Long et al. Supplementary Material

Supplementary Figure Legends

Supplementary Table 1. Primers used to design dsRNA.

Primers were used to generate double-stranded RNA (dsRNA) from S2R+ cells. RT-PCR produced 200-500 bp templates from total RNA extracted from a cell population and templates were reverse transcribed *in vitro* to produce dsRNA. All primers (unless noted by **) were designed using Snapdragon software and were generated including the T7 promoter sequence (TAATACGACTCACTATAGGG) at the 5' end. For primer sets denoted with **, primers were previously published and were generated as listed (see supplemental references for full citations.)

Supplementary Table 2. List of antibodies and primers used to validate dsRNA knockdown of candidate genes in *Drosophila* S2R+ cells

A. Antibodies used in Western blot validation of protein knockdown of candidate genes in S2 cells. Sources are listed for commercially available antibodies. For antibodies generated by individual research groups, the investigator name and original publication are referenced (see supplemental references for full citations.) **B.** Primer sets utilized for validation of candidate gene expression. Primers were designed using PrimerQuest (IDT DNA.) Quantitative real time PCR was performed using the Life Technologies 7900HT.

Movie 1. Control dsRNA treated cell.

Drosophila S2R+ cells were transfected with dsRNA for 72 hours. Twenty-four hours prior to plating, cells were transfected with EB1-GFP to mark the growing MT plus ends. Cells were plated on Concanavalin A for 1 hour to facilitate adhesion and spreading.

Images were collected of a single plane of focus using spinning disk confocal microscopy. Time-lapse image series were acquired for a period of 1 minute at a frame capture rate of every 750 ms using 400 ms exposure.

Movie 2. CLASP dsRNA treated cell.

Drosophila S2R+ cells were transfected with dsRNA for 72 hours. Twenty-four hours prior to plating, cells were transfected with EB1-GFP to mark the growing MT plus ends. Cells were plated on Concanavalin A for 1 hour to facilitate adhesion and spreading. Images were collected of a single plane of focus using spinning disk confocal microscopy. Time-lapse image series were acquired for a period of 1 minute at a frame capture rate of every 750 ms using 400 ms exposure.

Movie 3. Klarsicht dsRNA treated cell.

Drosophila S2R+ cells were transfected with dsRNA for 72 hours. Twenty-four hours prior to plating, cells were transfected with EB1-GFP to mark the growing MT plus ends. Cells were plated on Concanavalin A for 1 hour to facilitate adhesion and spreading. Images were collected of a single plane of focus using spinning disk confocal microscopy. Time-lapse image series were acquired for a period of 1 minute at a frame capture rate of every 750 ms using 400 ms exposure.

