Supporting Information

Zheng et al. 10.1073/pnas.1003953107

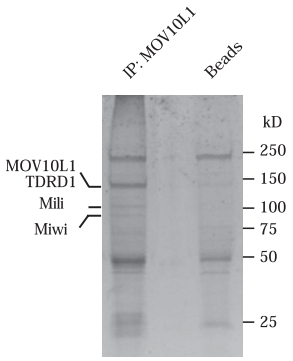

Fig. S1. Purification and characterization of MOV10L1-associated proteins from 18- to 20-d-old testes by immunoprecipitation (IP) and mass spectrometry. Protein identity for each band is indicated. In brief, to isolate MOV10L1-associated proteins, eight pairs of 18- to 20-d testes (≈300 mg) were homogenized in 3 mL radioimmunoprecipitation assay buffer (RIPA) in the presence of proteinase inhibitor mixture. Protein lysate was centrifuged twice. Supernatants were precleared with protein A agarose beads. Precleared lysate incubated with affinity-purified anti-MOV10L1 antibody, followed by binding to protein A agarose beads. Immunoprecipitated proteins were extensively washed, run on a 4–15% gradient SDS/PAGE gel, and stained with SYPRO Ruby (Bio-Rad).

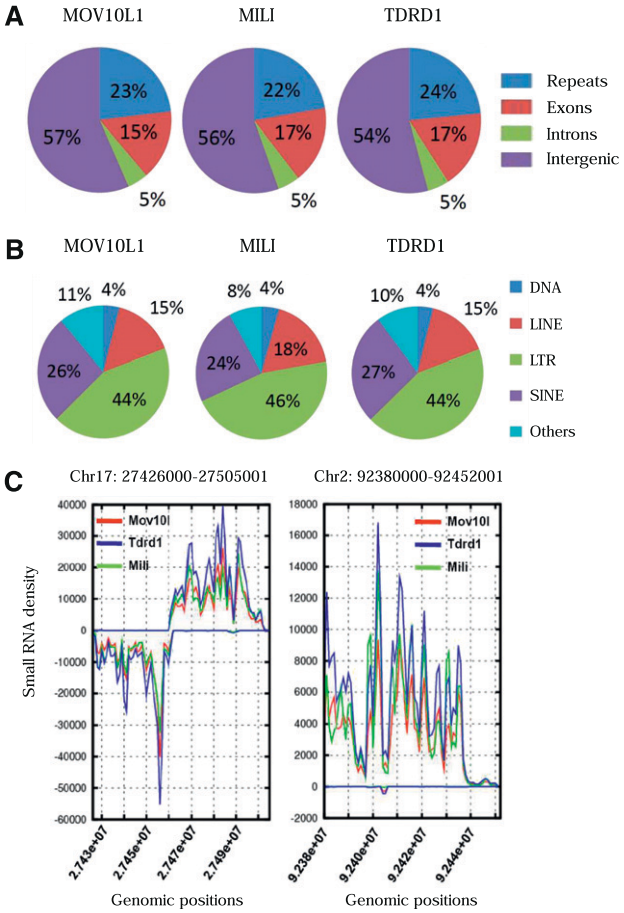

Fig. S2. Profiles of small RNA reads from MOV10L1, MILI, and TDRD1 libraries. (*A*) Annotation profile of MOV10L1 reads resembles those in MILI and TDRD1 libraries. (*B*) Pie charts showing representation of repeat piRNAs belonging to different transposon classes in indicated libraries. (*C*) Common genomic origins of MOV10L1-, MILI-, and TDRD1-associated piRNAs. All three libraries have similar read-depths (≈2 million). The Piwi-interacting RNA (piRNA) density for the top two pachytene clusters was plotted. The chromosome 17 (Chr17) cluster is ≈79 kb in length and bidirectional, with piRNAs arising from both strands in a nonoverlapping manner, whereas the Chr2 cluster is ≈72 kb and unidirectional, with most piRNAs deriving from the top strand.

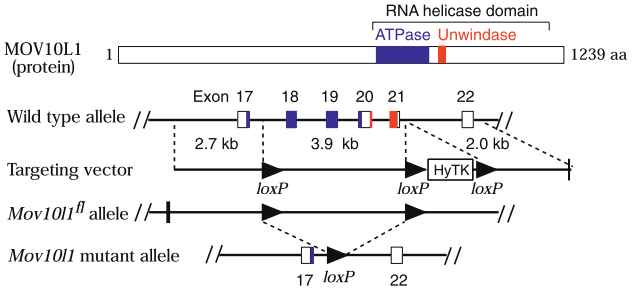

Fig. S3. Targeted inactivation of the *Mov10l1* gene. *Top:* Illustration of the predicted RNA helicase domain of mouse MOV10L1 protein. The mouse *Mov10l1* gene consists of 27 exons and spans a >72-kb genomic region. This deletion removes the ATPase and unwindase domains, which are highly conserved among RNA helicases.

