Supporting Fig. 1. Ajuba LIM-domains function as a co-repressor. *A*, Transient transcription assay in 293T cells with the reporter in *IA*, and expression constructs encoding wild type Gfi1 or Ajuba, and Ajuba-Pre-LIM, Ajuba-LIM1, Ajuba-LIM2,3 and Ajuba-LIM-3. Data shown as relative CAT activity and fold repression. Statistical analysis compared values of Gfi1 alone to those of Gfi1 with Ajuba (and mutant) cotransfection. *P < 0.05

Supporting Fig. 2 **Gfi1 transcriptional repression activity is dependent upon histone deacetylase enzymes**. Transient transcription assay, *A*, and immunoblot analysis, *B*, in EL4 T cells treated with either vehicle or TSA for 24 hours, with the reporters in Figure *IA*. The relative CAT activity and fold repression corresponding to each increase in TSA treatment is compared to the preceding value. Antisera against Gfi1, HDAC1, or β -Actin were used for Immunoblot. *C*, Transient transcription assay in 293T cells cotransfected with reporters in Figure 1A, and expression vectors encoding Flag-epitope-tagged Gfi1 or the SV40SWAP Gfi1-SNAGdomain mutant. Cells were treated for 24 hours with the HDAC inhibitors TSA or Sodium butyrate, or vehicle control. The relative CAT activity and fold repression of TSA or Sodium butyrate-treated samples is compared to that of vehicle-treated Gfi1 control. *D*, Immunoblot analysis of whole cell lysates corresponding to *IC*, with anti-Flag monoclonal antibody.

Supporting Fig. 3. **Ajuba knock-down in U937 depresses** *GFI1. A*, TaqMan analyses on U937 cells transduced with three independent lentiviral shRNA specific for *GFI1*, *AJUBA* or a nontargeting control. Representative immunoblot demonstrates lower levels of GFI1 and AJUBA with one of three independent shRNA constructs.



Montoya et al. Supplementary Figure 1



Montoya et al. Supplementary Figure 2



Montoya et al. Supplementary Figure 3