

# **Opposing kinesin complexes queue at plus tips to ensure microtubule catastrophe at cell ends**

## **Appendix Contents**

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**Appendix Table S1. List of strains used in this study**

**Figure 1B**

JM8018	<i>leu1-32::SV40-GFP-atb2:leu1 h<sup>-</sup></i>	F. Chang
JM8427	<i>klp5::natR klp6::hygR leu1-32::SV40-GFP-atb2:leu1 h<sup>-</sup></i>	this study
JM8334	<i>mcp1::kanR leu1-32::SV40-GFP-atb2:leu1 h<sup>+</sup></i>	this study
JM8547	<i>mcp1::kanR klp5::natR klp6::hygR leu1-32::SV40-GFP-atb2:leu1 h<sup>?</sup></i>	this study

**Figure 1C-E**

JM6823	<i>natR::Z:adh15-mCherry-atb2 h<sup>-</sup></i>	K. Sawin
JM9441	<i>klp5-mNeonGreen:kanR klp6-mNeonGreen:hygR natR::Z:adh15-mCherry-atb2 h<sup>+</sup></i>	this study
JM9877	<i>mcp1::natR klp5-mNeonGreen:kanR klp6-mNeonGreen:hygR natR::Z:adh15-mCherry-atb2 h<sup>?</sup></i>	this study

**Figure 1F**

JM9513	<i>klp5-mNeonGreen:kanR klp6-mNeonGreen:hygR natR::Z:adh15-mCherry-atb2 cut11-mCherry:ura4 h<sup>?</sup></i>	this study
JM9877	<i>mcp1::natR klp5-mNeonGreen:kanR klp6-mNeonGreen:hygR natR::Z:adh15-mCherry-atb2 h<sup>?</sup></i>	this study

**Figure EV1A**

JM7175	<i>klp5-GFP:kanR natR::Z:adh15-mCherry-atb2 h<sup>-</sup></i>	this study
JM7129	<i>mcp1-GFP:hygR natR::Z:adh15-mCherry-atb2 h<sup>-</sup></i>	this study

**Figure EV1B**

JM2943	<i>klp5::kanR h<sup>+</sup></i>	T. Toda
JM3089	<i>klp6::ura4 h<sup>-</sup></i>	T. Toda
JM7129	<i>mcp1-GFP:hygR natR::Z:adh15-mCherry-atb2 h<sup>-</sup></i>	this study
JM7185	<i>mcp1-GFP:hygR natR::Z:adh15-mCherry-atb2 klp5::kanR h</i>	this study
JM7187	<i>mcp1-GFP:hygR natR::Z:adh15-mCherry-atb2 klp6::ura4 h</i>	this study
JM7208	<i>mcp1-GFP:hygR natR::Z:adh15-mCherry-atb2 klp5::kanR klp6::ura4 h</i>	this study
JM8520	<i>mcp1-GFP:hygR natR::Z:adh15-mCherry-atb2 klp5::kanR klp6::ura4 lys1::klp5:kanR his3::klp6:blaR h<sup>-</sup></i>	this study
JM8521	<i>mcp1-GFP:hygR natR::Z:adh15-mCherry-atb2 klp5::kanR klp6::ura4 lys1::kanR his3::blaR h<sup>-</sup></i>	this study
JM8583	<i>mcp1-GFP:hygR natR::Z:adh15-mCherry-atb2 klp5::kanR klp6::ura4 lys1::klp5(G277A,E299A):kanR his3::klp6(G296A,E298A):blaR h<sup>-</sup></i>	this study

**Figure EV1C**

JM9325	<i>mcp1::natR lys1::nmt1-GFP-mcp1:ura4 klp5-13Myc:kanR h<sup>-</sup></i>	this study
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**Figure EV1D**

JM196	<i>cdc25-22 h<sup>+</sup></i>	lab stock
JM8009	<i>mcp1::kanR cdc25-22 h<sup>-</sup></i>	this study
JM5963	<i>klp5::natR klp6::ura4 cdc25-22 h<sup>?</sup></i>	lab stock
JM9321	<i>mcp1::hygR cdc25-22 klp5::natR klp6::ura4 h<sup>-</sup></i>	this study

