

Supplemental Materials

Molecular Biology of the Cell

Vasudevan et al.

Video - 13BC-KO cells grown with shaking move slower than wildtype cells

Wildtype (WT) and double knockout (13BC-KO) cells are grown with shaking at 160 rpm and a video is recorded for 10 sec at a maximal speed by Axiocam HS camera mounted on a Nikon Observer.A1 microscope with 10X lens and are shown side by side.

Video - Wildtype cells increase their motility upon treatment with IBMX

Video of wildtype cells without or with IBMX (1 mM, 20 min) recorded under a dissecting microscope with 40X magnification for 10 seconds using Moticam 480 digital camera.

Video - 13BC-KO cells increase their motility upon treatment with IBMX

Video of 13BC-KO cells without or with IBMX (1 mM, 20 min) recorded under a dissecting microscope with 40X magnification for 10 seconds using Moticam 480 digital camera.

Video - Cilia of wildtype cells have an apparently normal waveform

High-speed video recording of motile cilia in a wildtype cell acquired at 500 frames/s by a Photronics 1280 PCI FastCam on a Nikon Eclipse E600 microscope and replayed here at 30 frames/s.

Video - Cilia of wildtype cells have a less coordinated waveform

High-speed video recording of motile cilia in a 13BC-KO cell acquired at 500 frames/s by a Photronics 1280 PCI FastCam on a Nikon Eclipse E600 microscope and replayed here at 30 frames/s.

Video - GFP-Kin13Bp localizes to basal bodies, cortical microtubules and subset of cilia in 13BC-KO rescue B cell

Video of 13BC-KO cell rescued with GFP-Kin13Bp transgene (13BC-KO rescue B) captured using a TIRF system home-built around a Nikon eclipse Ti-U inverted microscope equipped with a 60 × NA 1.49 TIRF objective. GFP-Kin13Bp is seen to localize to the basal body, contractile vacuole pore, cortical microtubules and cilia (marked with red arrow at 9 s mark)

Video - GFP-Kin13Cp localizes to basal bodies, cortical microtubules and subset of cilia in 13BC-KO rescue C cell

Video of two 13BC-KO cells rescued with GFP-Kin13Cp transgene (13BC-KO rescue C) captured using a TIRF system home-built around a Nikon eclipse Ti-U inverted microscope equipped with a 60 × NA 1.49 TIRF objective. GFP-Kin13Cp localizes to the basal body and contractile vacuole pore in the first cell whereas in the second cell,

GFP-Kin13Cp localizes to the basal body and cilia (marked with red arrow on 6 - 9 s mark)

Video - GFP-Kin13Bp localizes to the basal body, cortical microtubules and in assembling cilia of regenerating 13BC-KO rescue B cell

Video of 13BC-KO rescued with GFP-Kin13Bp cell that was deciliated and allowed to regenerate for 0, 30 and 100 minutes captured using a TIRF system home-built around a Nikon eclipse Ti-U inverted microscope equipped with a 60 × NA 1.49 TIRF objective.. GFP-Kin13Bp is seen to localize to the basal body, and cortical microtubules at 0 min whereas at 30 min, GFP-Kin13Bp is seen to localize to the basal body, cortical microtubules and in assembling cilia (marked with red arrows) and at 100 min, GFP-Kin13Bp localizes to the basal body and cortical microtubules but not in cilia.

Video - GFP-Kin13Bp localizes to the basal body, cortical microtubules and in assembling cilia of regenerating 13BC-KO rescue C cell

TIRF video of 13BC-KO rescued with GFP-Kin13Cp cell that was deciliated and allowed to regenerate for 0, 30 and 100 minutes captured using a TIRF system home-built around a Nikon eclipse Ti-U inverted microscope equipped with a 60 × NA 1.49 TIRF objective. GFP-Kin13Cp is seen to localize to the basal body, and cortical microtubules at 0 min whereas at 30 min, GFP-Kin13Cp is seen to localize to the basal body, cortical

microtubules and assembling cilia (marked with red arrows) and at 100 min, GFP-Kin13Cp localizes to the basal body and cortical microtubules but not in cilia.

Homologs of kinesin-13 in *Tetrahymena thermophila* are well conserved

Multiple sequence alignment of members of kinesin-13 family from *Tetrahymena thermophila* (KIN13A, KIN13B and KIN13C), *Homo sapiens* (KIF2A, KIF2B and MCAK), *Mus musculus* (KIF2A, KIF2B and MCAK), *Xenopus laevis* (KIF2A and MCAK), *Giardia intestinalis*, *Leishmania major* (KIN13-2), *Trypanosoma brucei* (KIN13-2) and *Chlamydomonas reinhardtii* (CrKIN13) using ClustalW (1.8.3) tool.

Loss of Kin13Ap causes defects in the nuclear composition

(A) Confocal immunofluorescence images of cells labeled with a mixture of anti- α -tubulin mAb (12G10) and anti-polyglycylation antibodies (polyG) (red), and DAPI (blue) for DNA. Bar = 20 μ m. (B) A graph showing the nuclear organization in wildtype and 13A-KO cells. N = 50 cells. Abbreviations: mic, micronucleus; mac, macronucleus.

13BC-KO cells swim slowly, have reduced rates of phagocytosis and outer doublet microtubules of 13BC-KO cells have a normal density of outer dynein arms

Paths traced by cells swimming for 10 seconds of wildtype cells grown without (A) or with shaking (B) and 13BC-KO cells grown without (C) and with shaking (D). (E) A

graph showing the percentage of cells that have taken up India Ink after 20 min and 60 min. (F) The axonemes of wildtype and 13BC-KO cells were reactivated with 1mM ATP, negatively stained with uranyl acetate and viewed in TEM. Bar = 0.2 μ m

Cells lacking Kin13Bp and Kin13Cp have a normal ultrastructure of cilia

Images of wildtype and 13BC-KO cells obtained using transmission electron microscopy showing cross-sections of the middle segments of cilia (A and B), cross-section of the distal segments with singlet peripheral microtubules (C and D), longitudinal sections of axonemes (E and F), and cross-sections of oral ciliary membranelles (G and H). Bar = 20 μ m.

Coomassie gel image showing the amount of loading for total cilia, axoneme and M+M fractions

Total cilia, axoneme and M+M fractions of wildtype and 13BC-KO cells loaded onto 10% SDS-PAGE gels stained with Coomassie blue.

Deletion of MEC17, KIN13B and KIN13C is synthetically lethal

(A). Immunofluorescence images of progeny recovered from mating of either wildtype strains or knockout heterokaryons, fixed at different time points after the induction of mating and labeled with an anti- α -tubulin antibody 12G10 and anti-acetyl-K40 α -tubulin

antibody 6-11 B-1. Note that by 48 hr the triple knockout cells have lost most their cilia.

(B). TIRFM images of the triple knockout cells lacking KIN13B, KIN13C and MEC17 that were rescued by introduction of either GFP-KIN13B or GFP-KIN13C. Arrowheads mark cilia. Abbreviations: bb, basal body; cvp, contractile vacuole pore. (C) Viabilities of progeny of double and triple knockout heterokaryons (more details in Table 1 in the main text).

