



Figure S3. Transfer of tumor-derived gp96 to lymph node cells *in vivo*. (A and B) Gp96 was purified from gp96-EGFP transfected CMS5 cells using previously described methods (6). Two fractions of elution buffer from the DEAE column were analyzed by SDS-PAGE and stained with Coomassie Blue. Proteins on the gel were then transferred to an Immobilon-P membrane, blotted with anti-gp96 or anti-EGFP antibodies, and developed in X-ray films. The gel (A) and films (B) were imaged with the Kodak Image Station. (C) Following implantation of cells into the foot pad of BALB/c mice, draining lymph nodes were isolated and stained for lymphocytic marker CD45 and analyzed by flow cytometry for EGFP as shown in Figure 6. A representative plot shows the CD45 gated population. (D and E) Representative histogram shows the EGFP-containing CD45⁺ cells in the non-draining lymph node (ndLN) or draining lymph node (dLN) following implantation of CMS5 cells expressing gp96-EGFP, and RAP or an irrelevant protein, into BALB/c (D) or CD91^{-/-} (E) mice. (F and G) MFI of all mice from one experiment are shown. Denominator is the MFI (CMS5-EGFP before implantation) - MFI (untransfected CMS5 before implantation). These numbers are imported into the formula described in the Materials and Methods to obtain the normalized signal.