Supplemental Materials

Molecular Biology of the Cell

Gache et al.

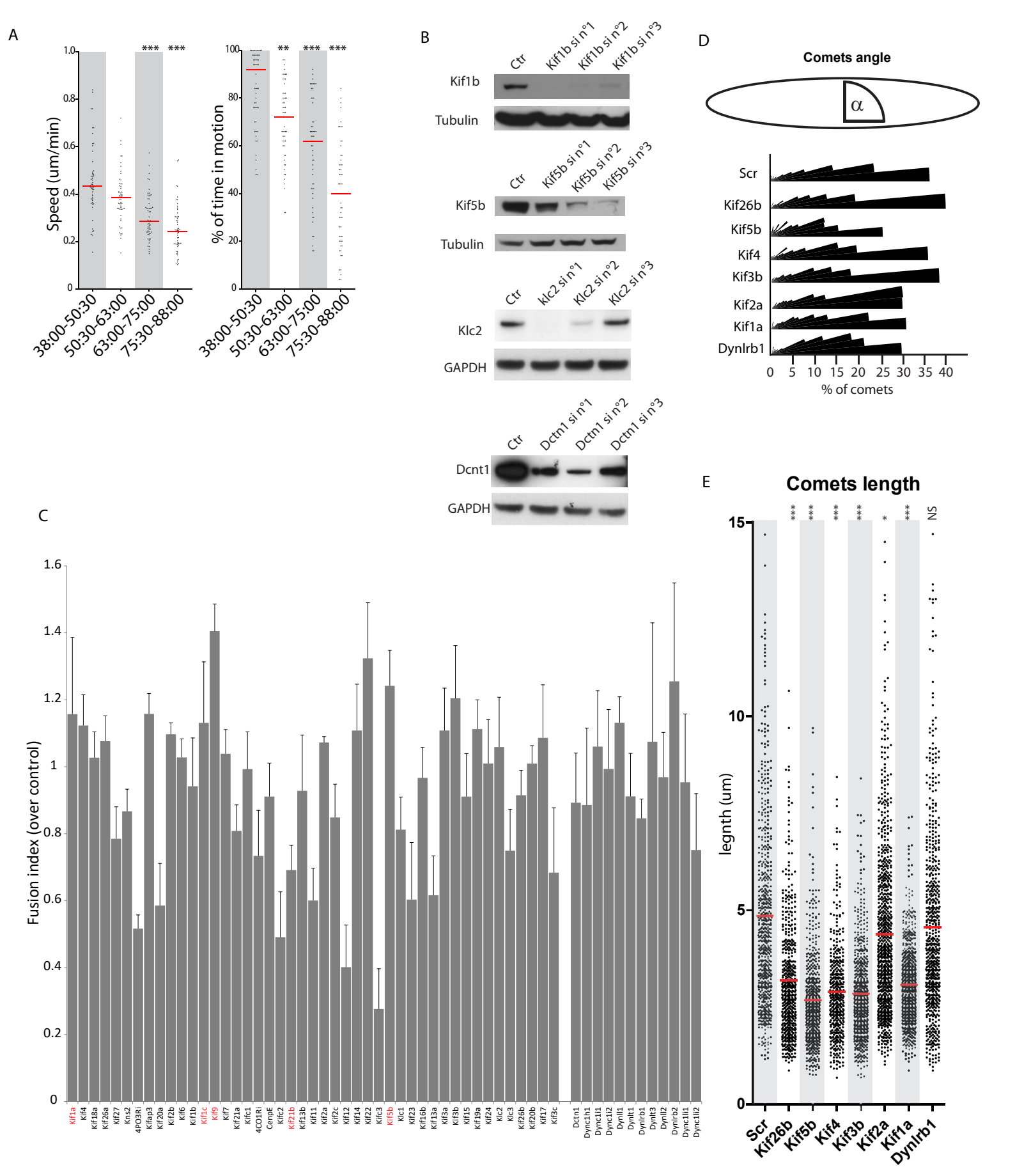
A
Speed (um/min)
% of time in motion
38:00-50:30
50:30-63:00
63:00-75:00
75:30-88:00
B
Ctr
Kif1b si n°1
Kif1b si n°2
Kif1b si n°3
Kif1b
Tubulin
Kif5b si n°1
Kif5b si n°2
Kif5b si n°3
Kif5b
Klc2 si n°1
Klc2 si n°2
Klc2 si n°3
Klc2
GAPDH
Dctn1 si n°1
Dctn1 si n°2
Dctn1 si n°3
Dcnt1
D
Comets angle
α
Scr
Kif26b
Kif5b
Kif4
Kif3b
Kif2a
Kif1a
Dynlrb1
% of comets
E
Comets length
legnth (um)
NS
C
Fusion index (over control)