CLUSTAL W (1.83) multiple sequence alignment

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T.THERMOPHILA_KIN13A MFDYPAVVI-----DNGSERCKAG---IAGEYAPRADFPS-----IVGRPKYQNLMLQKGE 48
T.THERMOPHILA_KIN13B MLRKQIN-----NILQNQQIARTEYD---ESPQN-----VQIQQLDSFDSQN-- 39
T.THERMOPHILA_KIN13C MKGTAAEK-----HKERLKNA---RDVQMS--KKEAQINKL TDEEVVNEL-----KDKALPVYGTROEKL 55
H.SAPIENS_KIF2A MATAN-----FGKIQIG---IYVEIK---RSDGRIHQ-----MVTSLNEDNESVTVEWIENGDTKGKE 53
H.SAPIENS_KIF2B MASQFCLPESPLSPLKPLKPHFGDIQEG---IYVAIQ---RSDKRIHLA---VVTEINRENYWVTVEWEKAVKKGKK 70
H.SAPIENS_MCAK MAMDSSL-----QARLFPG---LAIKIQ---RSNGLIHPA---NVRTVNLEKSCVSVWEAEGGATKGKE 55
M.MUSCULUS_KIF2A MATAN-----FGKIQIG---IYVEIK---RSDGRIHQ-----MVTSLNEDNESVTVEWIENGDTKGKE 53
M.MUSCULUS_KIF2B MASQFCLPLAPRLSPLKPLKSHFTDFQVG---ICVAIQ---RSDKRIHLA---VVTEINRENSWVTVEWEKGVKKGKK 70
M.MUSCULUS_MCAK MESLH-----ARLFPG---LSINIQ---RSNGLIHPA---NISTVNVEKSCVSVWEIEGGTTKGKE 52
C.REINHARDTII_KIN13 MVSSVA-----RG-----LEDAGLK--- 16
G.INTESTINALIS_KIN13 MSDLVYQ-----WLESANLQ--- 15
L.MAJOR_KIN13.2 MRAESSGS-----E-----SQSRQ--- 14
T.BRUCETI_KIN13.2 MTSL----- 4
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T.THERMOPHILA_KIN13A FYIGDDALANKALFNLYPIENGLVTNYDNMEQIWRHCFDNELQVDPSSQQPCM-LT--ESAMTPK--LYREKMTNIMFET 123
T.THERMOPHILA_KIN13B -----SGRL----- 43
T.THERMOPHILA_KIN13C DRLKMYGMIIPNVQQQQ-QQ-----G-NNLP-PS-NNNGGL----- 88
H.SAPIENS_KIF2A IDLESIFSLNPDLVPDEEIEP-----S-PETP-PPPASSAKVNKIVKNRR----- 96
H.SAPIENS_KIF2B IDLETILLNLPALDSAEHPMP-----P-PPLSPLALAPSSAI---RDQR----- 110
H.SAPIENS_MCAK IDFDVAAINPELLQLLPLHP-----K-DNLP-LQ--ENVTIQK--QKRR----- 94
M.MUSCULUS_KIF2A IDLESIFSLNPDLVPDEEIEP-----S-PELP-PP-SSSKVNKIVKNRR----- 95
M.MUSCULUS_KIF2B IELETILLNLPALASLEHQRS-----R-RPLRPVSVVPSTAI---GDQR----- 110
M.MUSCULUS_MCAK IDIDVAAINPELLQLLPLRP-----K-DSLPL-LQ--ENVTVPK--QKRR----- 91
C.REINHARDTII_KIN13 -----RFV-----PAF---SGVS-DQ--AFLGLMM--SDYASY----- 41
G.INTESTINALIS_KIN13 -----QYY-----PAFEQ-QGIT-PQ--RFITITI--QDYGAL----- 42
L.MAJOR_KIN13.2 ----- 14
T.BRUCETI_KIN13.2 ----- 4

T.THERMOPHILA_KIN13A FDVPSFVYQIQAVALSLYSSCGVTGIVLDSGEGVTN---AVPIFEGCALRHAIQKNY-----LAGRDLDYCMKLMYE 192
T.THERMOPHILA_KIN13B -----EY 45
T.THERMOPHILA_KIN13C ----- 88
H.SAPIENS_KIF2A -----TVASIKNDPPSRDN-----RVVGSARARP-----SQFPE 125
H.SAPIENS_KIF2B -----TATK-----WV-----AMIPQ 121
H.SAPIENS_MCAK -----SVNS--KIPAP-K-----ESLSRSRSTRM-----STVSE 119
M.MUSCULUS_KIF2A -----TVAAVKNDPPPRDN-----RVVGSARARP-----SQLPE 124
M.MUSCULUS_KIF2B -----TATK-----WI-----AMIPH 121
M.MUSCULUS_MCAK -----SVNS--KIPAL-K-----EGLRSRSTRM-----STVSE 116
C.REINHARDTII_KIN13 -----GVVE-----LED----- 48
G.INTESTINALIS_KIN13 -----GIQA-----LPD----- 49
L.MAJOR_KIN13.2 ----- 14
T.BRUCETI_KIN13.2 ----- 4

T.THERMOPHILA_KIN13A VGLNFQSSVEREVIR-DIKE-KYCYVALDYAEALKAYQNNSSKHKQYQFPDGKMITIQDQRFVPELLFKPFDIGNEQKG 270
T.THERMOPHILA_KIN13B DSE-----RYQ-Q---VDNVVDQGNQNN-N----- 65
T.THERMOPHILA_KIN13C NQ-----NFNNN---NINVDFNDQQ----- 106
H.SAPIENS_KIF2A -----SAQ-Q---NGSVSDISPVQA-AK--KEFGP----- 151
H.SAPIENS_KIF2B KNQ-----TAS-G---DSLDRVVP----- 136
H.SAPIENS_MCAK LRI-----TAQ-E---NDMEVELPAAAN-SR--KQFSVPPAP----- 149
M.MUSCULUS_KIF2A -----SAQ-Q---NGSVSDISPVQA-AK--KEFGP----- 150
M.MUSCULUS_KIF2B RNE-----TPS-G---DSQTLMIP----- 136
M.MUSCULUS_MCAK VRI-----PAQ-E---NEMEVELPVPTN-SR--KQFAIPSHP----- 146
C.REINHARDTII_KIN13 -----KQRLFRLIKSIAA-----SVKSS-DAPA-----P----- 71
G.INTESTINALIS_KIN13 -----KQKLFRLITTLKSR-EN---ILEQQPSAPNTG---ATPQSVPS----- 85
L.MAJOR_KIN13.2 ----- 14
T.BRUCETI_KIN13.2 ----- 4

T.THERMOPHILA_KIN13A ISELAFHSIMSCDIDLRRNLYENIVLSGGTTMFDGFAERISKDINALAPLSIKAE----- 325
T.THERMOPHILA_KIN13B ----- 65
T.THERMOPHILA_KIN13C ----- 106
H.SAPIENS_KIF2A ----- 151
H.SAPIENS_KIF2B ----- 136
H.SAPIENS_MCAK -----TRPSCP-----AVAEIPLRMVSEEMEEQVHSIRGSSS 181
M.MUSCULUS_KIF2A ----- 150
M.MUSCULUS_KIF2B ----- 136
M.MUSCULUS_MCAK -----RASCS-----TVTELPLLMVSEEAEEQAHSTRSTSS 177
C.REINHARDTII_KIN13 ----- 71
G.INTESTINALIS_KIN13 ----- 85
L.MAJOR_KIN13.2 ----- 14
T.BRUCETI_KIN13.2 ----- 4

T.THERMOPHILA_KIN13A -VIALPQRKYSAFIGGSILSSLSFQSKWITKAEYDEEIEQKQK-INETKKEKEKN----SED--M--MQQE----- 387
T.THERMOPHILA_KIN13B --KKKQTK-----KSSLQNMKEKMEQKRQERREK-----MNQMK 97
T.THERMOPHILA_KIN13C ---KKQNAF-----KDPTVEKIQKMEQKREERRAK-----MEEAK 138

