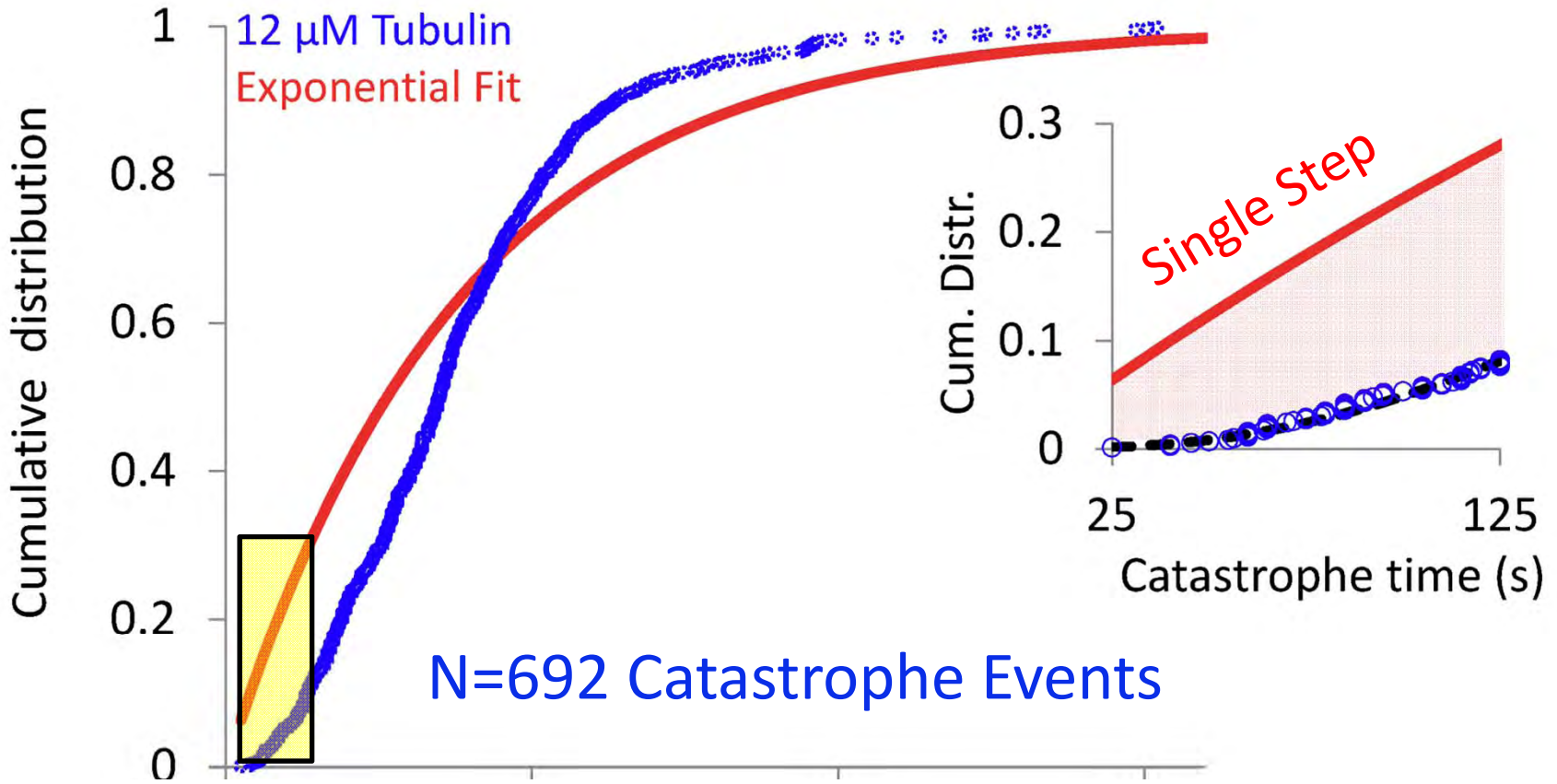


A Simple Model Describes Catastrophe as a Single Step Transition



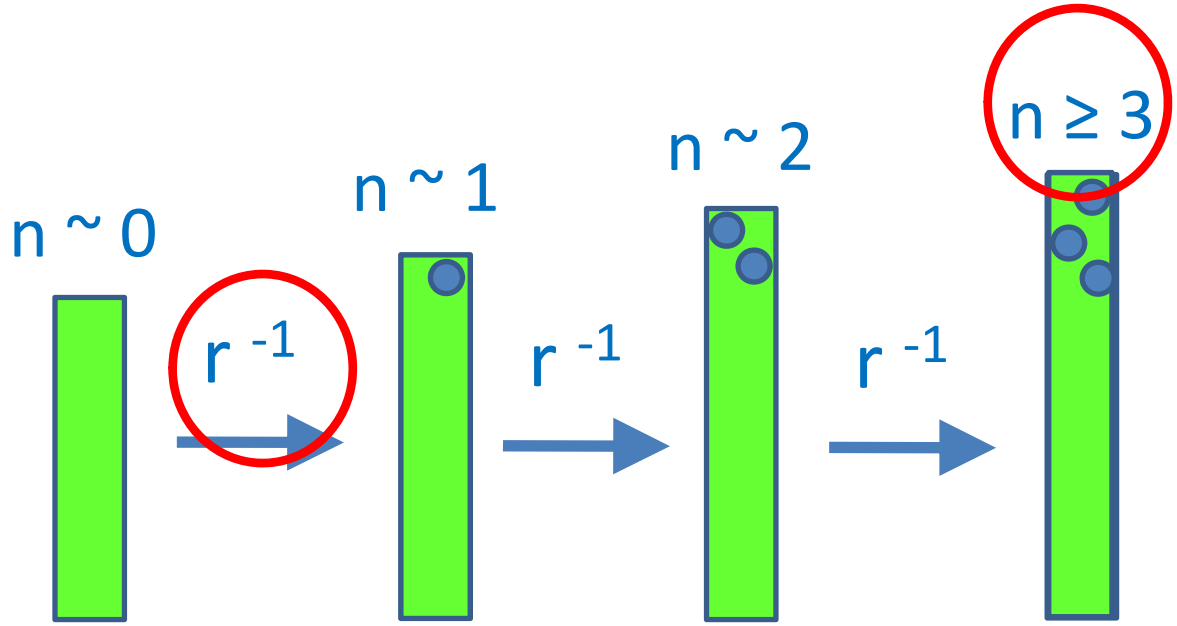
$$p_+(t) \sim \exp(-t/r)$$ Exponential Distribution

Catastrophe Time Data is Not Consistent with a Single Step Model



Odde *et al*, *Biophys J*, 1995 (DIC)

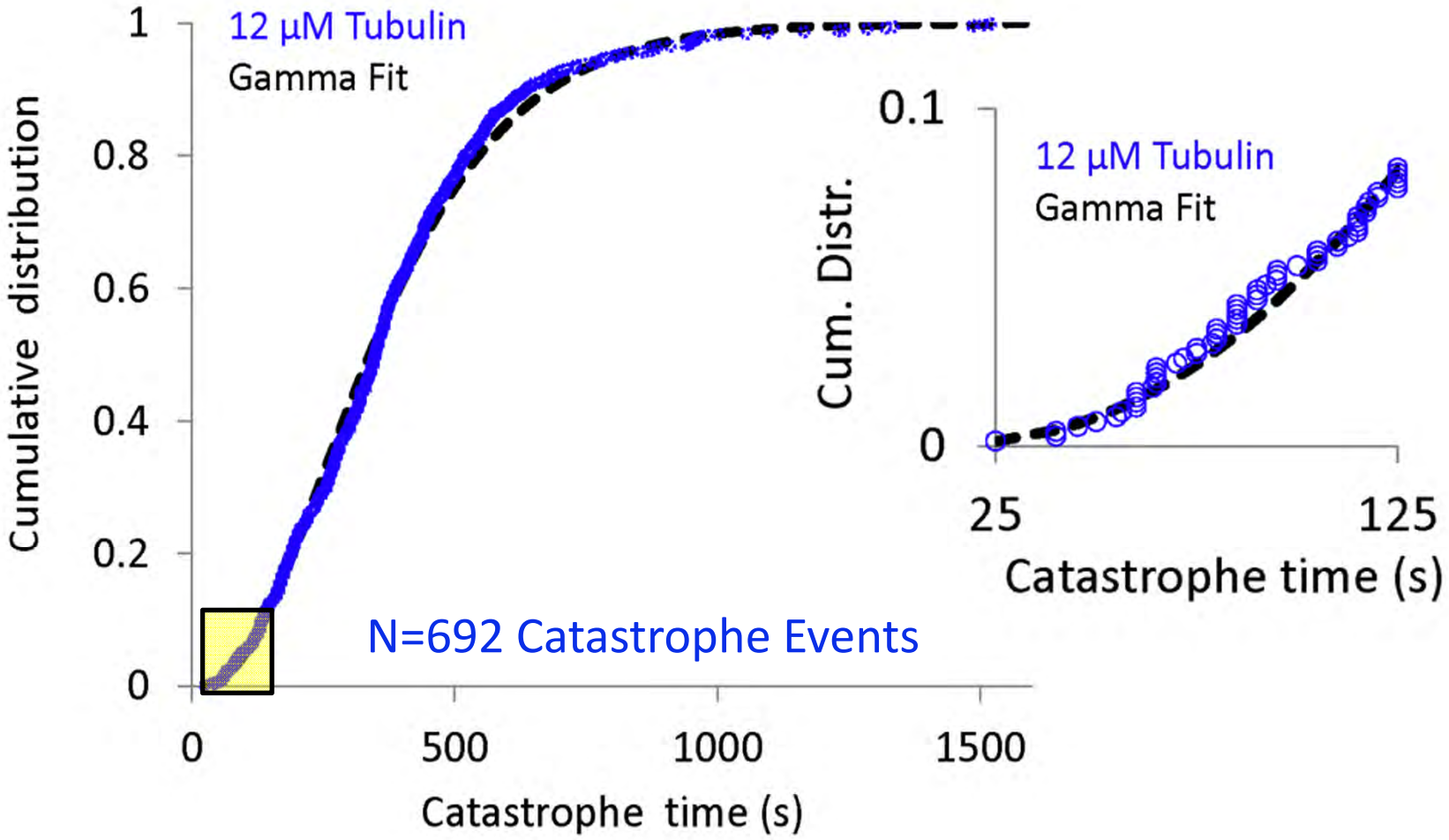
A Gamma Distribution Describes a Multiple-Step Transition Process