Fig S1

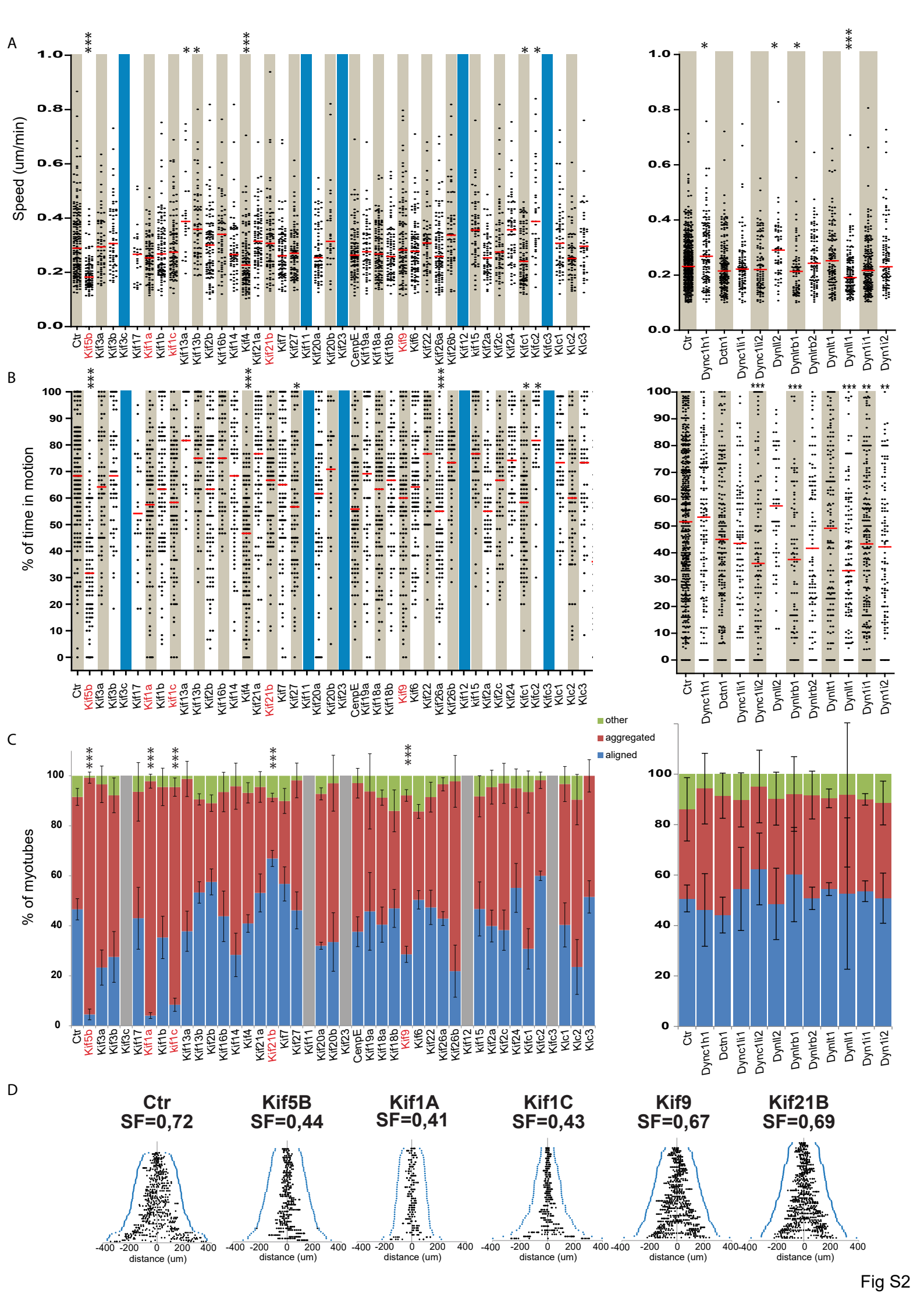
A
Speed (um/min)
B
% of time in motion
C
% of myotubes
other
aggregated
aligned
Ctr Kif5b Kif3a Kif3b Kif3c Kif17 Kif1a Kif1b kif1c Kif13a Kif13b Kif2b Kif16b Kif14 Kif4 Kif21a Kif21b Kif7 Kif27 Kif11 Kif20a Kif20b Kif23 CenpE Kif19a Kif18a Kif18b Kif9 Kif6 Kif22 Kif26a Kif26b Kif12 kif15 Kif2a Kif2c Kif24 Kifc1 Kifc2 Kifc3 Klc1 Klc2 Klc3
Ctr Dync1h1 Dctn1 Dync1li1 Dync1li2 Dynll2 Dynlrb1 Dynlrb2 Dynlt1 Dynll1 Dyn1i1 Dyn1i2
D
Ctr SF=0,72
Kif5B SF=0,44
Kif1A SF=0,41
Kif1C SF=0,43
Kif9 SF=0,67
Kif21B SF=0,69
distance (um)