**Figure EV1E**

JM7175	<i>klp5-GFP:kanR natR::Z:adh15-mCherry-atb2 h<sup>-</sup></i>	this study
JM7202	<i>mcp1::hygR klp5-GFP:kanR natR::Z:adh15-mCherry-atb2 h<sup>-</sup></i>	this study
JM7177	<i>klp6-GFP:hygR natR::Z:adh15-mCherry-atb2 h<sup>-</sup></i>	this study
JM7205	<i>mcp1::kanR klp6-GFP:hygR natR::Z:adh15-mCherry-atb2 h<sup>-</sup></i>	this study

**Figure EV2A**

JM3707	<i>fta3-GFP:kanR h<sup>-</sup></i>	X. He
JM3725	<i>sid4-TdTomato:hygR h<sup>+</sup></i>	I. Hagan
JM5061	<i>fta3-GFP:kanR sid4-tdTomato:hygR h<sup>-</sup></i>	lab stock
JM6143	<i>klp5::ura4 fta3-GFP:kanR sid4-tdTomato:hygR h<sup>?</sup></i>	lab stock
JM7141	<i>mcp1::natR fta3-GFP:kanR sid4-tdTomato:hygR h<sup>-</sup></i>	this study
<b>Figure EV2B</b>		
JM2589	<i>cdc13-117:cdc13-GFP:LEU2 h<sup>-</sup></i>	M. Yanagida
JM5785	<i>cdc13-117:cdc13-GFP:LEU2 sid4-TdTomato:hygR h<sup>-</sup></i>	this study
JM9138	<i>klp5::ura4 cdc13-117:cdc13-GFP:LEU2 sid4-TdTomato:hygR h<sup>+</sup></i>	this study
JM9131	<i>mcp1::natR cdc13-117:cdc13-GFP:LEU2 sid4-TdTomato:hygR h<sup>-</sup></i>	this study
<b>Figure EV2C</b>		
JM2794	<i>ade6-M210 + Ch16(ade6-M216) h<sup>-</sup></i>	J-P. Javerzat
JM6343	<i>klp6::ura4 ade6-M210 + Ch16(ade6-M216) h<sup>?</sup></i>	this study
JM7181	<i>mcp1::natR ade6-M210 + Ch16(ade6-M216) h<sup>-</sup></i>	this study
<b>Figure EV2D</b>		
JM2881	<i>dam1::kanR h<sup>+</sup></i>	lab stock
PR100	<i>h<sup>+</sup></i>	P. Russell
JM2943	<i>klp5::kanR h<sup>+</sup></i>	lab stock
JM7111	<i>mcp1::kanR h<sup>-</sup></i>	this study
<b>Figure EV2E</b>		
JM3253	<i>klp5::kanR h<sup>-</sup></i>	lab stock
JM2958	<i>dam1:hygR h<sup>+</sup></i>	lab stock
JM2115	<i>dis2::ura4 h<sup>+</sup></i>	M. Yanagida
JM8832	<i>bub3::hygR h<sup>+</sup></i>	lab stock
JM6523	<i>mcp1::natR h<sup>-</sup></i>	this study
<b>Figure EV2F</b>		
JM9441	<i>klp5-mNeonGreen:kanR klp6-mNeonGreen:hygR natR:Z:adh15-mCherry-atb2 h<sup>+</sup></i>	this study
JM9877	<i>mcp1::natR klp5-mNeonGreen:kanR klp6-mNeonGreen:hygR natR:Z:adh15-mCherry-atb2 h<sup>?</sup></i>	this study
<b>Figure EV2G</b>		
JM7129	<i>mcp1-GFP:hygR natR:Z:adh15-mCherry-atb2 h<sup>-</sup></i>	this study
<b>Figure 2A</b>		
JM9513	<i>klp5-mNeonGreen:kanR klp6-mNeonGreen:hygR natR:Z:adh15-mCherry-atb2 cut11-mCherry:ura4 h<sup>?</sup></i>	this study
JM3010	<i>tea2-GFP:kanR h<sup>-</sup></i>	P. Nurse
JM8007	<i>tea2-GFP:kanR natR:Z:adh15-mCherry-atb2 h<sup>-</sup></i>	this study
<b>Figure 2B-C</b>		
JM8007	<i>tea2-GFP:kanR natR:Z:adh15-mCherry-atb2 h<sup>-</sup></i>	this study
JM8070	<i>klp6::hygR tea2-GFP:kanR natR:Z:adh15-mCherry-atb2 h<sup>?</sup></i>	this study
JM8053	<i>mcp1::hygR tea2-GFP:kanR natR:Z:adh15-mCherry-atb2 h<sup>?</sup></i>	this study
<b>Figure 2D</b>		
JM9441	<i>klp5-mNeonGreen:kanR klp6-mNeonGreen:hygR natR:Z:adh15-mCherry-atb2 h<sup>+</sup></i>	this study
JM2895	<i>tea2::his3 his3-D1 h<sup>-</sup></i>	P. Nurse
JM9961	<i>tea2::his3 klp5-mNeonGreen:kanR klp6-mNeonGreen:hygR natR:Z:adh15-mCherry-atb2 his3-D1 h<sup>?</sup></i>	this study
<b>Figure 2E-F</b>		