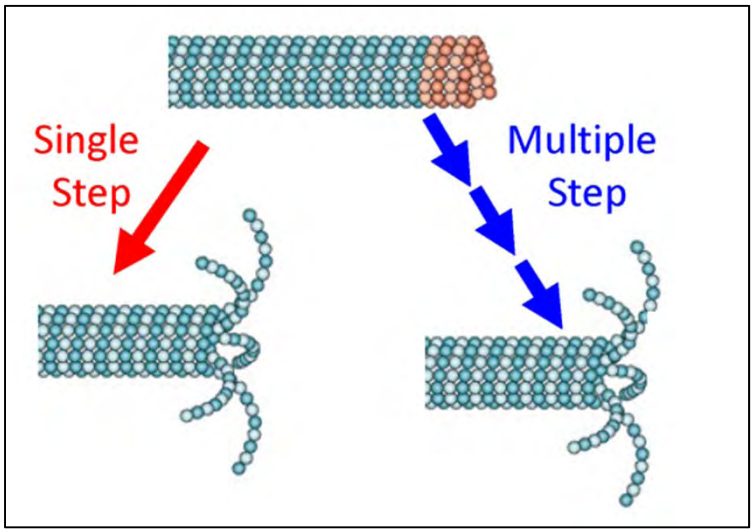
$$p_+(t) \sim x^{n-1} \frac{\exp(-t/r)}{(n-1)!}$$

Gamma Distribution

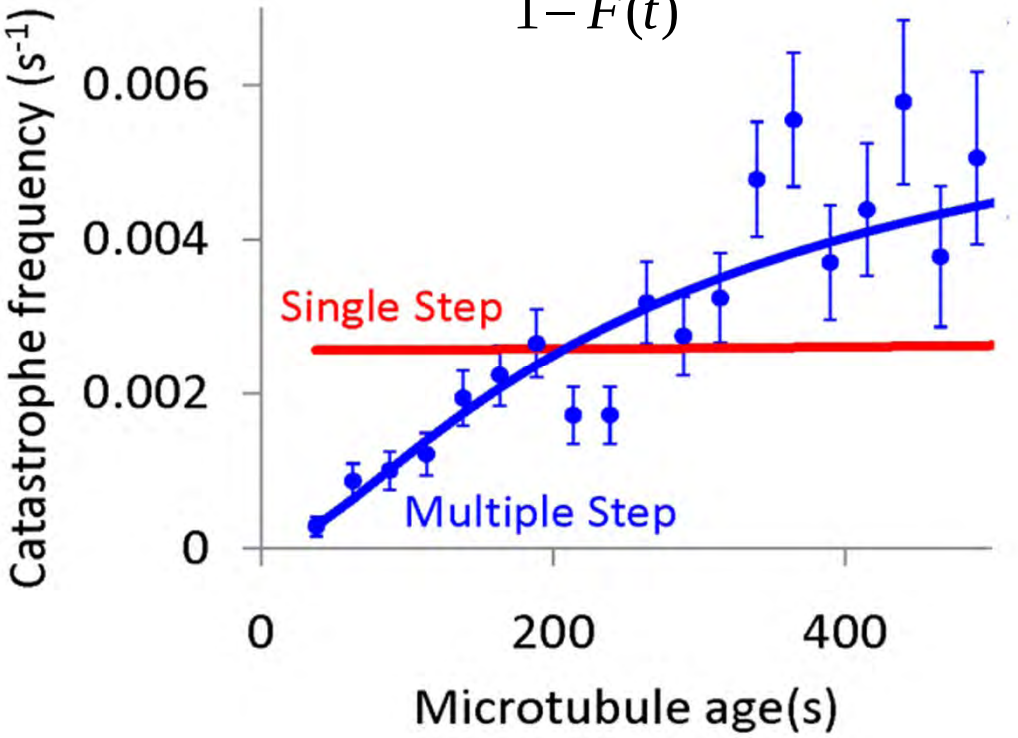
Catastrophe Time Data is Consistent with a Multi-step Model



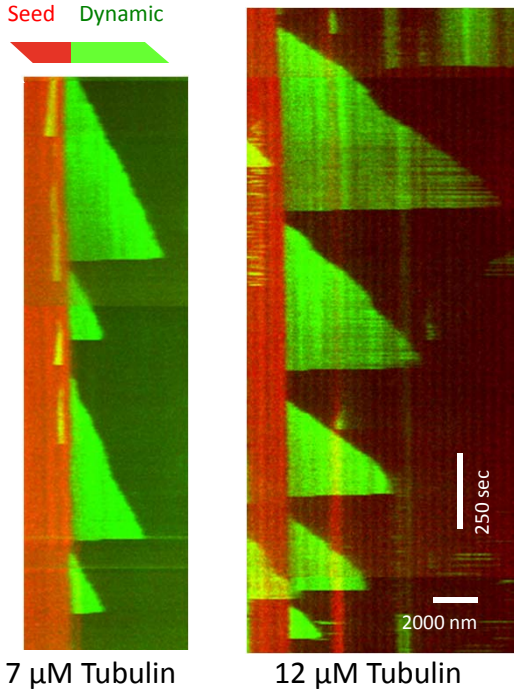
Catastrophe Frequency Increases with Microtubule Age

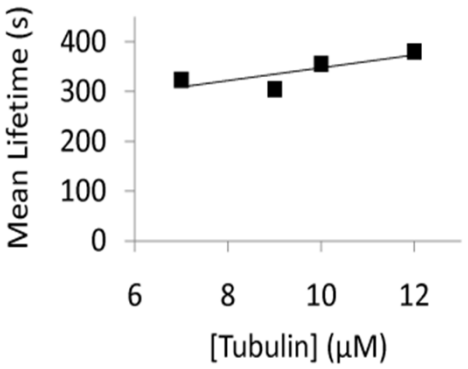


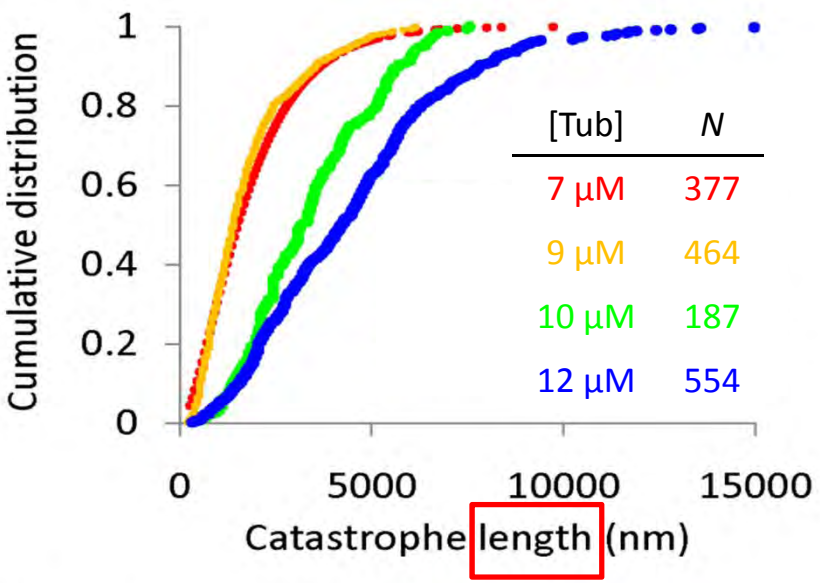
$$f_{\pm}(t) = \frac{dF(t)/dt}{1 - F(t)}$$

