Figure S1. Polypeptide sequence analysis of *Drosophila* Mobs. (A) Crystal structure of human Mob1B (NCBI CN3D 4.1 structure) revealing general multi-domain alpha helical organization of Mob proteins and zinc binding site. (B) Amino acid sequence alignment of *Drosophila melanogaster* Mob proteins with human Mob1B indicates that *Drosophila* Mobs are significantly divergent from each other. Highly conserved residues are colored black, while less well conserved residues are colored orange. Helical domains and b-strands of Human Mob1B as determined from crystal structure analysis are overlayed on the linear amino acid sequence. The four amino acid residues (2 Cysteines and 2 Histidines) necessary for Zn^{2+} coordination (black arrowheads) are conserved between *Drosophila* Mob proteins.

Figure S2. Validation of DMob4 rescued animals. PCR analysis of DMob4$^{EY\Delta L3}$ homozygous mutants rescued to adult stages with *actin-GAL4:UAS-DMob4* indicates that the rescued animals carry the two copies of the EYΔL3 mutant chromosome which harbors a 357 b.p. deletion that spans the *DMob4* initiator methionine.

Figure S3. Ubiquitous expression of DMob4 by immunohistochemistry with anti-DMob4 antisera. (A-D, F-G, I-K) Embryonic expression pattern of DMob4. (A) *En face* view of syncytial blastoderm embryo. (B) Longitudinal section of embryo at syncytial blastoderm stage embryo. (C-D) *En face* views of DMob4 localization patterns during mitosis and cellularization. (F,G) DMob4 expression

in a whole-mount stage 16 embryo. Strong expression of DMob4 is seen in the CNS. (G) High magnification view of the CNS. Anti-DMob4 labels neurons of the longitudinal connectives and central commissure tracts. (E,H) Larval expression of DMob4. (E) DMob4 expression in the salivary gland epithelium is largely cytoplasmic. (H) DMob4 in the CNS is strong in the ventral nerve cord and peripheral nerves. (I-K) *DMob4*$^{EY_{\Delta}L3}$ homozygous mutant embryo (St. 16) stained for DMob4 (red) to indicate specificity of the DMob4 antiserum. Embryo co-stained with Futsch/22C10 (green) to reveal embryo orientation and the CNS. DMob4 staining observed in CNS of control embryos (F), is absent in *DMob4* nulls (I).