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Gene	Abbrev	CG #	Ortholog	Forward	Reverse	Source
4EHP		CG33100	eIF4E2	GGACGCATGTTGTGTTGTGT	TGGGTTTTTGGTGGATTCTC	DRSC41089
Abi		CG9749		CTTCCAGGAAAGCTCAGTGC	CCGTGGATTCTGTGTGTTTG	
Abl		CG4032		TAGCTACAACAAATCGGGGG	AGCTCGGCCAGAGTGTTAAA	
APC1		CG1451	APC	AGATACGCCAGCATTGCTCT	TCTTCGGACTTGCTCCTGTT	
Bifocal		CG1822		CCACAAAGGTAACGGTAAAACC**	TAATACGACTCACTATAGGGGCTGATTCTCTGGCTGTTCC	Goshima et al., 2007[1]
C3G		CG42328	Rapgef1	ATTATGCCCATGAGCAAGGA	CCCGTCCTCCTCTCTCTTCT	
Cadherin N	CadN	CG7100	N-cadherin	TAATACGACTCACTATAGGATTCCTCATCACTGCTCGCT**	TAATACGACTCACTATAGGTCGCAGTACTCGTCTTGGTG**	Kiger et al., 2003[2]
Calmodulin	Cam	CG8472	Calmodulin	TGACTTCCCTGAATTCCTTACC	TAATACGACTCACTATAGGGGTCGCCATCGATATCAGCC	Goshima et al., 2007[1]
Cappuccino	Capu	CG3399		TAATACGACTCACTATAGGAATGAAATGGAGCAAGGTGG**	TAATTAACCCTCACTAAAGGGCGAAATGTGGGCATAATCT**	Kiger et al., 2003[2]
Capulet	Capt	CG33979	Formin	AGAGCTGAGCCAATACC	ATAAGTGACCTTCTTCA	
CG17272		CG17272		CATTTGGCCAACTTTATTGTGC	TAATACGACTCACTATAGGGACGATCCAAGAACTGGTTTCC	Goshima et al., 2007[1]
CG31957		CG1957	eIF1AD	TTTCGATGCCCAACAAATTC	TAATACGACTCACTATAGGGTTCAGTCCTCTTCGCTGGAT	
Clip190		CG5020	Clip170	GGCAAGGAGAATCTTTGGCAG	GAACTGCTTTTGCTGAAGGGA	
Disabled		CG9695	Dab	TAATACGACTCACTATAGGACAAGCAGCGGATAACCATC**	AATTAACCCTCACTAAAGGTGGGTAACGAACCTCTTTGG**	Kiger et al., 2003[2]
eIF2B		CG4153	eIF2B4	AGGACAACAGCTCAACCT	AGGTTGTTTTTGCTCGTATG	
eIF3-S10		CG9805	eIF3A	GAGGCTGAGGACGAAAAGC	CAGTTCCCTCCAGTTTCCC	
eIF4E		CG4035		AGCGCCCCCAGCACC	TCTTCTTGAATAGCGAGTAGT	DRSC11342
Enabled	Ena	CG15112		TACCACCAGCAACAGCATC	GTTCGAGGGCGAAACGC	
Fax		CG4609		TACCTGGACTCGGGACTCA	CACTTGTCCTTCATGCGAGA	
Fmr1		CG6203	Fxr1	CGTGCCCGAGAGTATGAAAT	ATTGTGCGCTGAAACTCCTT	
Fps85D		CG8874	FER	TCTCGTTGACTTGGAGCACA	TCTCTGGCTCTGGAGTTGGT	DRSC39364
GFP				AGAGATGTTAATGGGCACAAATTTTCT	AGATTTGTATAGTTATCCATGCCATG	
Jaguar	Jar	CG5695	Myosin 6	AGACCACCTAATCAAATATAGTTATATTTAC	AGACCACTCAGATCCGAAAATCTTCGAGCCC	Petritsch et al., 2003[3]
Karst		CG12008	Beta spectrin	TAATACGACTCACTATAGGTTTACACCGATGCCAATGAA**	TAATTAACCCTCACTAAAGGACGCTATCTGCGTTCTCGTT**	Kiger et al., 2003[2]
Klarsicht	Klar	CG170469		CAGGGCACTCTCACTGATC	AGCACCAGCCCTCGAC	Sepp et al., 2008[4]
Klp10A		CG1453	KIF2A	ATGGACATGATTACGGTG	CATCGATCTCCTTGCGATT	
Krasavietz	Kra/ Exba	CG2922		GCGAATGGAACAAGAAGGAG	CTGACTGAAGCCATTCGACA	
LAR		CG10443		ATGGGTCTGCAGATGACAG	CCACGGCGAGTGGATTG	DRSC 01707
Mekk1		CG7717	Map3K4	AAATCACCCAGCACTTGGAC	CGCCCTTATATGCACCTTGT	