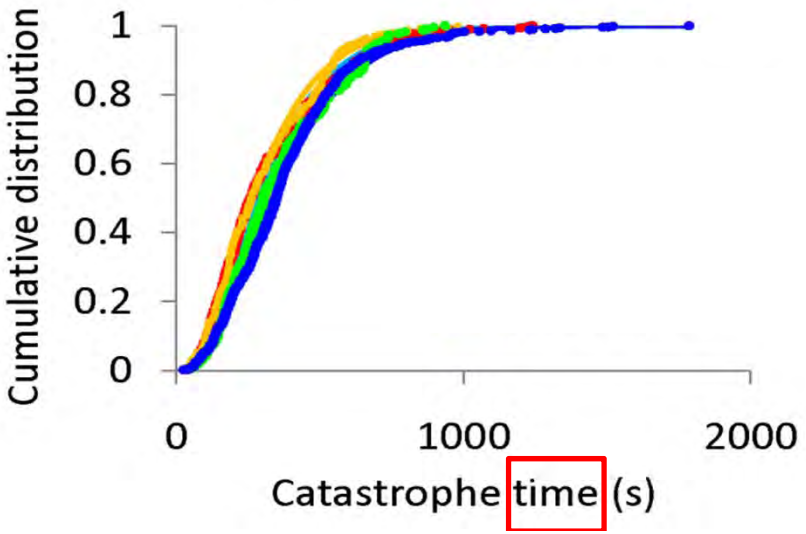


Catastrophe Frequency is Regulated by Microtubule Age Rather Than Length

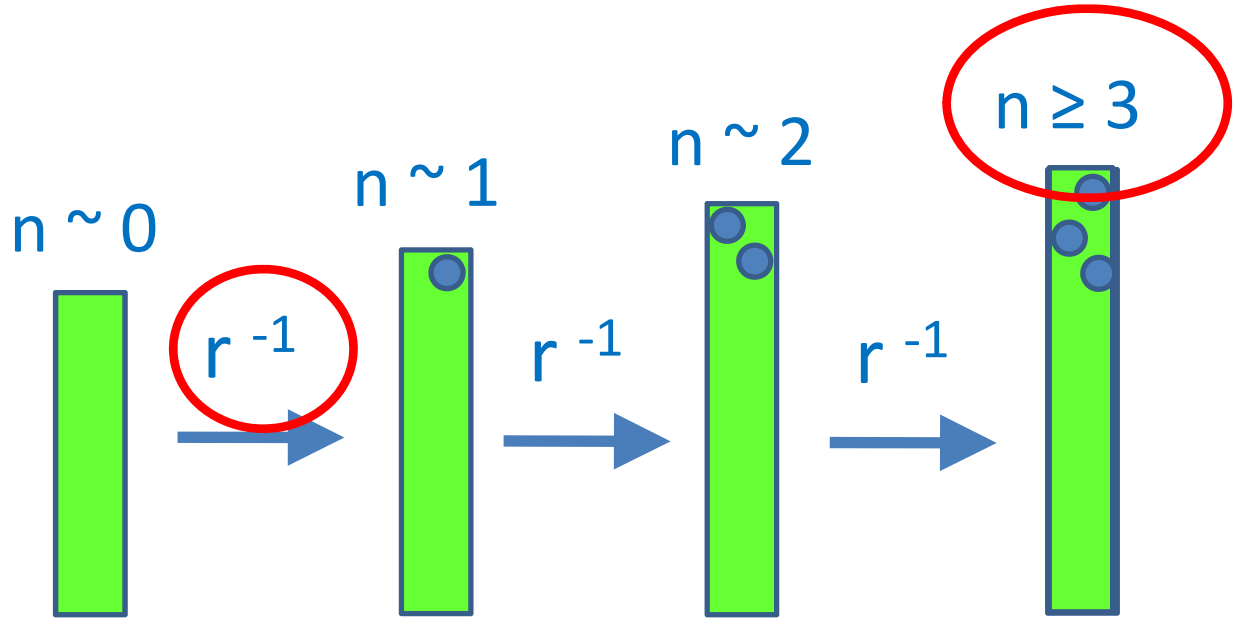




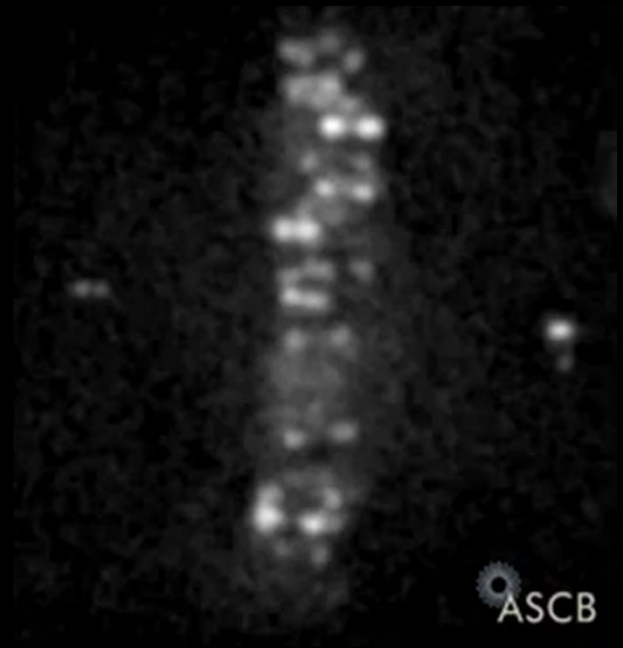




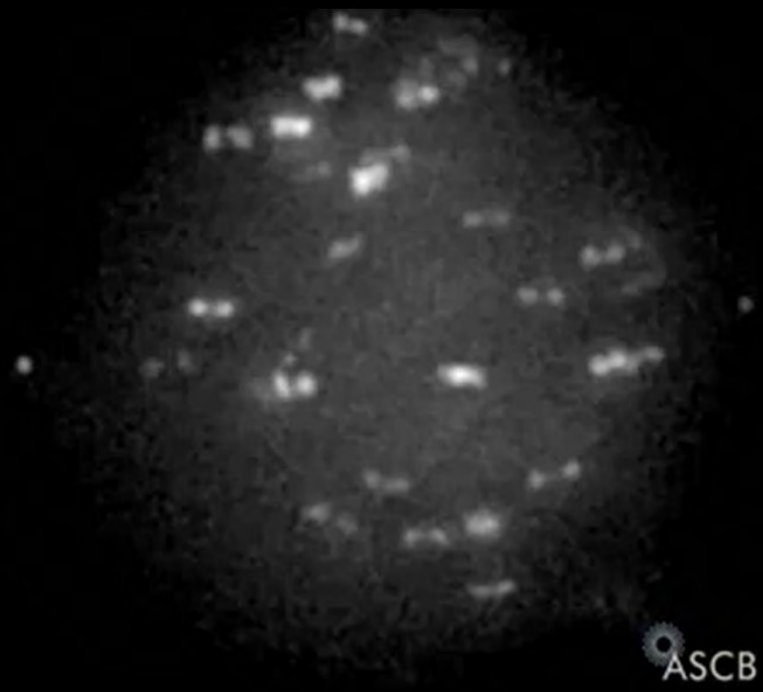
Do Depolymerizing Kinesins Regulate Catastrophe?



The Kip3 Homologue regulates microtubule-mediated chromosome alignment during mitosis

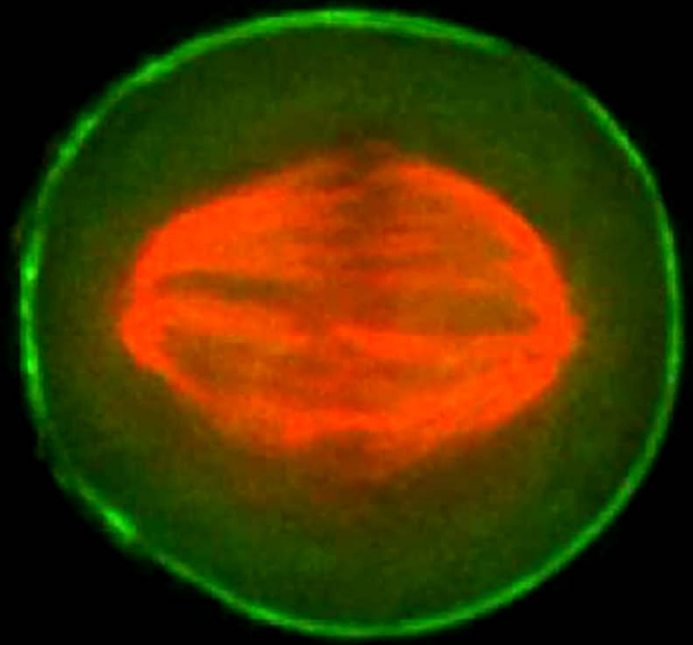


HeLa Mitosis Wild-type.
J. Stumpff et al Dev Cell (2008)

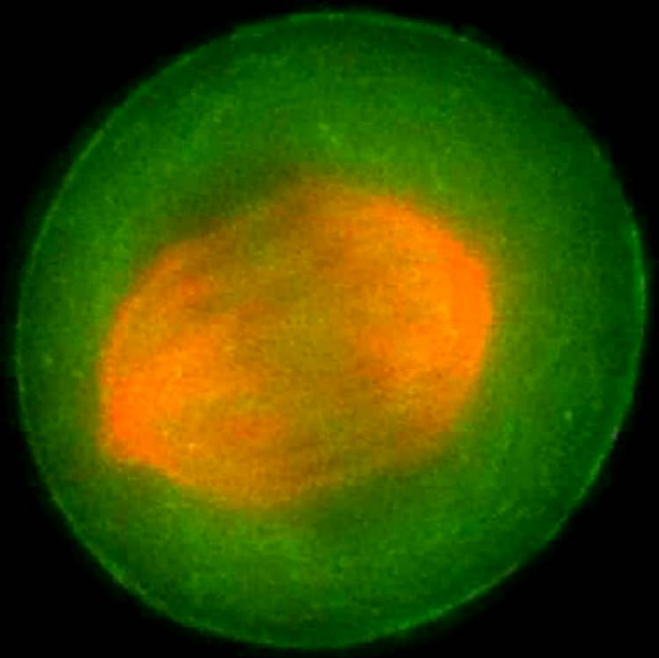


HeLa Mitosis. Kinesin-8 Rundown
J. Stumpff et al Dev Cell (2008)

