Craige et al., http://www.jcb.org/cgi/content/full/jcb.201006105/DC1

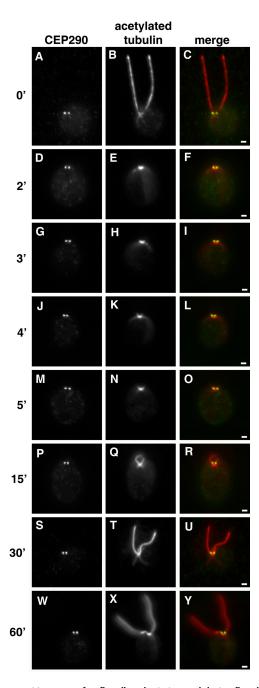


Figure S1. **CEP290 remains associated with the transition zone after flagellar abscission and during flagellar regeneration.** Wild-type cells were deflagellated by adding acetic acid to pH 4.5. After 45 s, the pH was neutralized to pH 7.0 with KOH. The cells were then fixed at the indicated time points after the addition of acid and processed for immunofluorescence with anti-CEP290 and anti-acetylated tubulin antibodies. Bar, 1 µm.

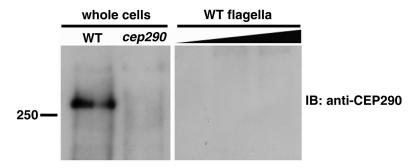


Figure S2. **CEP290** is **not present in isolated flagella.** Western blotting of whole cells and isolated wild-type flagella probed with antibody to CEP290. Increasing amounts of flagella were loaded (cell body/flagella ratios of 1:1, 1:4, and 1:16). The position and molecular mass (in kD) of a standard protein are indicated on the left.

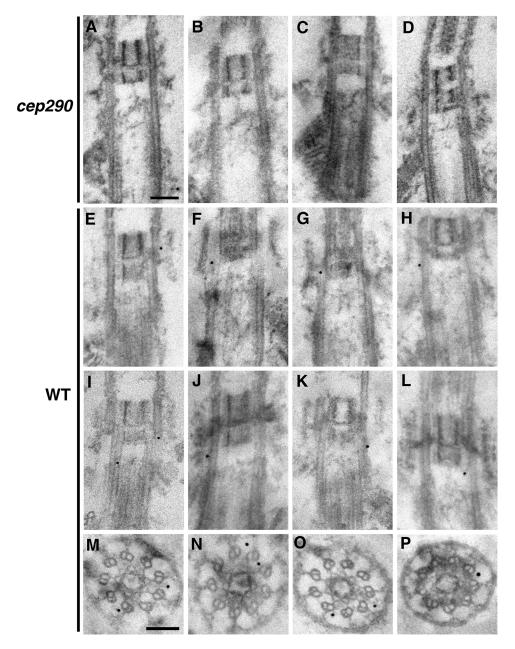


Figure S3. Immunogold localization of CEP290. (A) Detergent-extracted cytoskeletons were labeled with antibodies to CEP290, followed by labeling with 12-nm gold secondary antibody. Representative EM images for the cep290 mutant (A–D) and wild-type transition zones (E–P) are shown in longitudinal views (A–L) and in cross section (M–P). Bars, 100 nm.

## anti-HA 6-nm gold anti-CEP290 12-nm gold

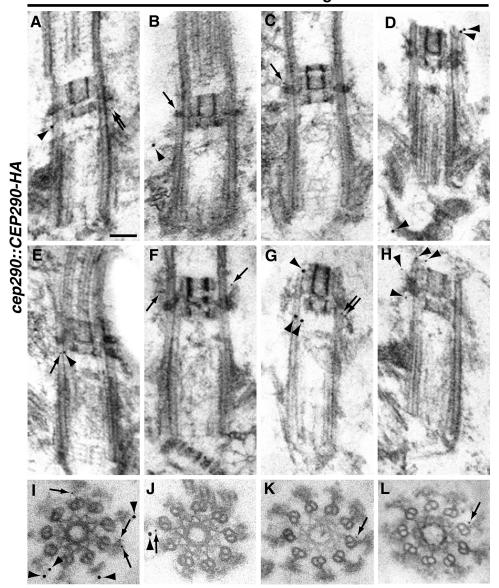
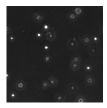


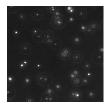
Figure S4. Immunogold localization of CEP290-HA. The CEP290-HA rescued strain (cep290::CEP290-HA) was double-labeled with antibodies to HA (6-nm gold particles, arrows) and CEP290 (12-nm gold particles, arrowheads). Bar, 100 nm.