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Minispindles	Msps	CG5000	TOG	GATCCAAGGTGAATCATAATGCC	GAGACAATGAGGACGATGATGG	
NCD		CG7831	KIFC1	GCTCTAAGCACAGAAGTGGTGC	CCATTCGAATCTCCATGTCC	
Orbit/MAST	CLASP	CG32435	CLASP	ATCAGCACCACCACAAACAA	TGTGAGAAGTGAGGATTGCG	
P150glued		CG9206	Dynactin 1	AGACCACCAGCGATTCAGTGC	AGATTCTTCTCCGCTAGATCC	
P190RhoGap		CG32555		TAATACGACTCACTATAGGGAAGAAGTCCTTCAGTGCCG**	TAATTAACCCTCACTAAAGGAAACCAGAGCCATGTGTTCC**	Kiger et al., 2003[2]
Par1		CG8201	MARK3	GGTTCGCCTAACATGCAAAT	CTCCTTCTCCTTCATGCGTC	DRSC 31375
Peanut		CG8705	Septin7	CGCCTCCAACGG	TCCTGAAGGTGC	Somma et al., 2002[5]
Phospholipase D	Pld	CG12110	PLD3	CGTCGTAATCGACCAAACCT	CTTCATCGCGTTGTTCTTCA	DRSC 38086
PlexinA		CG11081	PlexinA2	ATTGTAGTCATTAAACTCTCGG	TCTAATACGACTCACTATAGGGATACATTCCAACCAAAAACAG	DRSC17220
Pod-1		CG4532	Coronin7	TTCTTTGACTTCGCCTGGAGT	CCTTGCCCGTAACAAACAGT	
PP2A-B'		CG7913	PP2R5c/d	GAGTGCCGCCGGTCAC	TAATACGACTCACTATAGGGGATAACGAGGCGTTAGATCC	DRSC16337
Puckered	Puc	CG7850		TATAGAAACACACCCCGCCT	TCGTAGGCCTCCTGGAGTA	DRSC 31024
Rab5		CG3664	Rab5A	TAATACGACTCACTATAGGGTCCTGGCCAGCCGTGT**	TAATACGACTCACTATAGGGGCAACCACTCCACGCA**	Kiger et al., 2003[2]
Rapgap1		CG34374	Rap1gap	ACCACCGAGGAGGAACTTTT	GTCGGGTGAGAATGGAGTGT	
RfC38		CG6258	RfC3	GACGCATCAAATTGAGATTAGT	CGTGAGTAGCTCGTACAG	DRSC 01997
Rhogef2		CG9635		ATGGATCACCCATCAATCAAAAAACGG	TGTCCCGATCCCTATGACCACTAAGGC	Rogers et al., 2004[6]
Roundabout	Robo	CG13521	Robo2	CACTATTCATGGACCCCACC	GCAGGTCTCACTGGAAGAGG	
Roughened	Rap1	CG1956	Rap1A	AAAGCCCAACCAACAACAAG	TTGATGCAACTGATCGTGGT	
SF2		CG6987	SFRS	CGATCGGAAGCGAGAGT	TAATACGACTCACTATAGGGCGACGATGCGGTGAAG	DRSC16845
Shortstop	Shot	CG18076	MACF1	CCACGGAGTACCATCAGTT	CAGCCCTTAACCACACGAAT	
Su{dx}		CG4244	Ubiquitin ligase	TTCTCCCAGCCATCTGGTA	TAATACGACTCACTATAGGGACACAACCAGCTGCTGTTT	DRSC00791
Syndecan	Sdc	CG10497		CACTCTGCGTATGTGGGTGT	CCTGCTCTTTCTGCTTTTGC	
TACC		CG9765		AGTGAGAAGGAGCAGCAAGC	CATCTTGTCGTAGCGCTGT	DRSC36042
Tra2		CG10128	Tra2a/b	GATAGTTGTCATAGGGCGAAGC	TAATACGACTCACTATAGGGACTTCTGACATACAGACACAGCG	Goshima et al., 2007[1]
Tribbles	Trbl	CG5408	Trib	TGACAGATCTGGTGGAATATGG	TAACAGTAGCGGTCAAAACAGC	Goshima et al., 2007[1]
Wallenda		CG8789	MapKKK 12/13	AGTGGCAGGCTAAAGAACGA	GCTTGAGAGAGTTGTTGCCC	
Wasp		CG1520	WASL	CAAACGACAAGAGAAACGCA	CCTGCCTTCACGAAGAACTC	
Zipper		CG15792	Myosin heavy chain 10	TTCAGCTTGGCCAGGTGT	CGCGAGAAGCTTGACAC	Sepp et al., 2008[4]

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A.

	Citation	Source and catalog #	Notes
Abi	Lin et al., 2009[7]		Generous gift of J.L. Juang (NHRI)
Abl	Wills et al., 2000[8]		
APC		Santa Cruz Biotechnology (sc15803)	
CadN		DSHB (DN-Ex#8)	
Capulet	Wills et al., 2000[8]		
Clip190	Lantz and Miller, 1998[9]		Generous gift of Kathryn Miller (Wash. Univ)
Disabled		DSHB (P4D11)	
eIF4E	Lachance et al., 2002[10]		Generous gift of Paul Lasko (McGill Univ.)
eIF3-S10		Abnova (ABVAPB6VV)	
Ena			
Fmr1		Abcam (ab10299)	
Jar	Kellerman and Miller, 1992[11]		Generous gift of Kathryn Miller (Wash. Univ)
Karst	Zarnescu and Thomas, 1999[12]		Generous gift of Graham Thomas (Penn State Univ.)
Klp10A	Rogers et al., 2004[13]		Generous gift of David Sharp (Albert Einstein)
LAR	Sun et al., 2001[14]		Generous gift of Kai Zinn (UCSF)
Msps	Lee et al., 2001[15]		Generous gift of Jordan Raff (Univ. of Oxford)
NCD	Hatsumi and Endow, 1992[16]		Generous gift of Sharon Endow (Duke Univ.)
Orbit (CLASP)	Lee et al., 2004[17]		
Peanut		DSHB (4C9H4)	
Pod-1	Rothenberg et al., 2003[18]		Generous gift of Yuh Nung Jan (UCSF)
PP2A-B'		Millipore (07-1221)	
Rab5		Abcam (ab21261)	
Robo		DSHB (13C9)	
Shot	Lee et al., 2000[19]		Generous gift of Peter Kolodziej (Vanderbilt Univ.)
TACC	Gergely et al., 2000[20]		Generous gift of Jordan Raff (Univ. of Oxford)

B.