MCAK Regulates Microtubule Length During Cell Division



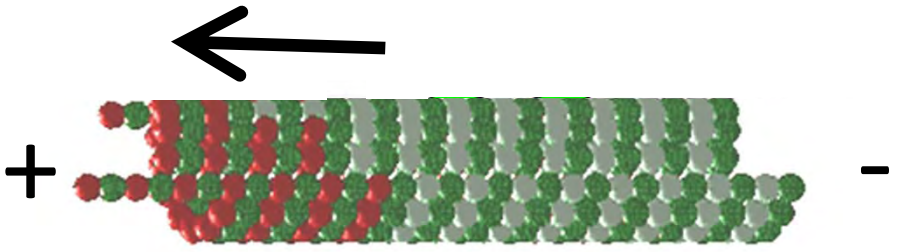
A HeLa cell expressing GFP–myosin IIA and mCherry– α-tubulin
Rankin & Wordeman, JCB, (2010)

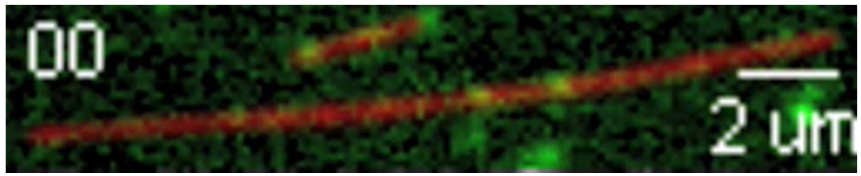


An MCAK-depleted HeLa cell expressing GFP–myosin IIA and mCherry– α-tubulin
Rankin & Wordeman, JCB, (2010)

Kip3 and MCAK are Molecular Motors

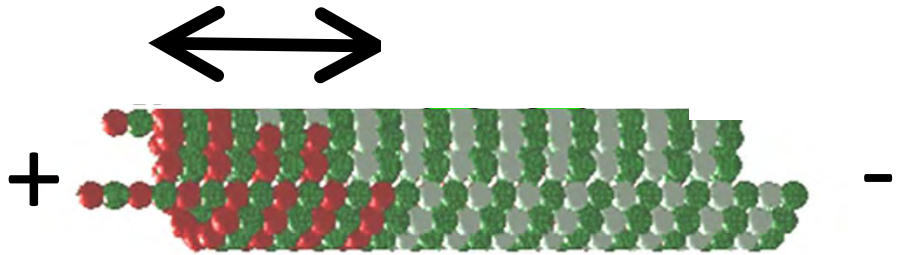
Kip3: Plus-End Directed

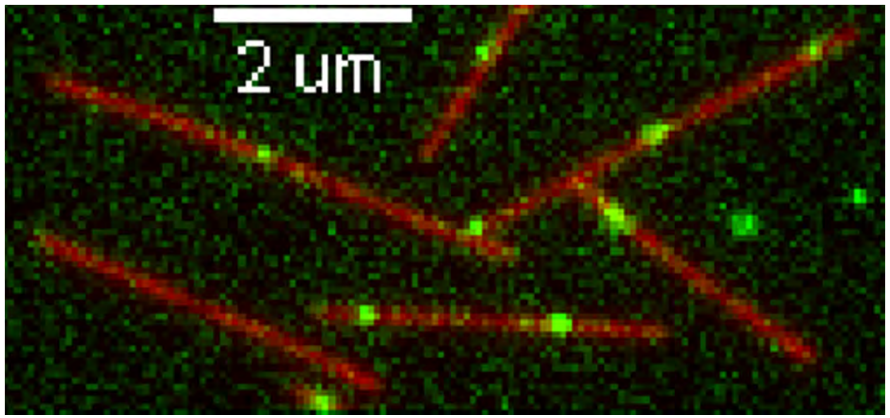




Varga et al, *Cell* (2009)

MCAK: Diffusive

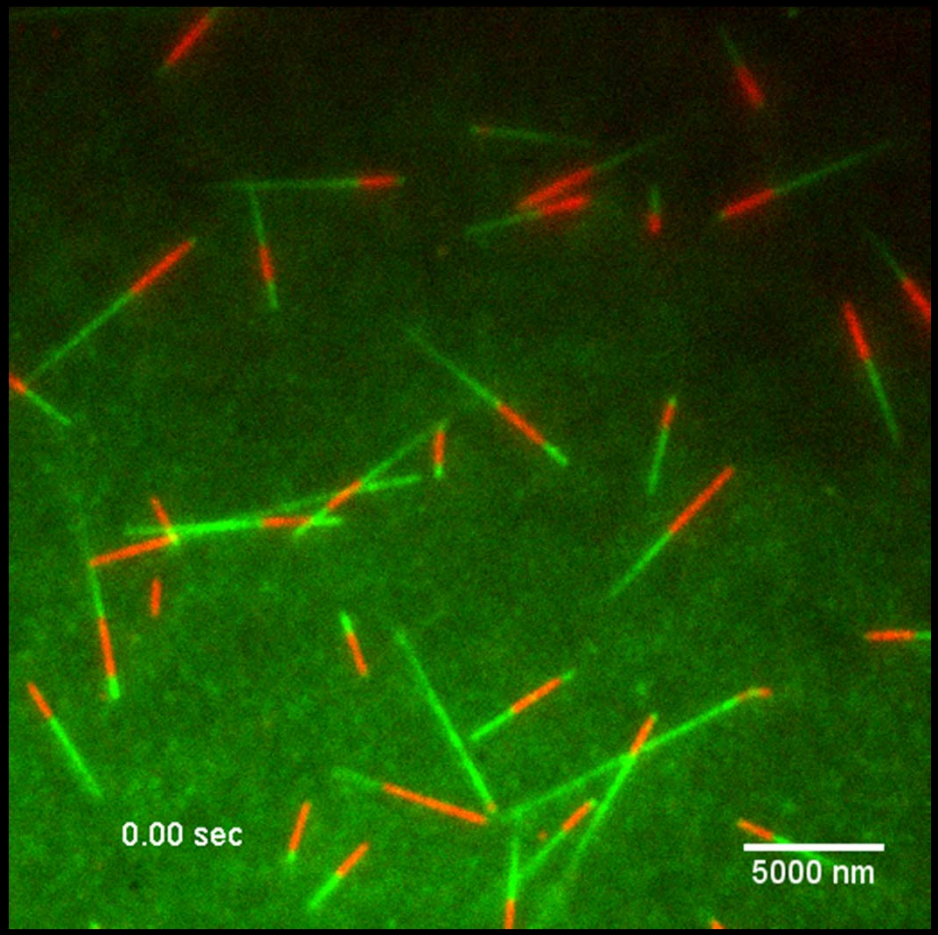




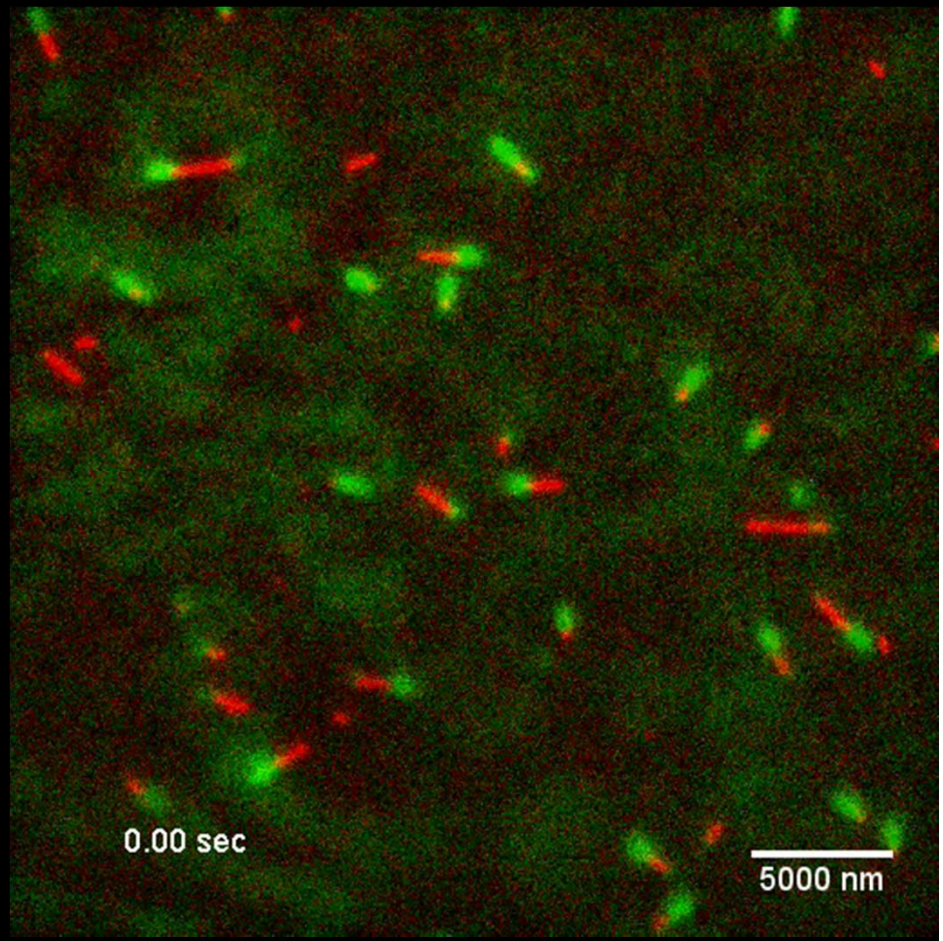
Helenius et al, *Nature* (2006)