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H.SAPIENS_KIF2A -----PSRR-----KSNVCVEKLEKQEKREKRRRLQ-----QQELR 181
H.SAPIENS_KIF2B SKPCLMKQK-----KSPCLWEIQKLEQEKREKRRRL-----QQEIR 171
H.SAPIENS_MCAK ANPVNSVRR-----KSCLVKEVEKMKMKREKKAQ-----NSEMR 216
M.MUSCULUS_KIF2A -----PSRR-----KSNVCVEKLEKQEKREKRRRLQ-----QQELR 180
M.MUSCULUS_KIF2B SNPCLMKRK-----KSPCLREIEKLEKQEKREKRRRL-----QLEIR 171
M.MUSCULUS_MCAK ANPGNSVRR-----KSCIVKEMEKMKNKREKRAQ-----NSELR 212
C.REINHARDTII_KIN13 -----ALEKPNAL-IDLLEN-----DG-----D-----LLAD----- 92
G.INTESTINALIS_KIN13 -----SH-----VSPHVAQGRFVGDQKQNDIQQAQDMSLYESYDGGYEPPYVSAQSGSPANGDDY 142
L.MAJOR_KIN13.2 -----SER----- 17
T.BRUCETI_KIN13.2 ----- 4

NECK DOMAIN

T.THERMOPHILA_KIN13A -----F-NIDD-----LSNEPRIKVVIRKRPVNKKEQA 414
T.THERMOPHILA_KIN13B QQKLEKEQMNKESNRNVDIDFEMLEQERLN-----FSLQHV-----PISNNRLSVIVRKRPLFSKEEE 156
T.THERMOPHILA_KIN13C REREKIMEHQALGKNIDVDFEILIEKRNFK-----EPLQQAHT-----PASHLKLVCVVRKRPFKKEET 199
H.SAPIENS_KIF2A EKRAQDV--DATN--PNYEMCIRDFRGLDYLRL- TTAD-----PIDEHRICVVRKRP LNKKETQ 239
H.SAPIENS_KIF2B ARRALDV--NTRN--PNYEMIMHIEEYRRLDSSKI-SVLE-----PPQEHRCVVRKRP LNQRETT 229
H.SAPIENS_MCAK MKRAQEQ--DSSF--PNWEFARMIKFRATLECHPL-TMTD-----PIEHRICVVRKRP LNQKELA 274
M.MUSCULUS_KIF2A EKRAQDV--DATN--PNYEMCIRDFRGLDYLRL- TTAD-----PIDEHRICVVRKRP LNKKETQ 238
M.MUSCULUS_KIF2B ARRALDI--NTGN--PNFETMRMIEEYRRLDSSKM-SSE-----PPEDHRICVVRKRP LNQRETT 229
M.MUSCULUS_MCAK IKRAQEQ--DSSF--PNWEFARMIKEFRVTMCSP-LTVT-----PIEHRICVVRKRP LNQKELA 270
C.REINHARDTII_KIN13 -----AGVNFQLSPL-ANQPKAPVPSGGGGVPPSEGEDPPKIRVVVRKRP ISKKERE 144
G.INTESTINALIS_KIN13 VIPTIPY--HPNA--PNPPNPRGIPVTNRTV-VPPV-DLFL-----NQIQSRIRVVIRKRP INPKELS 199
L.MAJOR_KIN13.2 -----TCGVSPG-GPGG-----PGASPISVAVRKRPIPRGR-E 49
T.BRUCETI_KIN13.2 -----C-----PI-TSSITVAIRKRPIANNG-N 25
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NECK DOMAIN

MOTOR DOMAIN

T.THERMOPHILA_KIN13A KSDVDIIEV--KT--GQSVIVKEMKYVQLFIL SAAKTDKLIKHKQIDDIIFINRQKVD-LTKYVEEHLFNFDHAFGENCTNE 490
T.THERMOPHILA_KIN13B EGELDSISC--S--NPDIRVHEPK-----YKVDGITKYVENQDFQDNFASKEESTE 204
T.THERMOPHILA_KIN13C NGEIDSVSV--S--NPQIRVLAPK-----FKVDGITKYVENYDFTDNFENEETQ 247
H.SAPIENS_KIF2A MKDLDVITI--PS--KDVVMVHEPK-----QKVD-LTRYLENQTFRFDYAFDDSPNE 287
H.SAPIENS_KIF2B LKDLDIITV--PS--DNVVMVHESK-----QKVD-LTRYLENQTFCFDHFADDDKASNE 277
H.SAPIENS_MCAK KKEIDVIVSI--PS--KCLLLVHEPK-----LKVD-LTKYLENQAFCDFAFDETASNE 322
M.MUSCULUS_KIF2A MKDLDVITI--PS--KDVVMVHEPK-----QKVD-LTRYLENQTFRFDYAFDDSPNE 286
M.MUSCULUS_KIF2B MKDLDIITV--PS--HNVMVHESK-----QKVD-LTRYLENQTFCFDHFADDDKASNE 277
M.MUSCULUS_MCAK KKEIDVIVSI--PS--KCLLLVHEPK-----LKVD-LTKYLENQAFCDFAFDETASNE 318
C.REINHARDTII_KIN13 RGDDDVDV--LAKNNTVVVNEEK-----VKVD-LTKYLEKHAFKFDESLDENVSNE 193
G.INTESTINALIS_KIN13 QNQDQVVT A--DG--WNQVSIHEPK-----VKVD-LTKYTDLHFKFDFHVFNEQSDNQ 247
L.MAJOR_KIN13.2 DDENDVRCGEG- GPSVTVYEPK-----TKLD-LTPIEPPSSFSYDHVFGAEACTNE 99
T.BRUCETI_KIN13.2 SGDKDIVTC--ED--CRTISVHEPK-----TRVD-LKAVVETSAFAFDYVFDESAND 73
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MOTOR DOMAIN

KVD DOMAIN

T.THERMOPHILA_KIN13A EVFIFYFKDTIYKHFYSYLKWSQHSQSLISVSIQVYMTVVRPIVTAAFQARVTCFAYGQSGKTHMLGSAV---NR 566
T.THERMOPHILA_KIN13B D-----VYKYSLQPLIKCIFEHGVVTCFAYGQSGKTFMTRG----- 242
T.THERMOPHILA_KIN13C D-----VYKYSLRPLLDHIMNQGVIITCFAYGQSGKTFMTRG----- 285
H.SAPIENS_KIF2A M-----VYRFTARPLVETIFERGMATCFAYGQSGKTHMGGDMSGKNDQC 334
H.SAPIENS_KIF2B L-----VYQFTAQPLVESIFRGMATCFAYGQSGKTYMGGDMSGTAQDC 324
H.SAPIENS_MCAK V-----VYRFTARPLVQTIFFEGGATCFAYGQSGKTHMGGDLGKAQNA 369
M.MUSCULUS_KIF2A M-----VYRFTARPLVETIFERGMATCFAYGQSGKTHMGGDMSGKNDQC 333
M.MUSCULUS_KIF2B L-----VYQFTAQPLVESIFRGMATCFAYGQSGKTHMGGDMSGTAQDC 324
M.MUSCULUS_MCAK V-----VYRFTARPLVQTIFFEGGATCFAYGQSGKTHMGGDLGKAQNA 365
C.REINHARDTII_KIN13 A-----VYQTVGVLVRTLFRNGRACCFAYGQSGKTYMTPS----- 231
G.INTESTINALIS_KIN13 E-----IYQYAAKPLIRSVFEKNCVTFAYGQSGKFTMMHK----- 286
L.MAJOR_KIN13.2 E-----VYRGCQPLLQNVREGGAVIFAFGQSGKTHMLGTGE----- 140
T.BRUCETI_KIN13.2 V-----VYKVCQPILLSVDVQNGGSSVVVIAFGQSGKTHMLGHGS----K 115
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MOTOR DOMAIN