	Foward Primer	Reverse Primer
4EHP	ACCCGGAAAGGATAAGGCACAGGA	TGTTGTGTGTGTGTGTGTGTGTGG
Bifocal	TCACAGAACCGGCGACAATCAGTA	ACCTCCAGTGGATCGGAATTGGT
C3G	TGTGCGGCGAATGGAAAGCTATTG	TGCCTGCCCGATTGTGGTAAATTG
Cam	ACCAGCAGCAACAAACAAGAGTCG	TTCTTGCTTCTCCCTCTGCTTGCT
Capu	CAAATCGATGGCCAAGCTGCATCA	TGAGAAGTGGTAGAAGCGCATGGT
CG17272	ATCCGCAGAATAAGGGCACCATCT	ATTGTTGTTCACATTGGCCTCCCG
eIF1AD (CG31957)	ACGTTCCTTGTTTCGATGCCCAAC	TCTCGGCTTTGACTTTATCGCCCT
eIF2B	ACCGCGATTACACATACGACGAGT	TCGCATCACGAACTTTGGCTTTCG
Fax	TGCCATGCTGGAGAACCATCTCAT	AGTTGACCTTGTAGCCCTTGAGCA
Fps85D	ATGTGAATGCCATCGAAAGTGCCG	AAGACAGGGAACGCTCTTGGAGT
Klarsicht	CACGCGATTGTCTTGCACCTCAAA	AAATCCGTTAGGGTGGCTGTACCA
Kra	AAGTCATCCAGAAGAAGCAGCCCA	TGGTCTTGATGCGTTGACCCGATA
Mekk1	GCGCGTGCTTACTAAAGTGCAAGT	TTGTTCGCACTCTTCGCCTGTTTG
P150glued	TAAAGCAGCGCAGGAACGAGGATA	AACTCCTTCTGAAGCGAAGCCTGA
P190RhoGap	AGACGTGTTTGTCGTCGGAACTGA	TACACTCATCCGATTTGCCTGGGT
Par1	TCGACTTAGAATGGCATCGGCTGT	GCTGTGTTGTTTCGTGCTCTGGTT
Phospholipase D	CTGCGGTTCGGCAAACATTAACGA	TCCGCTCGGATACTTCTTGCCATT
PlexinA	GCTTTGCCGAACTCCAAACTGAA	AGAAGCTCAGGACGTTCCCATTGT
Puc	GAGCGCGTGCATATGTGTGTGAAT	GCGCTTCTCGCTTTATCCGCATTT
Rap1	ACACACACACACATTCGCAACCAG	TTTGTGGCTCTCTCTGCACTTCCT
Rapgap1	ACCACAGACTACCTGGATGGCAAA	TCCCAACTGCTCATCCAGTCCAAT
Rfc38	ACTTCCACAAGGATCAAGCGGAGA	CACATGATGCGTGTCTTCTTGCCA
Rhogef2	TGTCGAGGTCAACCAGGCAGTAAA	TGATCGTCAGATTGCCGTCATGGA
Sdc	ACAAGTGCAGGAACAAAGTGTGCG	CAGTTGAGCCAAACCAACCCAGT
SF2	GGCCATTTGTCATTGTGAGCGTGA	ACTTTGCCGAACTTGTGGAACAGG
Su{dx}	AGCACTCACACACTCAGCCACTAA	AAGCCTCCTCGATTGTCACGCTTA
Tra2	TCTCCGCAACTACGTCGAACTTCA	TCGTATGCTCCTTTACAGCCCGTT
Trbl	AACTATTCGTCACCAGTCTCGCCA	ATGGTCTGCAGCATCTTGGCATTG
Wallenda	AGAGAGCGGATTGGAACTCGGTTT	TTCTGCTCTTCTTGGAGTTCCGCT
Wasp	TTTCTGCTTACCTTCGAGGGCAGT	CCGGTTGCGTTTCTCTTGTCGTTT
Zipper	AACAGATCGCCCAAGAGCGTGATA	TGGAGGGTCTTTCGCTTGTTCTCA

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