Kip3 Results in Shorter Catastrophe Lengths

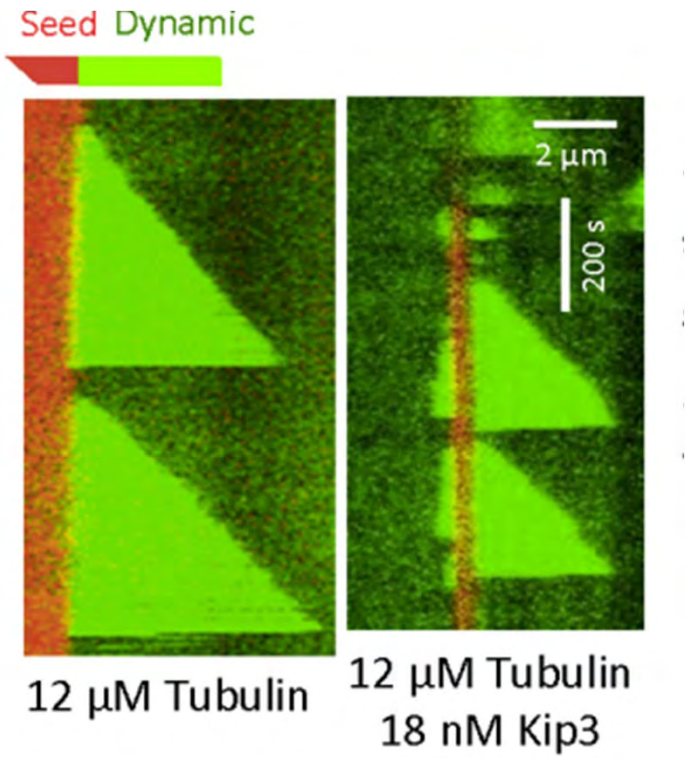
12 µM GTP-Tubulin

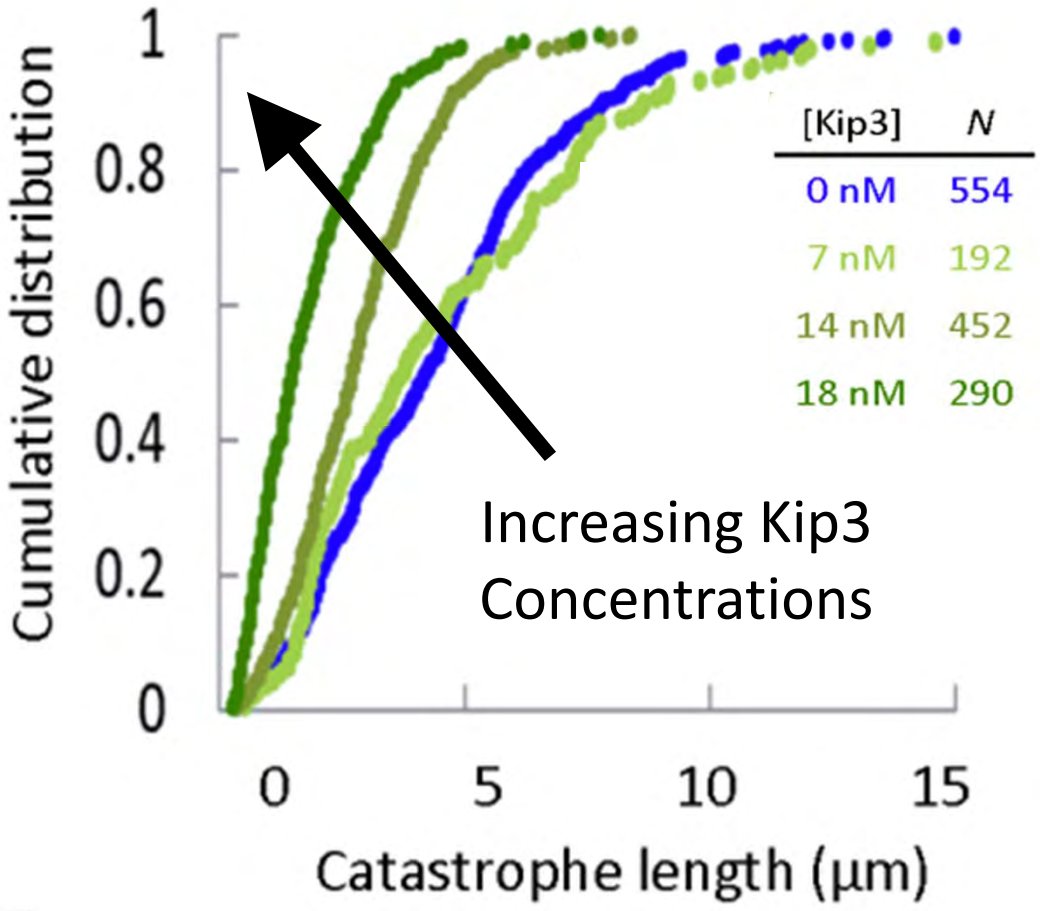


12 µM GTP-Tubulin with Kip3



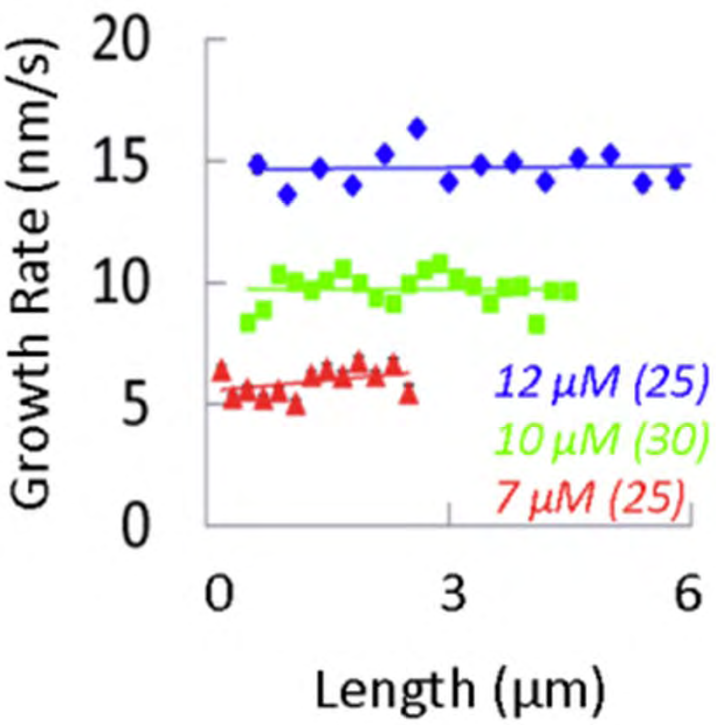
Kip3 Results in Shorter Catastrophe Lengths