T.THERMOPHILA_KIN13A VPGMYVLASHDIFLALRQPQ-----FSD--LQIFVFSFYEIYCGKLFLLNDRQILHPRDGGKQINIIIGLMEQRITNDVQ 639
T.THERMOPHILA_KIN13B ---LQHYINDIFSIIQK-----NHQFQLIMSFFEYGGKFDLLNERNQLNIEDKNNVQIQNLEKPVRDQLE 310
T.THERMOPHILA_KIN13C ---LQNYVDTMVKLSTHQ-----YSSMNLKFIFISFFEYGGRCYDILLNNKNQLVILEDKNGYVQIQNIVEKQAMSAE 357
H.SAPIENS_KIF2A SKGIYALAAADVFLMKKPN-----YKLELQVYATFFEIYSGKVFDDLNRKTKLRVLEDGQQVQVVLQREVEKCVED 409
H.SAPIENS_KIF2B SKGIYALVAQDVFLLLNST-----YEKLDLKVYGTFFEYGGKVDLLNNKLLQVLEDGQQVQVVLQREVEKCVED 399
H.SAPIENS_MCAK SKGLYAMASRDVFLKNQPC-----YRKLGLEVVYVTFEYNGKLFLLNKKAKLRVLEDGQQVQVVLQREVEKCVED 444
M.MUSCULUS_KIF2A SKGIYALAAADVFLMKKPN-----YKLELQVYATFFEIYSGKVFDDLNRKTKLRVLEDGQQVQVVLQREVEKCVED 408
M.MUSCULUS_KIF2B SKGIYALVAQDVFLLLNST-----YEKLELKVYGTFFEYGGKVDLLNNKLLQVLEDGQQVQVVLQREVEKCVED 399
M.MUSCULUS_MCAK SKGIYAMASRDVFLKNQPC-----YRNLNLEVYVTFEYNGKLFLLNKKAKLRVLEDGQQVQVVLQREVEKCVED 440
C.REINHARDTII_KIN13 ---LPIRASADIFFMAQQQ-----YRD--ISLVCSCFEIYGNKVFDDLNRKTKLRVLEDGQQVQVVLQREVEKCVED 301
G.INTESTINALIS_KIN13 DNGIYVLAACFDILYELRVYNGSQGNSK-FLVPVVSFFEYGGKLFLLNRRQRLQALDQGNVQITGLTEKQISSVDA 365
L.MAJOR_KIN13.2 RPLGLYSLAVTELLTME-----H--SMTASFYAEYAKLFDLLNDRAEVAKMDEYQNVHVIGTEIQVSSVDD 208

T. BRUCEI_KIN13.2 TIGLYGYAIRELIGEET-----T--RKLAVSFYEVYGSKLFDLLNGRTQLKMMQDEADNLRIVGLSEKVVTCDE 183
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MOTOR DOMAIN

T. THERMOPHILA_KIN13A LLKIIIEFGMSARVTAQNSANTDSSRSHAILQIQLKE-----QNNVY-GKISFIDLAGSERGADVIDQNK 702
T. THERMOPHILA_KIN13B MIEIIEQAAIIRTTTHATEANEESRSHAIQIVVKDT-----NGSTR-GKLMVDLAGSERAQDCQSNK 374
T. THERMOPHILA_KIN13C MIQLIEYGHNIRTTTHATASNDTSSRSHAIQIIMLRND-----KDKQV-GKLVLDLAGSERAQDCQSNR 421
H. SAPIENS_KIF2A VLKIIDIGNSCRTSGQTSANAHSSRSHAVFQIILRR-----KGKLG-GKFSIDLAGNERGADTSSADR 472
H. SAPIENS_KIF2B VLNLVLEIGNSCRTSRQTPVNAHSSRSHAVFQIILKS-----GRIMH-GKFSVLDLAGNERGADTTKASR 462
H. SAPIENS_MCAK VIKMLDMGSACRTSGQTFANSNSRSHACFQIILRA-----KGRMH-GKFSVLDLAGNERGADTSSADR 507
M. MUSCULUS_KIF2A VLKIIDIGNSCRTSGQTSANAHSSRSHAVFQIILRR-----KGKLG-GKFSIDLAGNERGADTSSADR 471
M. MUSCULUS_KIF2B VLNLVLEIGNSCRTSGQTSVNAHSSRSHAVFQIILKA-----GGKLG-GKFSVLDLAGNERGADTAKATR 462
M. MUSCULUS_MCAK VIKMINMGSA CRTSGQTFANSNSRSHACFQIILRT-----KGRMH-GKFSVLDLAGNERGADTSSADR 503
C. REINHARDTII_KIN13 VKALIEESAKNRSTGTAANADSSRSHIMQFALKRAAPAGGFRREDAPEPRV-GKISFIDLAGSERGADTFDNNR 380
G. INTESTINALIS_KIN13 MLNLIDSGTLRAVGATGANADSSRSHAILQIALKYT-----KSGKEY-SRISFIDLAGSERASDVQNSDR 430
L. MAJOR_KIN13.2 VNALMMRGGQLRAIGTTHANDRSSRSHAVLEIKLKLADS-----SSESQL-GRITFVLDLAGSERASDTAETA 275
T. BRUCEI_KIN13.2 VYKLIKSGESLRSSGSTLANDTSSRSHAVLEIKVLNY-----QGEPHGRVTLIDLAGSERAADTSSDT 248
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MOTOR DOMAIN

T. THERMOPHILA_KIN13A QTRKDGAEINKSLALKECIRALDQ-----KNYTPFRGSKLTVLKDSFTG--NCRTVMIGNISPCQSSSEHTLNTLR 774
T. THERMOPHILA_KIN13B QRRVEGANINQSLALKECIRAMDSG-----AQHVPRGSKLTVLKDSFSLKQSNHIMFACISPGSSSDHTVNTLR 449
T. THERMOPHILA_KIN13C QRRMEGAEINKSLALKECIRAMDTG-----AAHVPRFRASKLTVLKDSFQSKSDKSKIVMIACISPGSSSDHTVNTLR 496
H. SAPIENS_KIF2A QTRLEGAEINKSLALKECIRALGRN-----KPHTPFRASKLTQVLRDSFIGE--NSRTCMIAISPMSASCENTLNTLR 545
H. SAPIENS_KIF2B KRQLEGAEINKSLALKECTALGQN-----KPHTPFRASKLTQVLRDSFIGE--NSSTCMIAISPMTSCENTLNTLR 535
H. SAPIENS_MCAK QTRMEGAEINKSLALKECIRALGQN-----KAHTPFRASKLTQVLRDSFIGE--NSRTCMIAISPMSASCENTLNTLR 580
M. MUSCULUS_KIF2A QTRLEGAEINKSLALKECIRALGRN-----KPHTPFRASKLTQVLRDSFIGE--NSRTCMIAISPMSASCENTLNTLR 544
M. MUSCULUS_KIF2B KRQLEGAEINKSLALKECIRALGKN-----KSHTPFRASKLTQVLRDSFIGE--NSYTCMIAISPMTSCENTLNTLR 535
M. MUSCULUS_MCAK QTRMEGAEINKSLALKECIRALGQN-----KAHTPFRASKLTQVLRDSFIGE--NSRTCMIAISPMSASCENTLNTLR 576
C. REINHARDTII_KIN13 QTRLEGAEINKSLALKECIRALDSD-----ARHVPRGSKLTAVLRDSFVGD--QARTVMIANISPCSSSEHTLNTLR 453
G. INTESTINALIS_KIN13 QTRMEGAEINKSLALKECIRAMKSNDSKGAHIPFRGSKLTMVLRDSFIGE--NSQVTMIANISPNKSCDNTLNTLR 507
L. MAJOR_KIN13.2 KTRREGAEINKSLALKECIRAMSMR-----KRHIPFRGSKLTQVLRDSFVGD--RCKTCVIAISPQGHCEDTLNTLR 347
T. BRUCEI_KIN13.2 RGRHEGAEINKSLALKECIRAMSRN-----RRHIPFRASKLTQVLRDSFIGE--NCKTCFIATVSPQRHCEDTLNTLR 320
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MOTOR DOMAIN