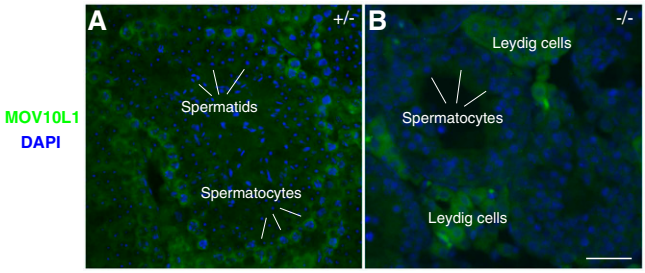

Fig. S4. Immunofluorescence analyses of MOV10L1 in testes from adult *Mov10l1*$^{+/-}$ (*A*) and *Mov10l1*$^{-/-}$ (*B*) mice. Leydig cells give strong autofuorescence. In *Mov10l1*$^{-/-}$ spermatocytes (*B*) the internally deleted mutant MOV10L1 protein was barely detectable by immunofluorescence analysis, owing to its sharply reduced abundance. (Scale bar, 25 μm.)

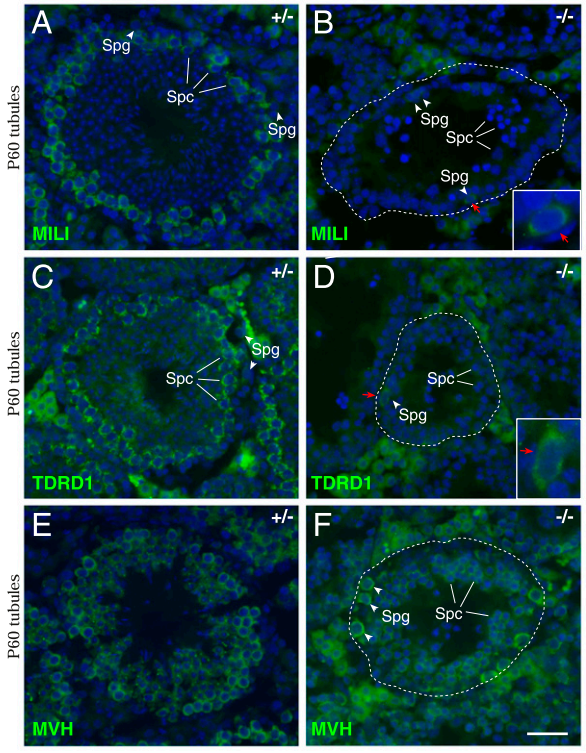

Fig. S5. MILI and TDRD1 persist in spermatogonia but not in spermatocytes in *Mov10l1*$^{-/-}$ testes. Frozen testis sections from 2-mo-old (P60) *Mov10l1*$^{+/-}$ (*A*, *C*, and *E*) and *Mov10l1*$^{-/-}$ (*B*, *D*, and *F*) were immunostained with anti-MILI, anti-TDRD1, and anti-MVH antibodies. Chromatin was stained with DAPI. *Mov10l1*$^{-/-}$ seminiferous tubules are demarcated with dash lines. *Insets* (*B* and *D*): Enlarged view of indicated spermatogonia. Spg, spermatogonia; Spc, spermatocytes. (Scale bar, 25 μm.)

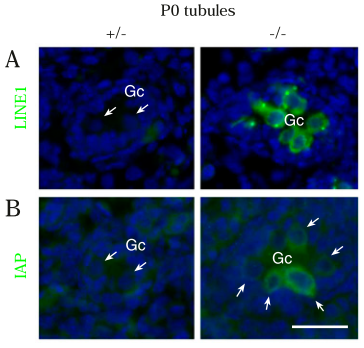

Fig. S6. Derepression of LINE1 and IAP retrotransposons in *Mov10l1*$^{-/-}$ perinatal gonocytes. Newborn (P0) testis sections were immunostained with anti-L1 ORF1p and anti-IAP antibodies. Gonocytes (Gc) are indicated by arrows. (Scale bar, 25 μm.)