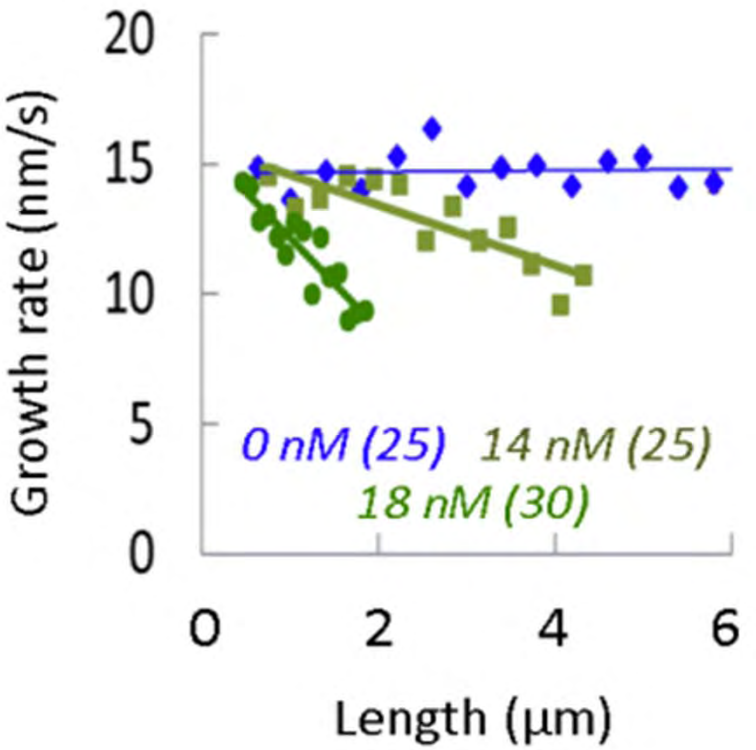


Kip3 Promotes Length-Dependent Slowing of Microtubule Growth

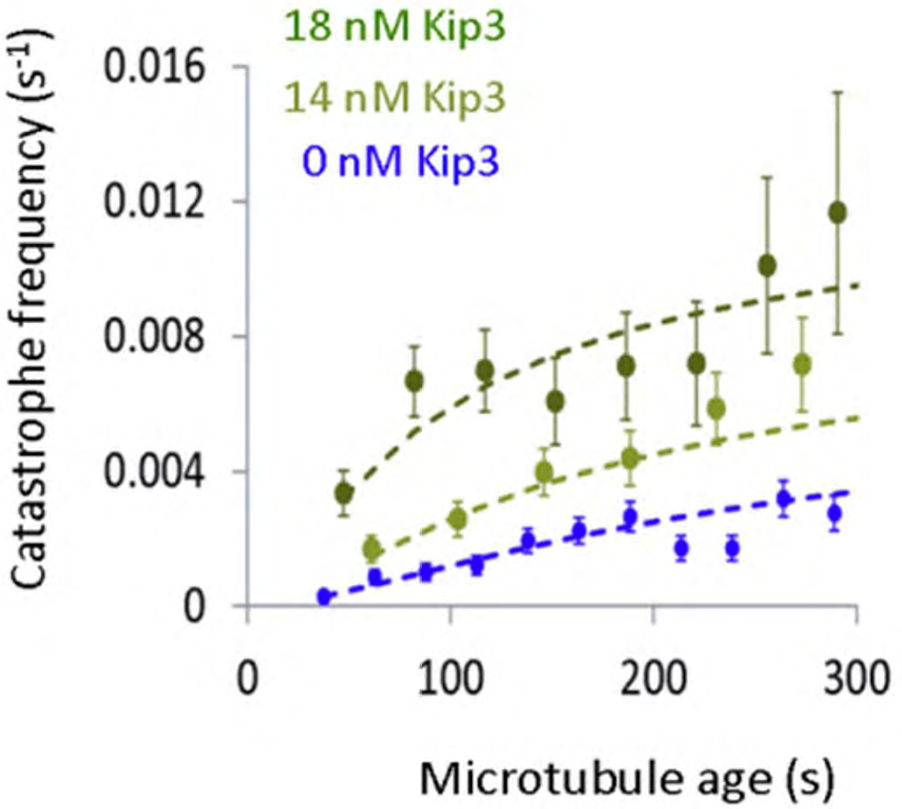
Tubulin Alone

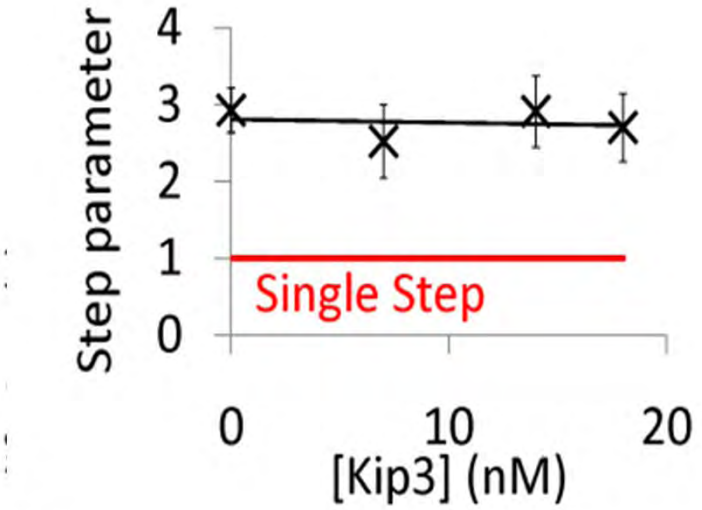


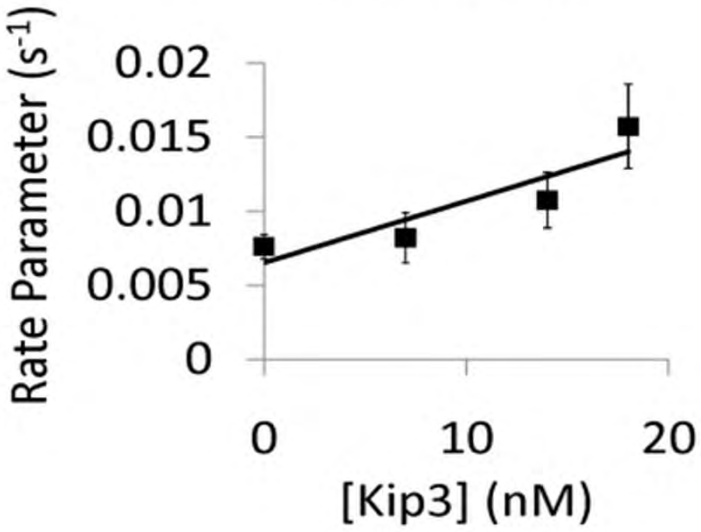
Tubulin with Kip3



Kip3 Promotes Catastrophe by Accelerating the Rate of Microtubule Aging