JM8018	<i>leu1-32::SV40-GFP-atb2:leu1 h<sup>-</sup></i>	F. Chang
JM8427	<i>klp5::natR klp6::hygR leu1-32::SV40-GFP-atb2:leu1 h<sup>-</sup></i>	this study
JM8334	<i>mcp1::kanR leu1-32::SV40-GFP-atb2:leu1 h<sup>+</sup></i>	this study
JM8952	<i>tea2::his3 leu1-32::SV40-GFP-atb2:leu1 his3-D1 h<sup>+</sup></i>	this study
JM9439	<i>tea2::his3 klp5::natR klp6::hygR leu1-32::SV40-GFP-atb2:leu1 his3-D1 h<sup>?</sup></i>	this study
JM9202	<i>tea2::his3 mcp1::hygR leu1-32::SV40-GFP-atb2:leu1 his3-D1 h<sup>+</sup></i>	this study
<b>Figure EV3A</b>		
JM9993	<i>tea2-GFP:kanR natR::Z:adh15-mCherry-atb2 cut11-mCherry:ura4 h<sup>?</sup></i>	this study
JM8070	<i>klp6::hygR tea2-GFP:kanR natR::Z:adh15-mCherry-atb2 h<sup>?</sup></i>	this study
JM8053	<i>mcp1::hygR tea2-GFP:kanR natR::Z:adh15-mCherry-atb2 h<sup>?</sup></i>	this study
<b>Figure EV3B</b>		
JM8018	<i>leu1-32::SV40-GFP-atb2:leu1 h<sup>-</sup></i>	F. Chang
JM8334	<i>mcp1::kanR leu1-32::SV40-GFP-atb2:leu1 h<sup>+</sup></i>	this study
JM2893	<i>tip1::kanR h<sup>-</sup></i>	P. Nurse
JM8392	<i>tip1::kanR leu1-32::SV40-GFP-atb2:leu1 h<sup>-</sup></i>	this study
JM8462	<i>mcp1::hygR tip1::kanR leu1-32::SV40-GFP-atb2:leu1 h<sup>?</sup></i>	this study
JM2894	<i>tea1::ura4 h<sup>-</sup></i>	P. Nurse
JM8929	<i>tea1::ura4 leu1-32::SV40-GFP-atb2:leu1 h<sup>+</sup></i>	this study
JM8944	<i>mcp1::hygR tea1::ura4 leu1-32::SV40-GFP-atb2:leu1 h<sup>+</sup></i>	this study
<b>Figure EV3C-D</b>		
JM9513	<i>klp5-mNeonGreen:kanR klp6-mNeonGreen:hygR natR::Z:adh15-mCherry-atb2 cut11-mCherry:ura4 h<sup>?</sup></i>	this study
JM9961	<i>tea2::his3 klp5-mNeonGreen:kanR klp6-mNeonGreen:hygR natR::Z:adh15-mCherry-atb2 his3-D1 h<sup>?</sup></i>	this study
<b>Figure EV3E</b>		
JM9513	<i>klp5-mNeonGreen:kanR klp6-mNeonGreen:hygR natR::Z:adh15-mCherry-atb2 cut11-mCherry:ura4 h<sup>?</sup></i>	this study
JM10020	<i>mcp1::natR tea2::his3 klp5-mNeonGreen:kanR klp6-mNeonGreen:hygR natR::Z:adh15-mCherry-atb2 his3-D1 h<sup>?</sup></i>	this study
<b>Figure 3A-B</b>		
JM9991	<i>tea2-GFP:kanR tip1-TdTomato:hygR h<sup>?</sup></i>	this study
<b>Figure 4A-B</b>		
JM9935	<i>klp5-mNeonGreen:kanR klp6-mNeonGreen:hygR tip1-TdTomato:hygR h<sup>?</sup></i>	this study
<b>Figure 4C</b>		
JM10018	<i>mcp1::natR klp5-mNeonGreen:kanR klp6-mNeonGreen:hygR tip1-TdTomato:hygR h<sup>?</sup></i>	this study
<b>Figure EV4</b>		
JM9935	<i>klp5-mNeonGreen:kanR klp6-mNeonGreen:hygR tip1-TdTomato:hygR h<sup>?</sup></i>	this study
<b>Figure EV5</b>		
JM10018	<i>mcp1::natR klp5-mNeonGreen:kanR klp6-mNeonGreen:hygR tip1-TdTomato:hygR h<sup>?</sup></i>	this study