KEC DOMAIN

T. THERMOPHILA_KIN13A YADRVKELKPTNQDLSEQ-VVNYNHKLEQ-----SSKAKKKKQNYQSSQQMASAPQAAAQKKEKEKEENDSNKKNQAP 802
T. THERMOPHILA_KIN13B YADRLKESNGIKQDV-I-KI-----IEEQSKAKKKKQNYQSSQQMASAPQAAAQKKEKEKEENDSNKKNQAP 517
T. THERMOPHILA_KIN13C YADRLKENKPPVKG-MP-RI-----DAPQLIDEMPSYINHNHNIKSD-NQLNDQPSQ----- 547
H. SAPIENS_KIF2A YANRVKELTVDPDA----- 559
H. SAPIENS_KIF2B YANRVKELNVDVRP----- 549
H. SAPIENS_MCAK YADRVKELSPHSGP----- 594
M. MUSCULUS_KIF2A YANRVKELTVNPAA----- 558
M. MUSCULUS_KIF2B YANRVKELALEARP----- 549
M. MUSCULUS_MCAK YADRVKELSPHSGP----- 590
C. REINHARDTII_KIN13 YADRVKELRDKADRTPGG-VTPGDDAY----- 480
G. INTESTINALIS_KIN13 YADRVKELQHGKGGI-IFK-N-----VLKMG-----QN-- 533
L. MAJOR_KIN13.2 YADRIKELKGPANPHNGVKPIPC--KT----- 372
T. BRUCEI_KIN13.2 YANRIRDLKAPSDGFSR-KISM--TC----- 344
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MOTOR DOMAIN

T. THERMOPHILA_KIN13A -----LMKSAQN-----SNFAYNLPQPPNFPYQPTQAQQPRTNYNFMAGDQMGNN 852
T. THERMOPHILA_KIN13B PVKNLKGKNKEERQYDHTQYLMNNAKNKENMQSDVNIPSIYQDKPS-----QQEK----- 570
T. THERMOPHILA_KIN13C --KEQLSN-----RRNHSSNQMDASVIFFYLLMLNYILFIYEIKPQ-----QQ----- 589
H. SAPIENS_KIF2A -----A-----GD----- 562
H. SAPIENS_KIF2B -----YHR-----GH----- 554
H. SAPIENS_MCAK -----S-----GE----- 597
M. MUSCULUS_KIF2A -----A-----GD----- 561
M. MUSCULUS_KIF2B -----YHH-----CV----- 554
M. MUSCULUS_MCAK -----S-----GE----- 593
C. REINHARDTII_KIN13 -----YAA-VARAAGPG-----QAAA----- 495
G. INTESTINALIS_KIN13 -----AADVILGTARDEN--DVYKAGIVGVNAAPS-----QQAR----- 566
L. MAJOR_KIN13.2 -----C-----GQP--I-- 377
T. BRUCEI_KIN13.2 -----PNC-----NGP--V-- 351

T. THERMOPHILA_KIN13A NNILSAHNNAL-----NPNLNANNMYPGQSEMFDIE-----PRQLNKTPQQQQSFAGIKMNQPPQSQ 912
T. THERMOPHILA_KIN13B -----KLNAAQAKSSQIQ-----KI----- 585
T. THERMOPHILA_KIN13C -----LPPKQNKTPLL----- 600
H. SAPIENS_KIF2A -----VR----- 564
H. SAPIENS_KIF2B -----Y----- 555
H. SAPIENS_MCAK -----Q----- 598
M. MUSCULUS_KIF2A -----VH----- 563

M.MUSCULUS_KIF2B -----S----- 555
M.MUSCULUS_MCAK -----Q----- 594
C.REINHARDTII_KIN13 -----P-----APERERERADLQREDRERASDRYSPPRQAGAFAGG----- 534
G.INTESTINALIS_KIN13 ----- 566
L.MAJOR_KIN13.2 ----- 377
T.BRUCETI_KIN13.2 ----- 351

T.THERMOPHILA_KIN13A QQYRPPSNQ-LGYQ-----NQMDQNTINSQTN----PINLNIEQI-ENWTA----- 953
T.THERMOPHILA_KIN13B ----PPIQQS-V----- 592
T.THERMOPHILA_KIN13C ----PPADKRIA----- 608
H.SAPIENS_KIF2A ----PIMHHP-P----- 571
H.SAPIENS_KIF2B ----PIGHEA-P----- 562
H.SAPIENS_MCAK ----LIQMET-E----- 605
M.MUSCULUS_KIF2A ----PIMHHP-P----- 570
M.MUSCULUS_KIF2B ----PPGHEV-P----- 562
M.MUSCULUS_MCAK ----PVQMET-E----- 601
C.REINHARDTII_KIN13 -----GGGGAGA----- 541
G.INTESTINALIS_KIN13 ---VPPASQA-PITA-----RQIQQLPQPHYNPYPPNSKP-AFEPRVET----- 608
L.MAJOR_KIN13.2 ---FIGDRHV-CKRQLVCPHCROQVD---KQELDTHMSE---CKESPMRC-QYCNERMLRSESVGHRRRCARAPIRCG 445
T.BRUCETI_KIN13.2 ---RPDASHT-CVRLSTRCPHCROQVVE---KHNLEGHIEE---CSEFPVRC-PRCNELLVRGDIPRHRRCRSLSLVRCP 419

T.THERMOPHILA_KIN13A ----- 953
T.THERMOPHILA_KIN13B -----QQIEQASTQNSTQQVVSSTSVSSASALALHREKRDRPQTQQKRQSSIGKKNANQTDTSNQNLNLLQKQI 665
T.THERMOPHILA_KIN13C -----KQIEEERIEK----- 618
H.SAPIENS_KIF2A -----NQIDDLLETQW----- 581
H.SAPIENS_KIF2B -----RMLKSHIGN----- 571
H.SAPIENS_MCAK -----EMEAC--SN----- 612
M.MUSCULUS_KIF2A -----SQIDDLLETQW----- 580
M.MUSCULUS_KIF2B -----LMIENDNTN----- 571
M.MUSCULUS_MCAK -----VMEAS--SN----- 608
C.REINHARDTII_KIN13 ----- 541
G.INTESTINALIS_KIN13 ----- 608
L.MAJOR_KIN13.2 ACGATVPRQLIDR----- 458
T.BRUCETI_KIN13.2 LCTCHVMRCGLEK----- 432

T.THERMOPHILA_KIN13A ----- 953
T.THERMOPHILA_KIN13B LNTDASLA--TNHNDHL SGVDQNKALSLKQLKQISQSNPVTQNNQGGQIRINSAHPSNNNSQNN---NTRDDNNNSF 740
T.THERMOPHILA_KIN13C ---EKLLQQQQQQAAQQQQQHMKRGQ---SSNLP SANKQPTTNQQQQQK-----NNSQNNLSKNSDDEGEQI 682
H.SAPIENS_KIF2A G-VGSSPQRD----- 590
H.SAPIENS_KIF2B --SEMSLQRD----- 579
H.SAPIENS_MCAK G---AL----- 615
M.MUSCULUS_KIF2A G-VGSSPQRD----- 589
M.MUSCULUS_KIF2B --SGKSLQRD----- 579
M.MUSCULUS_MCAK G---TS----- 611
C.REINHARDTII_KIN13 ----- 541
G.INTESTINALIS_KIN13 ----- 608
L.MAJOR_KIN13.2 ----- 458
T.BRUCETI_KIN13.2 ----- 432