Fig S2

Supplementary Figure 1. Interfering with nuclear movement inside myotubes.

A: Nuclear speed and percentage of time in motion during four successive intervals after differentiation. At least 57 nuclei were monitored from 3 different experiments. **B:** Expression levels after silencing of Kif1b, Dctn1, Kif5b and Klc2. **C:** Fusion index over control after depletion of the indicated motors. Average of three independent experiments is represented. **D:** Angles of EB1 comets were measured compared to the longest myotube axis. All angles were averaged on a 90° quadrant to facilitate reading, and sampled every 5°. **E:** The length of EB1 comets was measured over a period of 200s.

Supplementary Figure 2. Screening for nuclei behavior inside myotubes

Speed **(A)**, Time in motion **(B)** and nuclear distribution **(C)** of nuclei inside myotubes from differentiated GFP-H1-C2 cells in non-treated (ctrl), myotubes treated with 50 nM siRNA targeting kinesin and dynein members. Red line indicates the median. In average 110 nuclei were monitored from three different experiments. Nuclear distribution is quantified as "aligned" if >70% of nuclei are aligned along the same axis; "aggregated" if >70% of nuclei do not align along the same axis; "other" if nuclei are both aggregated and aligned in the same myotube. Silencing of kinesins required for cell cycle progression was not analyzed as their depletion induced cell death before differentiation was induced (blue bars). **D:** Representation of nuclei spreading in several myotubes after siRNA for the indicated proteins with the corresponding spreading factor. Each horizontal line represents a myotube, with the extremities depicted as blue dots. Each nucleus is a black dot.

Supplementary movie 1

Phase contrast time-lapse movies of myotubes in presence of 75 nM Nocodazole, or 100 nM Taxol or 200nM Jasplakinolide or 10 nM cytochalasine D as indicated.

Table1 Listing of siRNA sequences and Taqman probes used for each molecular motor.

RefSeq Accession Number	Gene Symbol	Sense siRNA Sequence	Antisense siRNA Sequence	Taqman Assay ID
NM_008440	Kif1a	GGACAUCAACUAUGCCUCUtt	AGAGGCAUAGUUGAUGUCCtc	Mm00492863_m1
		GGAAACAGAGAAGAUCAUUtt	AAUGAUCUUCUCUGUUUCCtt	
		CCAAGUCCUUCAUCGAAUAtt	UAUUCGAUGAAGGACUUGGtc	
NM_207682	Kif1b	CGGGCUGAUUCAACUGGUGtt	CACCAGUUGAAUCAGCCCGtt	Mm00801827_m1
		CCUCAAUGAAGACCCAUUAtt	UAAUGGGUCUUCAUUGAGGtt	

