ENCODE Antibody Validation Documentation
Transcription factor: GATA binding protein 2 (GeneID 2624)

From: Myers Lab, HudsonAlpha Institute for Biotechnology
Contact Person: Dr. Florencia Pauli (fpauli@hudsonalpha.org)

Transcription factor: GATA2 (GeneID 2624; ~50 kDa)

Antibody: GATA2 (CG2-96), Santa Cruz Biotechnology (sc-267)
Mouse monoclonal
Web: http://www.scbt.com/datasheet-267-gata-2-cg2-96-antibody.html

Validation 1: Immunoblot Analysis

For an antibody to meet ENCODE validation standards, a single band of the predicted size, or a band of no less than half the total signal, must be detected in a lane on a Western blot.

a. Vendor immunoblot analysis

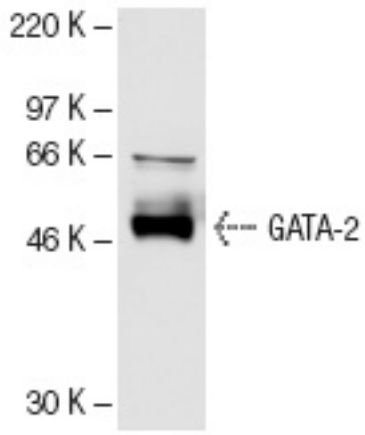

Figure Legend: Western blot analysis of GATA2 expression in TtT-97 nuclear extract.

b. Myers Lab immunoblot analysis

Western blot protocol

Whole cell lysates were immunoprecipitated using primary antibody, and the IP fraction was loaded on a 12% acrylamide gel and separated with a Bio-Rad PROTEAN II xi system. After separation, the samples were transferred to a nitrocellulose membrane with an Invitrogen iBlot system. Blotting with primary (same as that used for IP) and secondary HRP-conjugated antibodies was performed on an Invitrogen BenchPro 4100 system. Visualization was achieved using SuperSignal West Femto solution (Thermo Scientific).

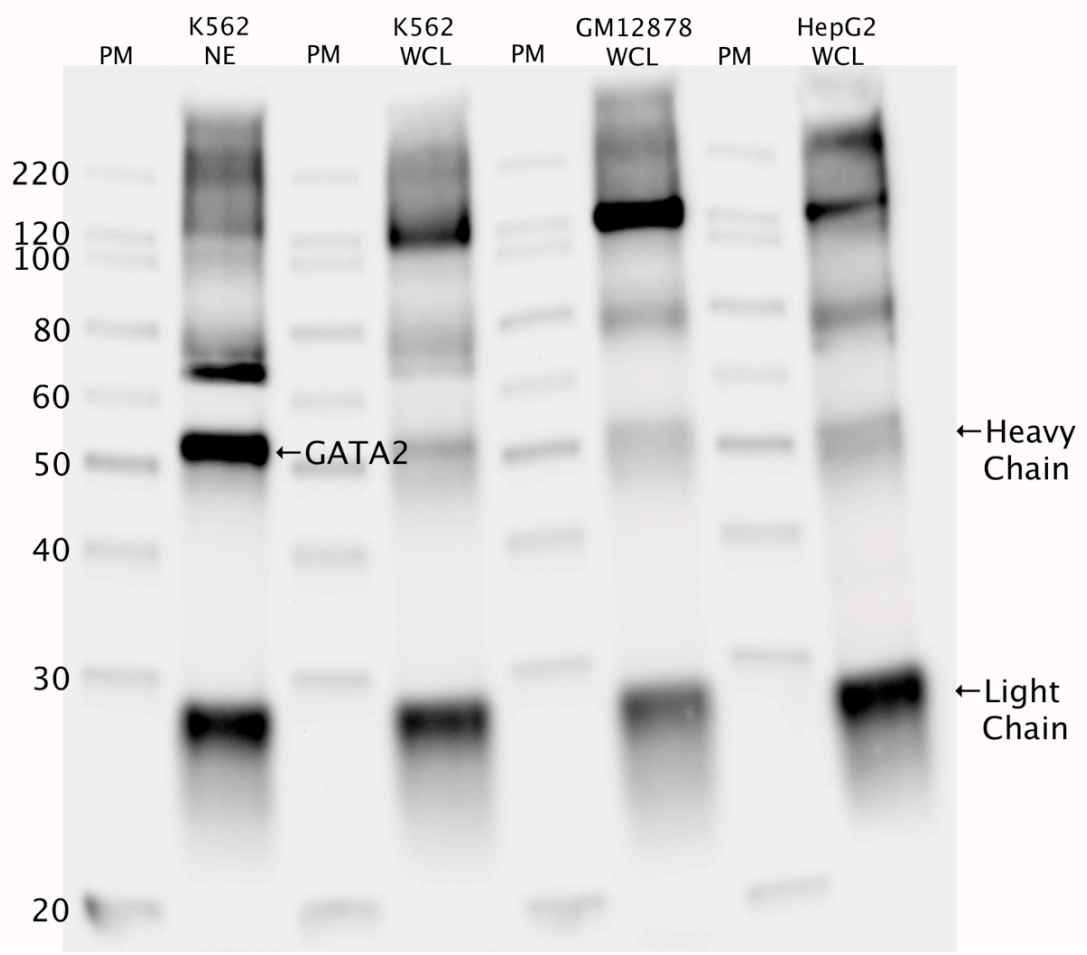

Figure Legend: GATA2 immunoblot: IP-western with sc-267 GATA2 antibody in nuclear lysate (NE) or whole cell lysates (WCL) of K562, GM12878, and HepG2. Heavy chain of IgG is indicated, and GATA2 band is indicated at ~50 kDa in K562 nuclear extract. GATA2 is expressed at very low levels in GM12878 and HepG2, and these lanes are included as negative controls.

Validation 2: In progress