All strains are *ura4-D18 leu1-32* unless otherwise stated.

**Appendix Table S2. List of oligonucleotides used in this study**

<i>Klp5.tagW</i>	CCTCATCTGGACACTATTGATCTGGAT
<i>Klp5.tagX</i>	GGGGATCCGTCGACCTGCAGCGTACGAGGTGGCTTTCTC TTCTTCGTT
<i>Klp5.tagY</i>	GTTTAAACGAGCTCGAATTCATCGATCGGATAAGCTTGAT GATATGA
<i>Klp5.tagZ</i>	GGTGGCAGACATATATGTACGCTT
<i>Klp6.tagW</i>	GTGGAAAAATCTTTAGATAAACATAAT
<i>Klp6.tagX</i>	GGGGATCCGTCGACCTGCAGCGTACGAAGCATTAGGAGT ATTCTCAGTCCCGCC
<i>Klp6.tagY</i>	GTTTAAACGAGCTCGAATTCATCGATGCTACTTATGCTACT TATGATTTAGCATTTCAT
<i>Klp6.tagZ</i>	GGTCAATCATTAAACCCGATTAAT
<i>Mcp1.delW</i>	AGCATTTTTTAACTATAATAAGC
<i>Mcp1.delX</i>	GGGGATCCGTCGACCTGCAGCGTACGAAATATCTTATGAT TTTAAACATTA AAAACAG
<i>Mcp1.delY</i>	GTTTAAACGAGCTCGAATTCATCGATTTTGTCTCTCGTTAC TCTACAATATTT
<i>Mcp1.delZ</i>	GGCTTAACGAATTCCTTGGATACCA
<i>Mcp1.tagW</i>	CCCAAAGGTGAATTTACTAATTCCTT
<i>Mcp1.tagX</i>	GGGGATCCGTCGACCTGCAGCGTACGATAGAAAAGTAGG TAGTTTAGAACCGGGGATCCGTCGACCTGCAGCGTACGA
<i>Mcp1.tagY</i>	GTTTAAACGAGCTCGAATTCATCGATATTAATGTATGAATA TAATGAAATAATTCGTAA
<i>Mcp1.tagZ</i>	CAATTTTCTTTGGTGTCTGACAAAGCGT
<i>Mcp1.pLys1U.FW</i>	AGTAGTCGACGTCCTCTTTTTTTTAAACGGGA
<i>Mcp1.pLys1U.RV</i>	TCATGGATCCCAAAGCACTTAAAATTA AAAAAA