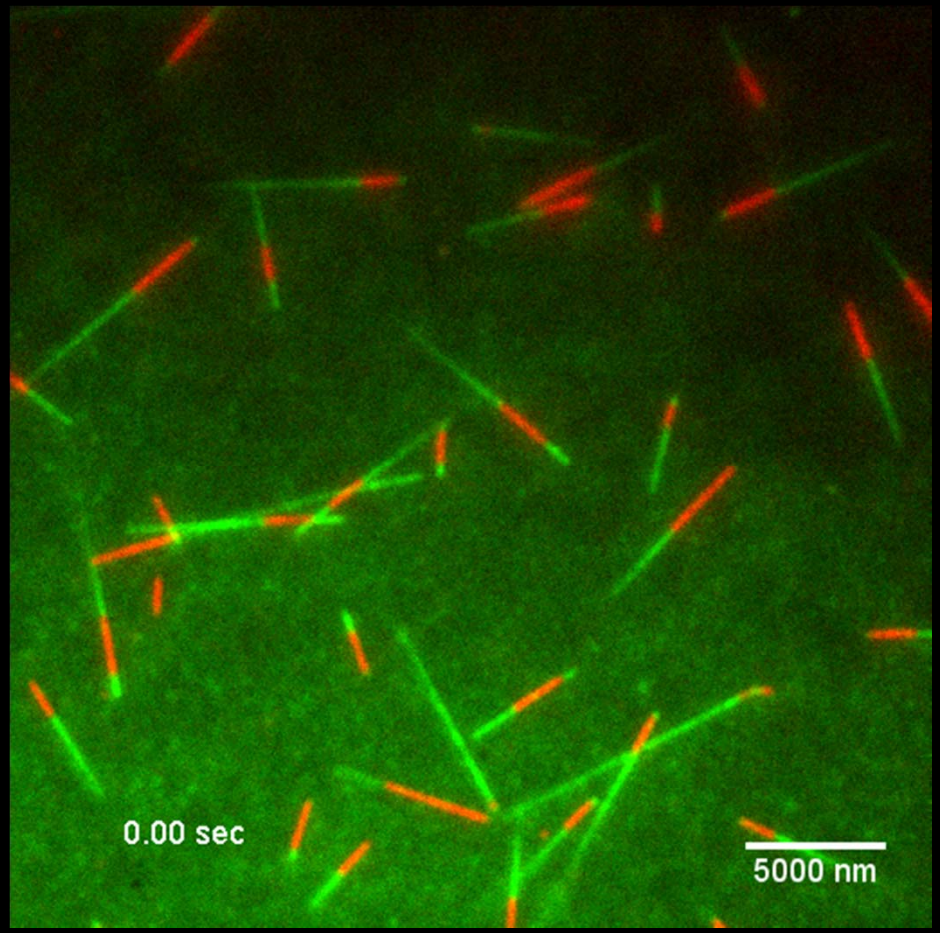




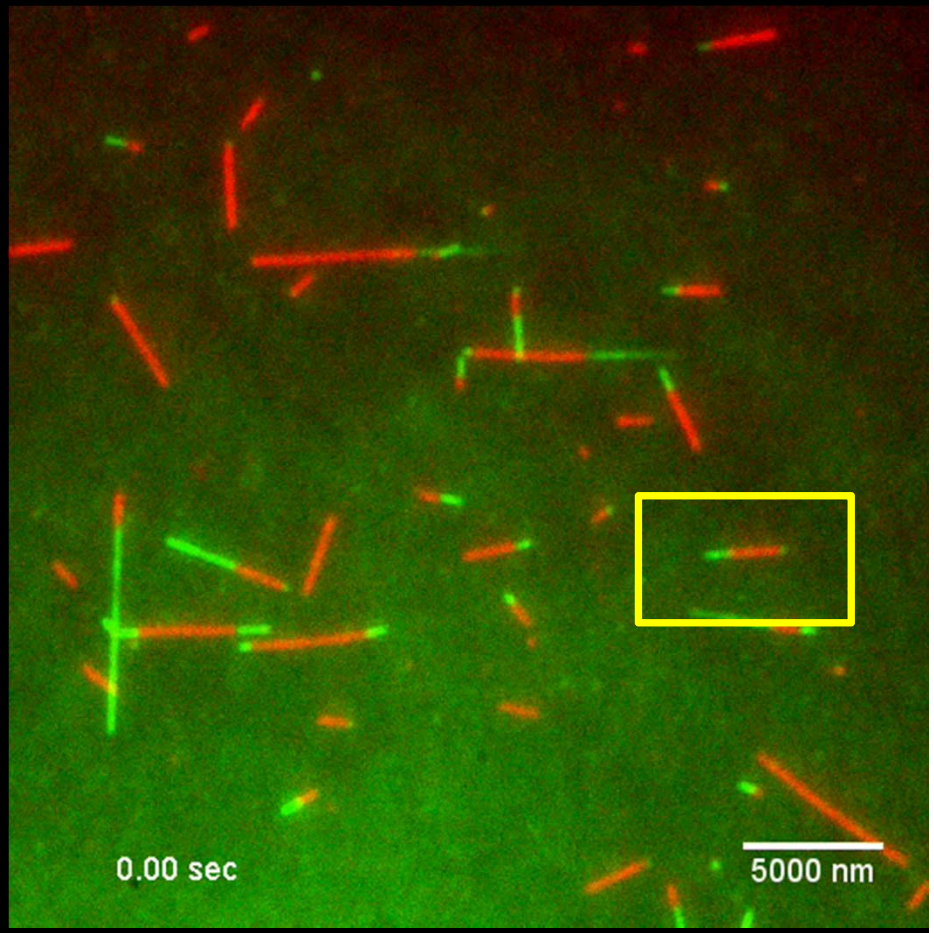


MCAK Also Results in Shorter Catastrophe Lengths

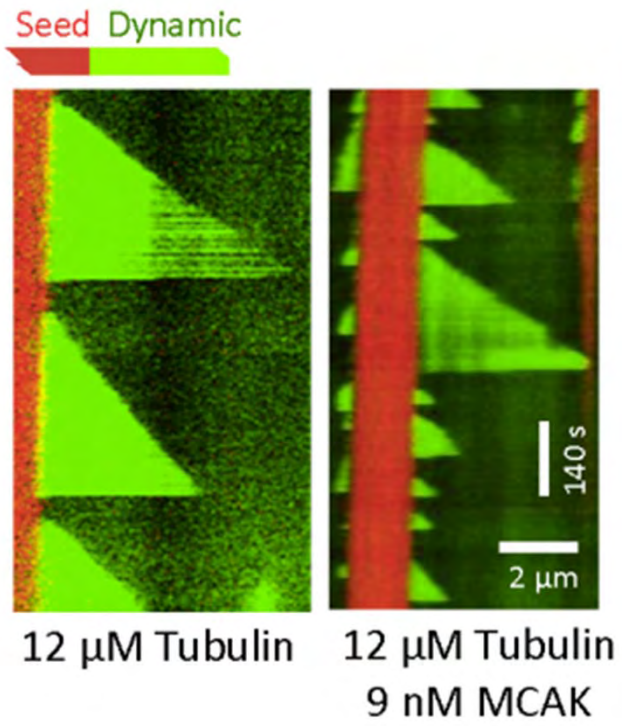
12 µM GTP-Tubulin

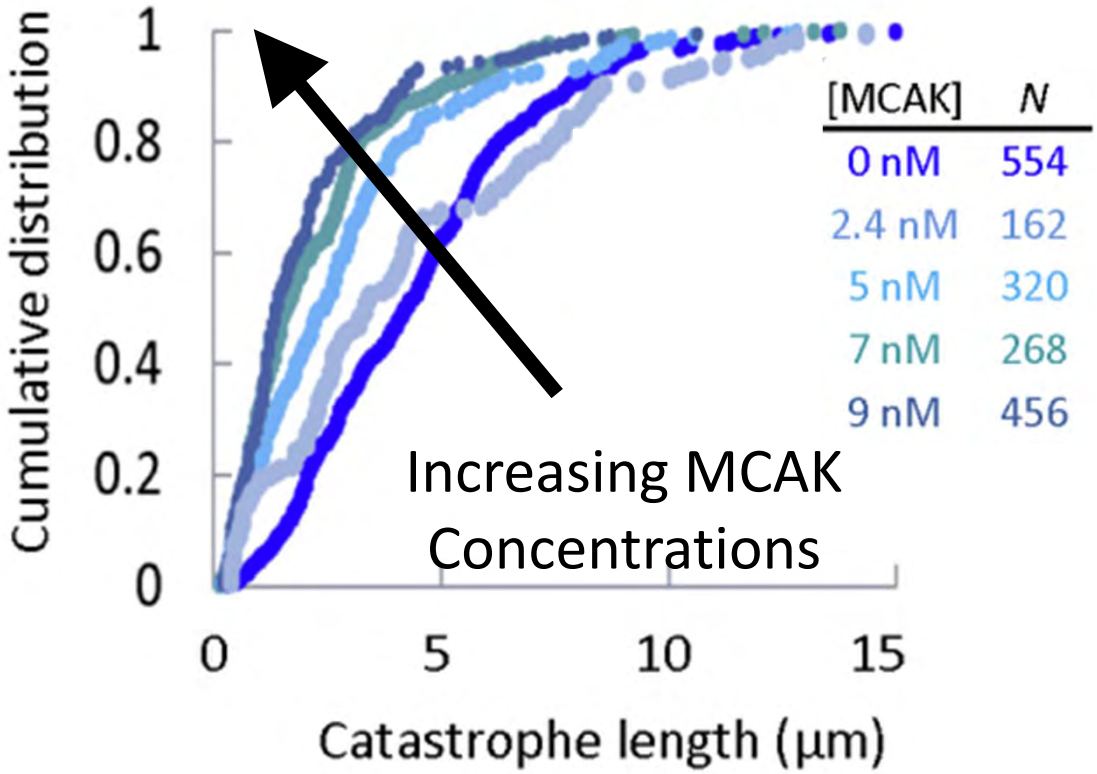


12 µM GTP-Tubulin with MCAK



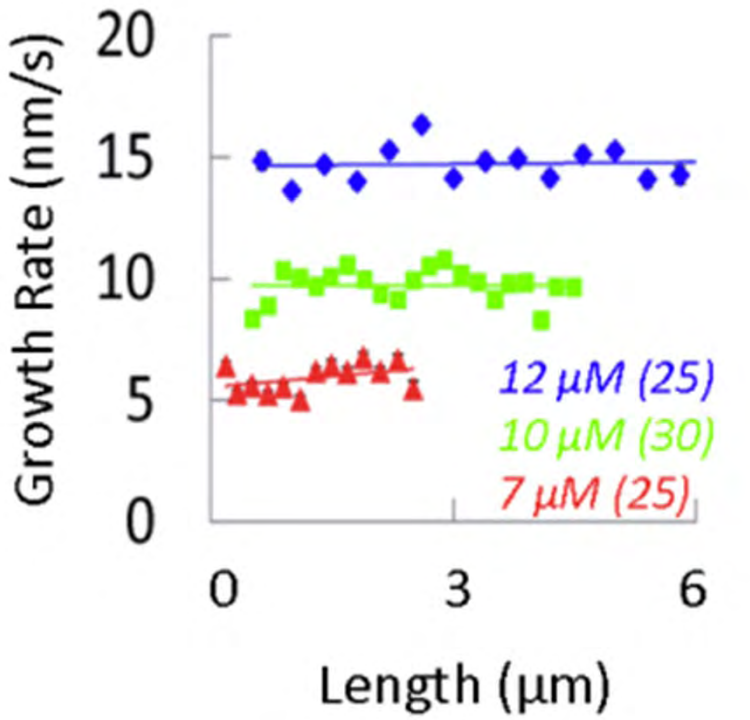
MCAK Results in Shorter Catastrophe Lengths



