Supplemental Figure 1: Western blots showing antibodies against Xenopus laevis (xNups) and mammalian nucleoporins (hsNups) and γ-TuRC.

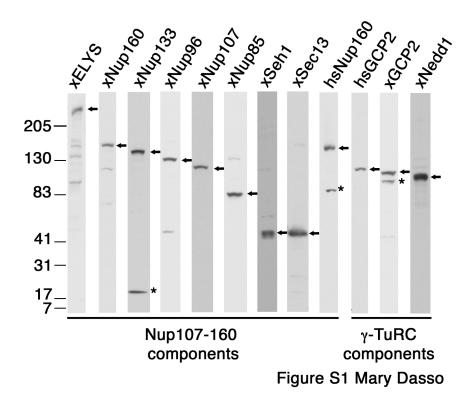
Supplemental Figure 2: γ-TuRC and Nup107-160 localization at

kinetochores.

a. Nocodazole-arrested HeLa cells were stained by indirect immunofluorescence with antibodies against Nup107 (upper panel, green), γ -tubulin (middle panel, green) and Nup160 (lower panel, green) in second column and CREST (third column, red) and counterstained with Hoechst 33342 dye to visualize chromosomal DNA (first column, blue). Inserts show magnified kinetochores stained by CREST and γ -tubulin or Nup107 or Nup160.

b. To test whether γ -TuRC associates with kinetochores of condensed chromosomes in XEEs, CSF extracts containing sperm chromatin were driven into interphase with 6 mM CaCl₂. After 60 minutes, mitosis was reestablished by addition of an aliquot of CSF-arrested extracts plus nocodazole. The mitotic chromosomes were analyzed by indirect immunofluorescence with antibodies against xNedd1, xGCP2 or γ -tubulin (second column, green) and the kinetochore marker Bub1 (third column, red) and counterstained with Hoechst 33342 dye to visualize chromosomal DNA (first column, blue). Similarly Nup107-160 complex members (Nup85 and Nup160) also stained Bub1 positive kinetochores. Scale bar=5 μ m

Supplemental Figure 3: Original western blots from figures 1, 3 and 4. The original uncropped western blot film scans for figures 1b, 1c, 3b, 4b and 4d with molecular weight markers are shown.



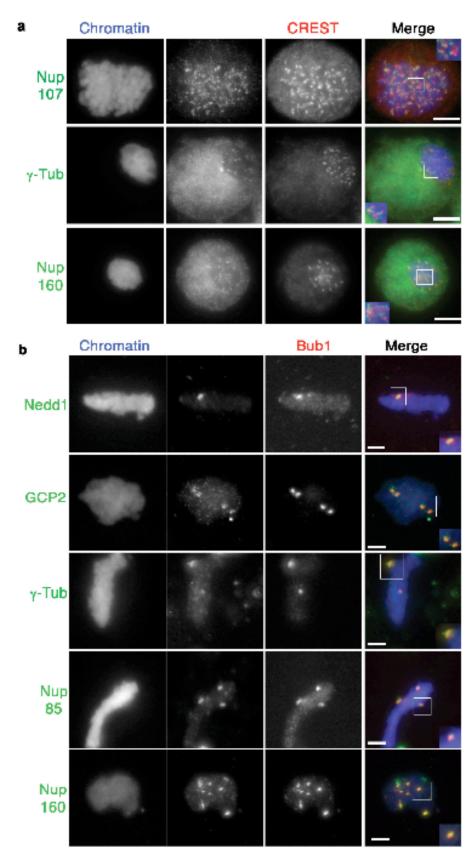


Figure S2 Mary Dasso