T.THERMOPHILA_KIN13A -----RNEEDL-QLI----- 962
T.THERMOPHILA_KIN13B NQLPKKGHNQQQKKTQQNDNI-STI---NNKNNRDSANHNSRHNSSSNNNNQINNNNFNQLNDKSLNDHILQKNK 816
T.THERMOPHILA_KIN13C QQVNVKGVK-----DDV-RCM---KET-----LKMCKNNKVSNE----- 711
H.SAPIENS_KIF2A -----DL-KLL---CE-----QNEEEVSPQ----- 606
H.SAPIENS_KIF2B -----EFIKIP--YVQ-----SEEQKEIEE----- 597
H.SAPIENS_MCAK -----IPGNLS-----KEEEELSSQ----- 630
M.MUSCULUS_KIF2A -----DL-KLL---CE-----QNEEEVSPQ----- 605
M.MUSCULUS_KIF2B -----EVIQIP-TVE-----KEEEKESDE----- 597
M.MUSCULUS_MCAK -----LTG-----NEEEELSSQ----- 623
C.REINHARDTII_KIN13 -----DGECSA-AAL----- 550
G.INTESTINALIS_KIN13 -----TDE---DDM----- 614
L.MAJOR_KIN13.2 -----HAQQECAEA-KVK---CR-----YCGCVQSRQL----- 482
T.BRUCETI_KIN13.2 -----HTLMDCGAK-LEK---CR-----YCGQGFPRHS----- 456

T.THERMOPHILA_KIN13A -----SQKHEQ-LISLILAE----- 976
T.THERMOPHILA_KIN13B EVQENIEKAQQHQIEQKTKDLILQIQIEK----- 845
T.THERMOPHILA_KIN13C -----FFDFHE-KVNTILEE----- 725
H.SAPIENS_KIF2A -----LFTFHE-AVSMVEM----- 620
H.SAPIENS_KIF2B -----VET-----L----- 601
H.SAPIENS_MCAK -----MSSFNE-AMTQIREL----- 644
M.MUSCULUS_KIF2A -----LFTFHE-AVSMVEM----- 619
M.MUSCULUS_KIF2B -----LTS----- 600
M.MUSCULUS_MCAK -----MSSFNE-AMTQIREL----- 637
C.REINHARDTII_KIN13 -----AERHDE-LMDSILLE----- 564
G.INTESTINALIS_KIN13 -----VRTHCD-LVDSIYEQ----- 628
L.MAJOR_KIN13.2 -----LAAHEQ-NCDAAKVACPHCLQFMKRRLDGHVATCVRNLSRAMHTP----- 527

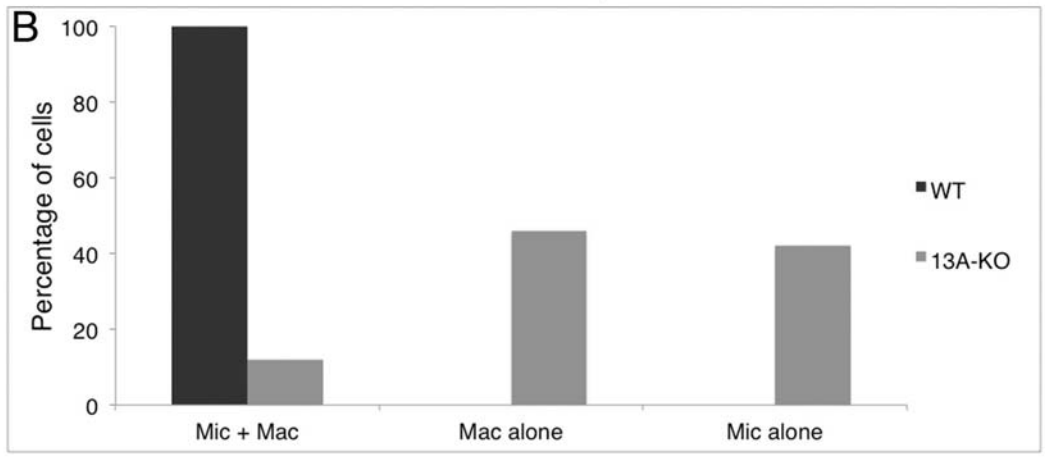
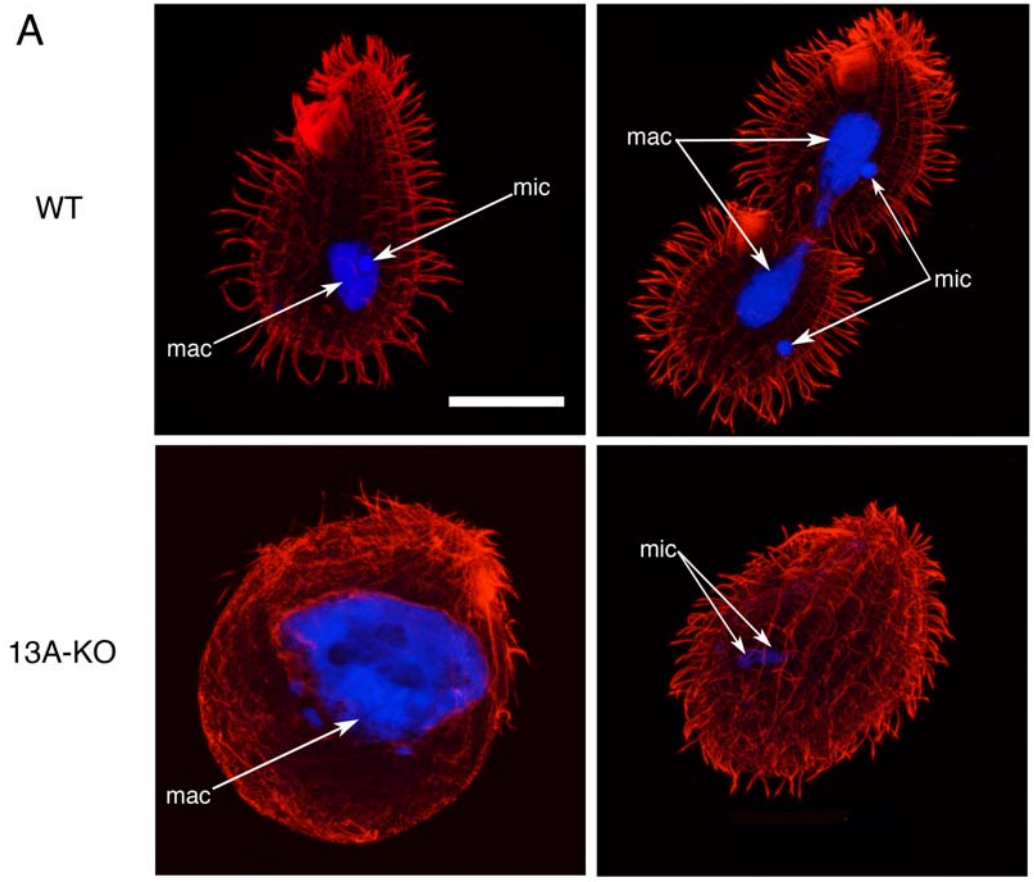
T. BRUCEI_KIN13.2 -----LKRHED-VCTMMKIACPYCLQYFRKVCDAHASVCRVNPNCRRVSPSRIRDSGEEVWKITNGKEWR 521