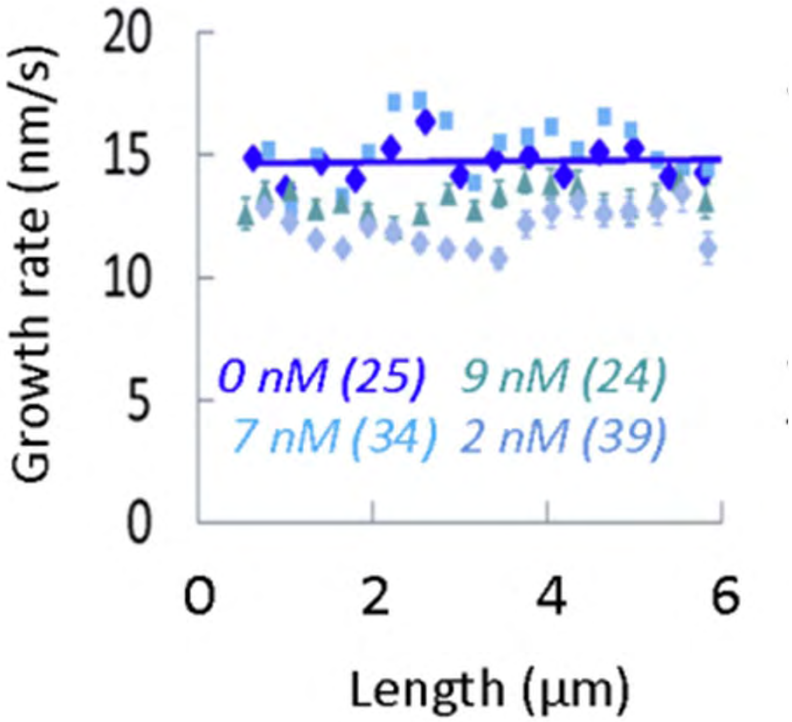


MCAK Does Not Change Microtubule Growth Rates

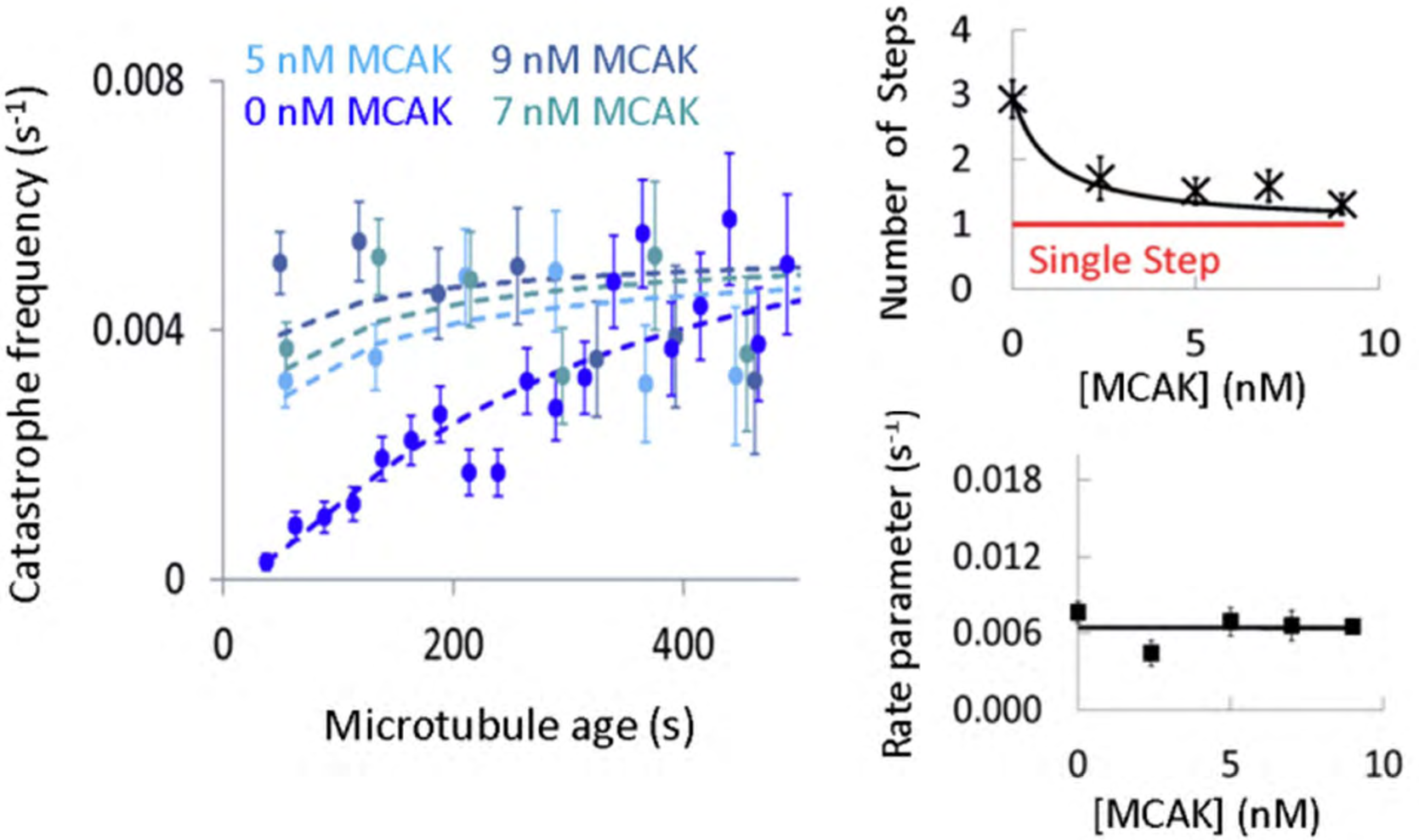
Tubulin Alone



Tubulin with MCAK



MCAK Promotes Catastrophe by Eliminating the Microtubule Aging Process

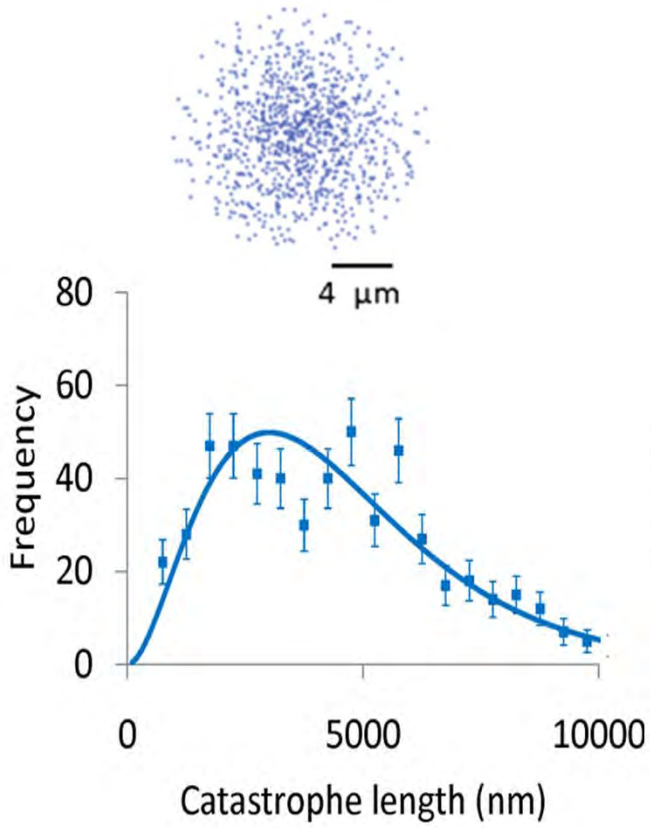


-In vitro Catastrophe Frequency is Regulated by the Age of the Microtubule

-The Depolymerizing Kinesins Kip3 and MCAK Promote Catastrophe by Modifying the Aging Process: Kip3 accelerates aging, while MCAK eliminates the aging process

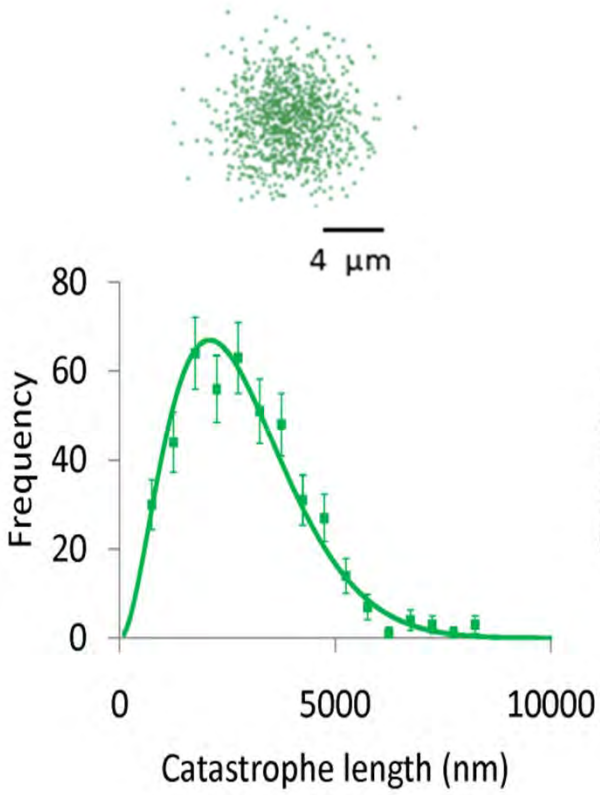
What are the Consequences for Microtubule Length Control?

Regulated Aging Allows for Fine-Tuned Microtubule Length Control



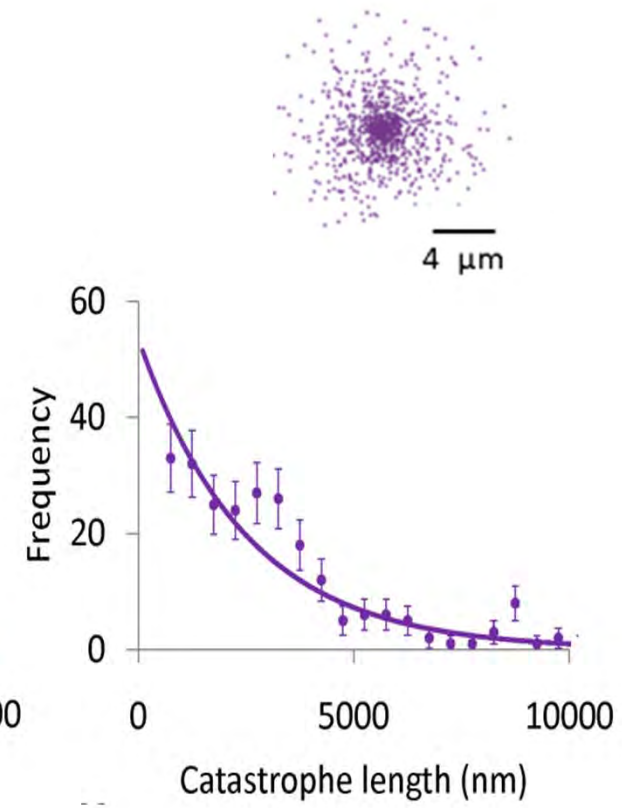
Control

Concentrated space exploration



Kip3

Length-dependent growth rates further concentrate space exploration



MCAK

Rapid Restructuring

THANKS

Joe Howard

Max Planck Institute for Molecular Cell Biology and Genetics
and the Howard Lab, particularly:
Marija Zanic, Chris Gell, Volker Bormuth

Whitaker International Scholars Program

Pew Scholars Program in the Biomedical Sciences

THE PEW CHARITABLE TRUSTS