		GGAUGGAAUUACAAGGGUUtt	AACCCUUGUAAUUCCAUCCtt	
NM_153103	Kif1c	CCUUCGACUAUUCUUACUGtt	CAGUAAGAAUAGUCGAAGGtg	Mm00462184_m1
		GGAAACAGAGAAGAUCAUAtt	UAUGAUCUUCUCUGUUUCCtg	
		CCAUGUUUUCCGCUUCAAUtt	AUUGAAGCGGAAAACAUGGtt	
NM_008442	Kif2a	GGGAAUUUAUGCAUUAGCAtt	UGCUAAUGCAUAAAUUCCCtt	Mm00515233_m1
		CGCAGAUCAAUUUUCAUAGtt	CUAUGAAAAUUGAUCUGCGtt	
		GCUCCUAAUGAAAUGGUUUtt	AAACCAUUUCAUUAGGAGCtg	
NM_134471	Kif2c	GGAGGUACCACAAAAGGCAtt	UGCCUUUUGUGGUACCUCCtt	Mm00728630_s1
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		GCAGAAAUUAACAAGUCUCtt	GAGACUUGUUAAUUUCUGCtc	
NM_008443	Kif3a	GGGCGACACAAGGUUUUUGtt	CAAAAACCUUGUGUCGCCCtc	Mm01288585_m1
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		CCGUAAUUGAUUCUUUACUtt	AGUAAAGAAUCAAUUACGGtc	
NM_008444	Kif3b	GGAUUUGUCUUCUUUUGUCtt	GACAAAAGAAGACAAAUCCtt	Mm00492891_m1
		GGUGGUAGAUGCGGAUGUGtt	CACAUCCGCAUCUACCACCtt	
		GGGUUUCAAUGGCACAAUUtt	AAUUGUGCCAUUGAAACCCtg	
NM_008445	Kif3c	GGAGAAUCCUGAAACAGGGtt	CCCUGUUUCAGGAUUCUCCtt	Mm00492900_m1
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		CCGGCUAUCUUUGAGAUGGtt	CCAUCUCAAAGAUAGCCGGtg	
NM_008446	Kif4	GGUGGUGGUUGGUAAUGAUtt	AUCAUUACCAACCACCACCtg	Mm00492908_m1
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		GGAUACAAUGCAACUGUCCtt	GGACAGUUGCAUUGUAUCCtt	
NM_177052	Kif6	CCUGGCAGAUGGAUUCGUAtt	UACGAAUCCAUCUGCCAGGtc	Mm00723857_m1
		GCUUCAACCCGGUCACACUtt	AGUGUGACCGGGUUGAAGCtt	
		CGAAUGUGGCUAUGACCUGtt	CAGGUCAUAGCCACAUUCGtt	
NM_010628	Kif9	GGACUUGGUUUAUGAAACAtt	UGUUUCAUAAACCAAGUCCtg	Mm00495130_m1
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		GGCUUGUCAGUGCAUCUCAtt	UGAGAUGCACUGACAAGCCtt	
NM_010615	Kif11	CCAUUUAAUCUGGCAGAGCtt	GCUCUGCCAGAUUAAAUGGtc	Mm01204225_m1
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		GGUCUACUGAUAUAAUCAAtt	UUGAUUAUAUCAGUAGACCtc	
NM_010616	Kif12	CCUGGCUAUUAGAUCGCGUtt	ACGCGAUCUAAUAGCCAGGtg	Mm00802885_m1
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		CCUUCACCUGGCUAUUAGAtt	UCUAAUAGCCAGGUGAAGGtc	
NM_010617	Kif13a	GGUAUCGUAUAUGGAGAUCtt	GAUCUCCAUAUACGAUACCtc	Mm00660179_m1
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		GCUGGAGAAUAAGCUAAUUtt	AAUUAGCUUAUUCUCCAGCtt	
NM_010620	Kif15	GGAGUCUGUAUUCUCAACAtt	UGUUGAGAAUACAGACUCCtg	Mm01341275_m1
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		GCUCCAAUCACUACGAGAATT	UUCUCGUAGUGAUUGGAGCTT	
NM_010626	Kif7	GGAGAACGGCUCAAAGAGATT	UCUCUUUGAGCCGUUCUCCGG	Mm01320530_m1
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		CCAUCUCCCAUGGUGGUUATT	UAACCACCAUGGGAGAUGGAG	
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NM_175214	Kif27	GGAUCUCUACUUCUAUAAGtt	CUUAUAGAAGUAGAGAUCCtt	Mm00723524_m1
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NM_001013380	dync1li2	GCAGGUUAAGUAGCUGACAtt	UGUCAGCUACUUAACCUGCtt	Dync1li2
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