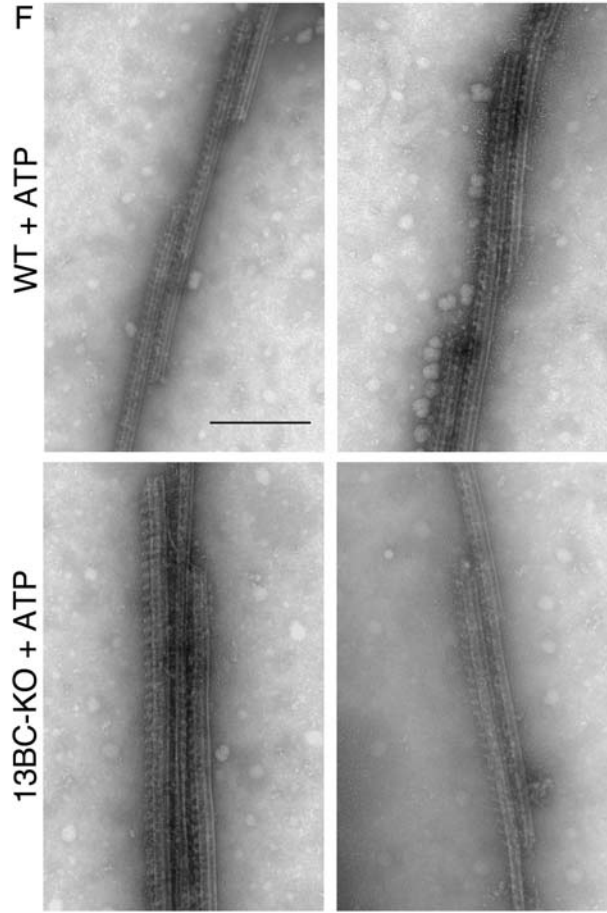
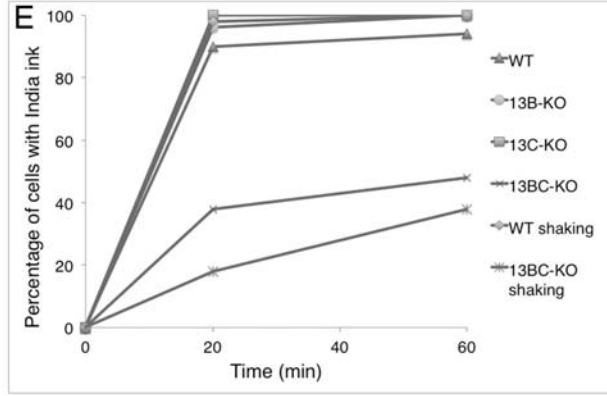
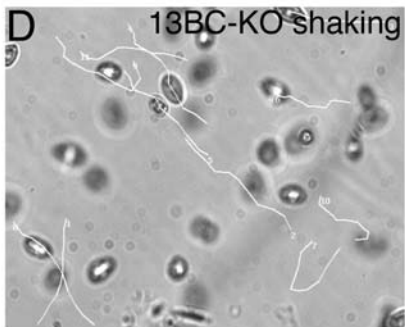
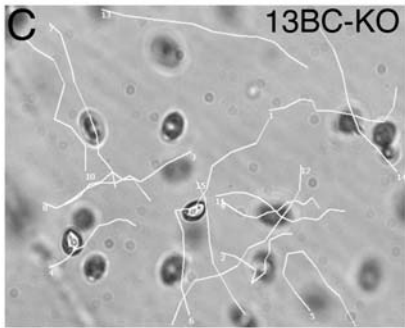
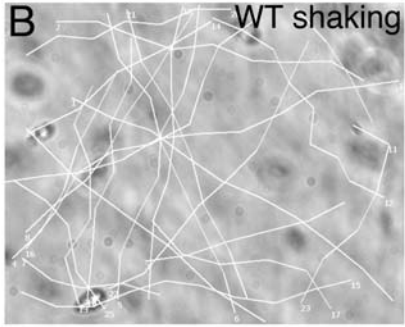
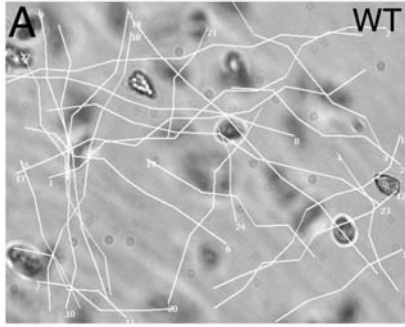
T. THERMOPHILA_KIN13A -----EEDV-----IASHRSHIDDMV----- 992
T. THERMOPHILA_KIN13B -----QEDM-----FSSHMSAIKEDA----- 861
T. THERMOPHILA_KIN13C -----QEEI-----FATHMAAIKEDA----- 741
H. SAPIENS_KIF2A -----EEQV-----VEDHRAVFQESI----- 636
H. SAPIENS_KIF2B -----PTLL-----GKDTTISGKGSS----- 617
H. SAPIENS_MCAK -----EKA-----MEELKEIIQGP----- 660
M. MUSCULUS_KIF2A -----EEQV-----VEDHRAVFQESI----- 635
M. MUSCULUS_KIF2B -----KKEPAASWSRSN----- 612
M. MUSCULUS_MCAK -----EERA-----LEELREIIQGP----- 653
C. REINHARDTII_KIN13 -----EENL-----VAFHRAKLEEDM----- 580
G. INTESTINALIS_KIN13 -----EDLI-----VRAHRRQVDSMM----- 644
L. MAJOR_KIN13.2 -----RSTALNIIASTSMETSQTFLASSSSSIQAQOSTPKSATAQTAASS---RASSSHNRNVAEEPAGGQVD---LVAA 595
T. BRUCEI_KIN13.2 QRPRLRNQSLKQLEAISRTKSSVQ-----LREGKPLGPLDNFSLPALH 567

T. THERMOPHILA_KIN13A -----ELTKQEMLLHDV---DKPASD--VDVY----- 1015
T. THERMOPHILA_KIN13B -----NLLSLESGLLTDCKQDGMDCD--IDSY----- 887
T. THERMOPHILA_KIN13C -----KLLTQSELSISKVQGTGFIDYD--IDLY----- 767
H. SAPIENS_KIF2A -----RWLEDEKALLEMT---EEVDYD--VDSY----- 659
H. SAPIENS_KIF2B -----QWLE---NIQERA---GGVHHD--IDFC----- 637
H. SAPIENS_MCAK -----DWLE---LSEMT---EQPDYD--LETF----- 679
M. MUSCULUS_KIF2A -----RWLEDEKALLEMT---EEVDYD--VDSY----- 658
M. MUSCULUS_KIF2B -----QWWE---AIQETA---EGVNGD--VDFC----- 632
M. MUSCULUS_MCAK -----NWL E---LSEMT---DQPDYD--LETF----- 672
C. REINHARDTII_KIN13 -----ETMRQEMALLQEV---DKPGSE--IDHY----- 603
G. INTESTINALIS_KIN13 -----QLVKEEVALHAI---ENDQVS--IDHW----- 667
L. MAJOR_KIN13.2 DEGTGPLNFTRAHSSRRSSAQLASGVSLANQSQSNRAPGAGGRWNAGALGADEVS---AACDSD--CGSV----- 660
T. BRUCEI_KIN13.2 APSSAPDRKHPVTSAFTE-----SLQSHP---NSEDDADKEVCRCTAPTISGEN 614

T. THERMOPHILA_KIN13A -----V-----QNLDAILQHKS---EINMLRQLQT---FRSHLKK----- 1046
T. THERMOPHILA_KIN13B -----V-----KKFESIVSKKL---LMYNLLQQKMDT---LK-KMRNEYNKYSLNQINNN 931
T. THERMOPHILA_KIN13C -----V-----KKLETVIKKKL---KMK-----HLQ----- 785
H. SAPIENS_KIF2A -----A-----TQLEAILEQKI---DIL TELRDKVKV---FRAALQ----- 689
H. SAPIENS_KIF2B -----I-----ARSLILEQKI---DALTEIQKLLK---LLADLH----- 667
H. SAPIENS_MCAK -----V-----NKAESALAQA---KHFSALRDVIKA---LRLAMQ----- 709
M. MUSCULUS_KIF2A -----A-----TQLEAILEQKI---DIL TELRDKVKV---FRAALQ----- 688
M. MUSCULUS_KIF2B -----I-----AQSLSVLEQKI---GVLTDIQKLLQS---LREDLQ----- 662
M. MUSCULUS_MCAK -----V-----NKAESAL TQQAQAKHFSALREV IKA---LRLAMQ----- 705
C. REINHARDTII_KIN13 -----V-----EQMSALLNIKR---QGIQELQAKLDT---FKAKLRE----- 634
G. INTESTINALIS_KIN13 -----L-----VKLSDILSRKE---EAITTLKGNLSA---FKQALQK----- 698
L. MAJOR_KIN13.2 -----VCPYSRYGCPVKVTRLNVA AHLKESMQHL---ELVTTYADRVDEQNMQLRRLVIHE----- 714
T. BRUCEI_KIN13.2 SSRVGGEGCVCPYAAAYGLHTVCDSSLEKHMKDSVEMHL---QLVRDYAERVSEENNIL-RERVNE----- 676