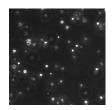
Video 1. Wild-type cell motility. Cells were imaged at RT at 30 frames per second; playback is in real-time.



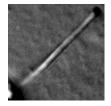
Video 2. cep290 mutant cell motility. Cells were imaged at RT at 30 frames per second; playback is in real-time.



Video 3. cep290::CEP290 rescued cell motility. Cells were imaged at RT at 30 frames per second; playback is in real-time.



Video 4. cep290::CEP290-HA rescued cell motility. Cells were imaged at RT at 30 frames per second; playback is in real-time.



Video 5. **DIC imaging of IFT in a wild-type cell.** Wild-type cells were immobilized in agarose (see Materials and methods) and imaged at 24 frames per second; playback is in real-time.



Video 6. **DIC imaging of IFT in a cep290 mutant cell.** cep290 mutant cells were immobilized in agarose (see Materials and methods) and imaged at 24 frames per second; playback is in real-time.

Table S1. Primers used in this study

Name	Sequence	Amplification of cep290 DNA
NPH6_Nf NPH6_Nr	5'-GGTCCATTGGAGAGAGACA-3' 5'-TGCGAATCTCATTGTTCTGC-3'	+
NPH_Cf NPH6_Cr	5'-CTGGGTGCTGTTGTTGT-3' 5'-TGACCTCATCCCAGAAGGAC-3'	_
NPH6_M1f NPH6_M1r	5'-GATGAGCCAGTCGCTGTGTA-3' 5'-AATGATTTGGGCTCATACGC-3'	-
NPH6_M2f NPH6_M2r	5'-CCCGTAATCACCCATTCATC-3' 5'-TGCAAACGTAAGCAAGCATC-3'	_
IEPH6_M3f IEPH6_M3r	5'-CCAATGTGCACACAAACA-3' 5'-ATCGCTGAAGGTCTGCATCT-3'	_
IEPH6_M4f IEPH6_M4r	5'-GCTGCCATATCACCACACAC-3' 5'-ACCATCCACTCGTTCACCTC-3'	_
IEPH6_M5f IEPH6_M5r	5'-AGGACAACCTGCTGTCCATC-3' 5'-TCTCGTCCTCCCAAGAGCTA-3'	_
IEPH6-M6f IEPH6_M6r	5'-GCCAAGGTTGAGGACCTGTA-3' 5'-AGCTCGGCCTTACACACAAC-3'	_
IEPH6_M7f2 IEPH6_M7r	5'-TTCATGACGACCATGCTCAC-3' 5'-TGCTGCAGCTCCTTGTAGTC-3'	_
AP61_Nf AP61_Nr	5'-TGGGACTGGACCTCACCTAC-3' 5'-TACATCCTCCCGATTCCTTG-3'	+
AP61_Cf AP61_Cr	5'-CTGCATGCCAACTACCTGAA-3' 5'-GGAGCAGAAGTGCAGAAACC-3'	+
_340104_Nf _340104_Nr	5'-AGCGCCTCTTAAACGTCAGA-3' 5'-ACTTGTTCCGCCAAGCTAGA-3'	_
:_340104_Cf :_340104_Cr	5'-TGTGACAGCGGACAAGACTC-3' 5'-CAAGCCACTCAAATGCTTCA-3'	-
_340036_Nf _340036_Nr	5'-CTTCAGCTCCACTCCAGGAC-3' 5'-GAAGTTGTCGACCGTGATGA-3'	-
C_340036_Cf C_340036_Cr	5'-GTGGATCAGGGTGTCATGTG-3' 5'-TAGCACACGCTCCTCTAGCA-3'	+

Names and sequences of primers used to map the deletion in the cep290 mutant strain. Plus and minus symbols indicate whether a PCR product was generated using cep290 mutant genomic DNA as template.