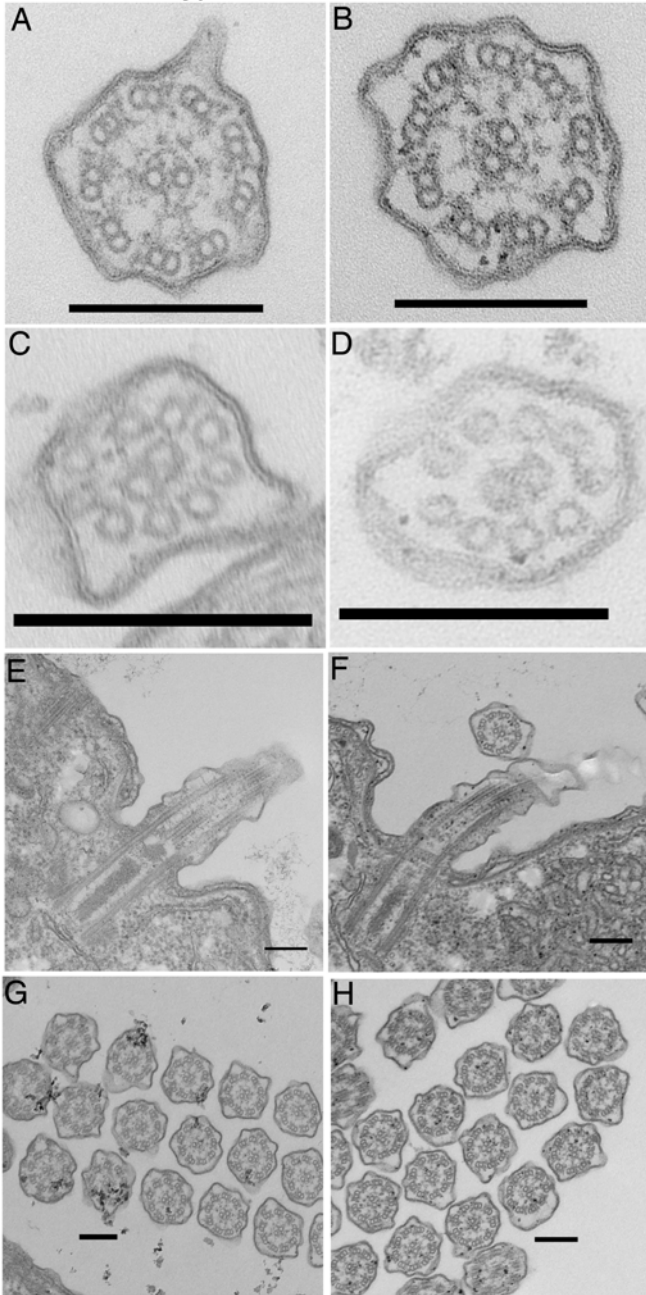
T. THERMOPHILA_KIN13A -----EELLSKKF-YEQRAQIMVDFLNSNTINPNNDQMLLDDLPD-LYDN 1091
T. THERMOPHILA_KIN13B HHEKNQQQQAQKQIEDPNKKRQS-----FL-----LKK-KIEN 963
T. THERMOPHILA_KIN13C -----EEEEISSK-----VKD-TFYF 800
H. SAPIENS_KIF2A -----EEEQASKQI-----N-----PKRPRAL 706
H. SAPIENS_KIF2B -----VK-----SKVE 673
H. SAPIENS_MCAK -----LEEQASRQI-----S-----SK-KRPQ 725
M. MUSCULUS_KIF2A -----EEEQASKQI-----N-----PKRPRAL 705
M. MUSCULUS_KIF2B -----KK-----SQVE 668
M. MUSCULUS_MCAK -----LEEQASKQI-----N-----SK-KRHQ 721
C. REINHARDTII_KIN13 -----EEALSRTV-H-----KIRT 647
G. INTESTINALIS_KIN13 -----EEELSHSI-DLN-----K-ARCK 714
L. MAJOR_KIN13.2 -----TDTLSSRV-T-M-----E-RLDK 729
T. BRUCEI_KIN13.2 -----GTTKASKL--H-----E-LESV 690



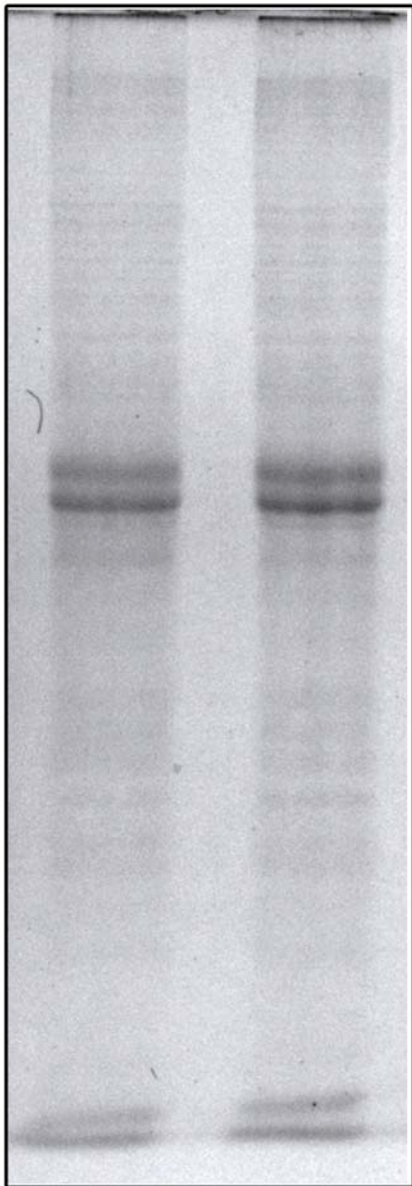


Wild type

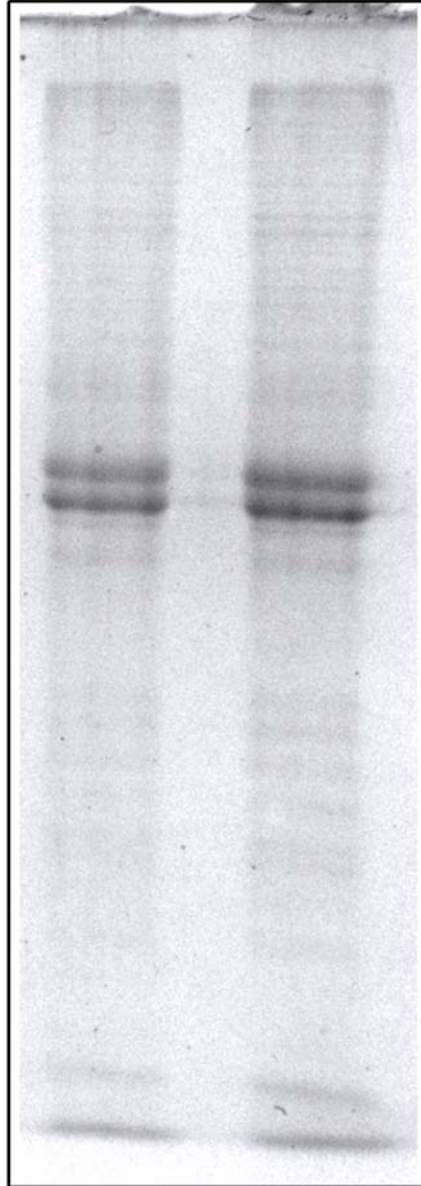
13BC-KO



Total cilia
WT 13BC-KO



Axoneme
WT 13BC-KO



M + M
WT 13BC-KO

