Supplemental Methods:

RNA extraction

RNA was extracted from confluent cells in T75 flasks or 6 well plates using the GenElute Mammalian Total RNA Kit (Sigma) or Trizol (Invitrogen). RNA concentration was determined by optical densitometry at 260/280 nm. Contaminating DNA was removed from RNA preparations with DNAse I using the DNA-free™ Kit (Ambion Inc).

qRT-PCR

First strand cDNA was synthesized from 1-2 µg of total RNA using SuperScript II Reverse Transcriptase (Invitrogen) with random hexamer primer. The PCR reaction included SYBR green PCR Master Mix (Applied Biosystems), primers, and the cDNA template. Real-time RT-PCR analysis was performed in an ABI Prism 7700 sequence detection system according to the manufacturer's instructions (Applied Biosystems). RT-PCR primers were designed with Primer-Express 2.0 (Applied Biosystems) or obtained from the literature. Primers used in RT-PCR are as per Supplementary Table. Target gene expression was normalized by subtracting the critical threshold (C_T) of the endogenous control (18s rRNA) from that of the target gene (ΔC_T). For intergroup comparisons, the relative change of the target gene equals $2^{\Delta\Delta CT}$, where $\Delta\Delta C_T = \Delta C_T$ of the tested sample (ex. *epi* $^{-/-}$) - ΔC_T of the control sample (ex. WT sample). Each qRT-PCR was performed in triplicate using three independent RNA isolates.

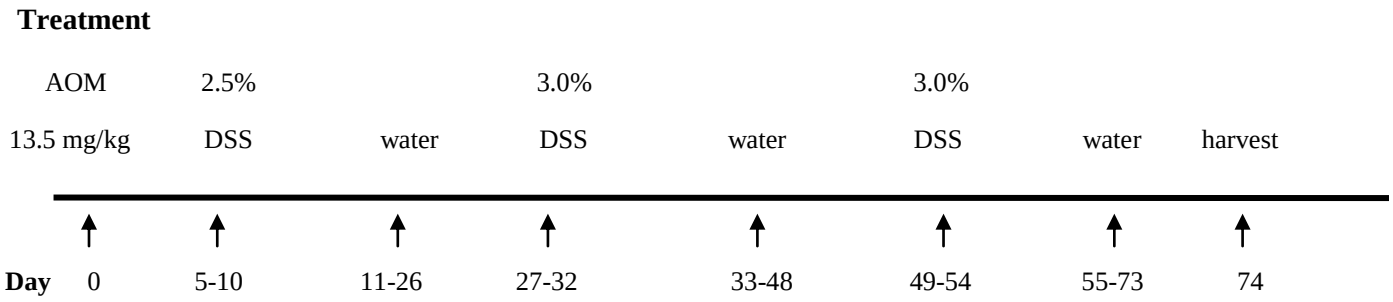
Immunoblot analyses

Protein concentrations were determined by using Bio-Rad D_c Protein Assay (Bio-Rad). Samples were prepared for immunoblot analysis as per (19) and briefly as follows: Aliquots of 50-100 µg of protein were separated on a 4-12% Bis Tris gels and subsequently electrotransferred for 2.5 hours onto PVDF membranes in NuPAGE Transfer Buffer (Invitrogen). After transfer, the

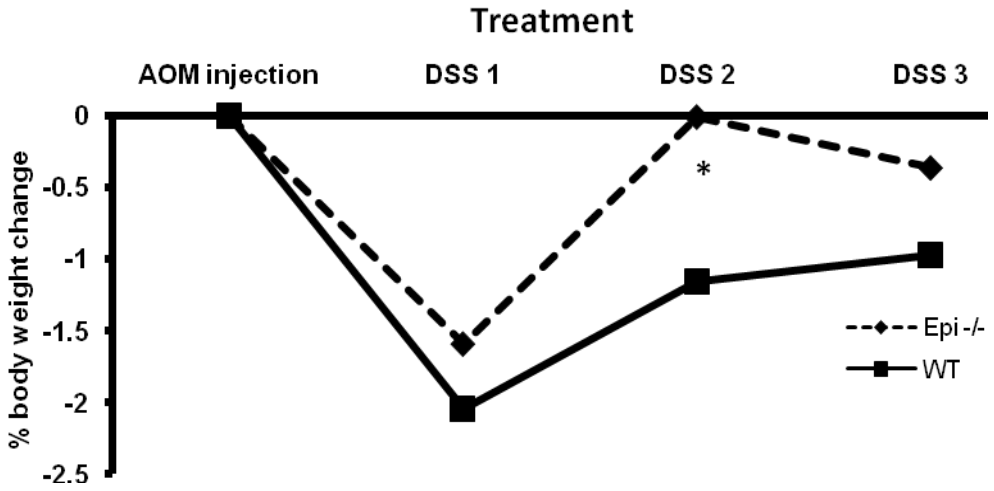
PVDF membrane was incubated overnight at 4ºC in blocking solution (150 mM NaCL, 10 mM Tris-HCL, .05% Tween-20, 5% non-fat dry milk, 5% bovine serum albumin) and then incubated 2.5 hours with polyclonal rabbit anti-mouse syntaxin 2 (epimorphin; 1:1000; Synaptic Systems). After reaction with the secondary antibody, PVDF membranes were treated with enhanced chemiluminescent agent (Amersham Biosciences). Prestained molecular mass markers were included in each gel. Blots were sequentially probed with smooth muscle α-actin (ab5694 Abcam) to control for differences in loading.

ELISAs

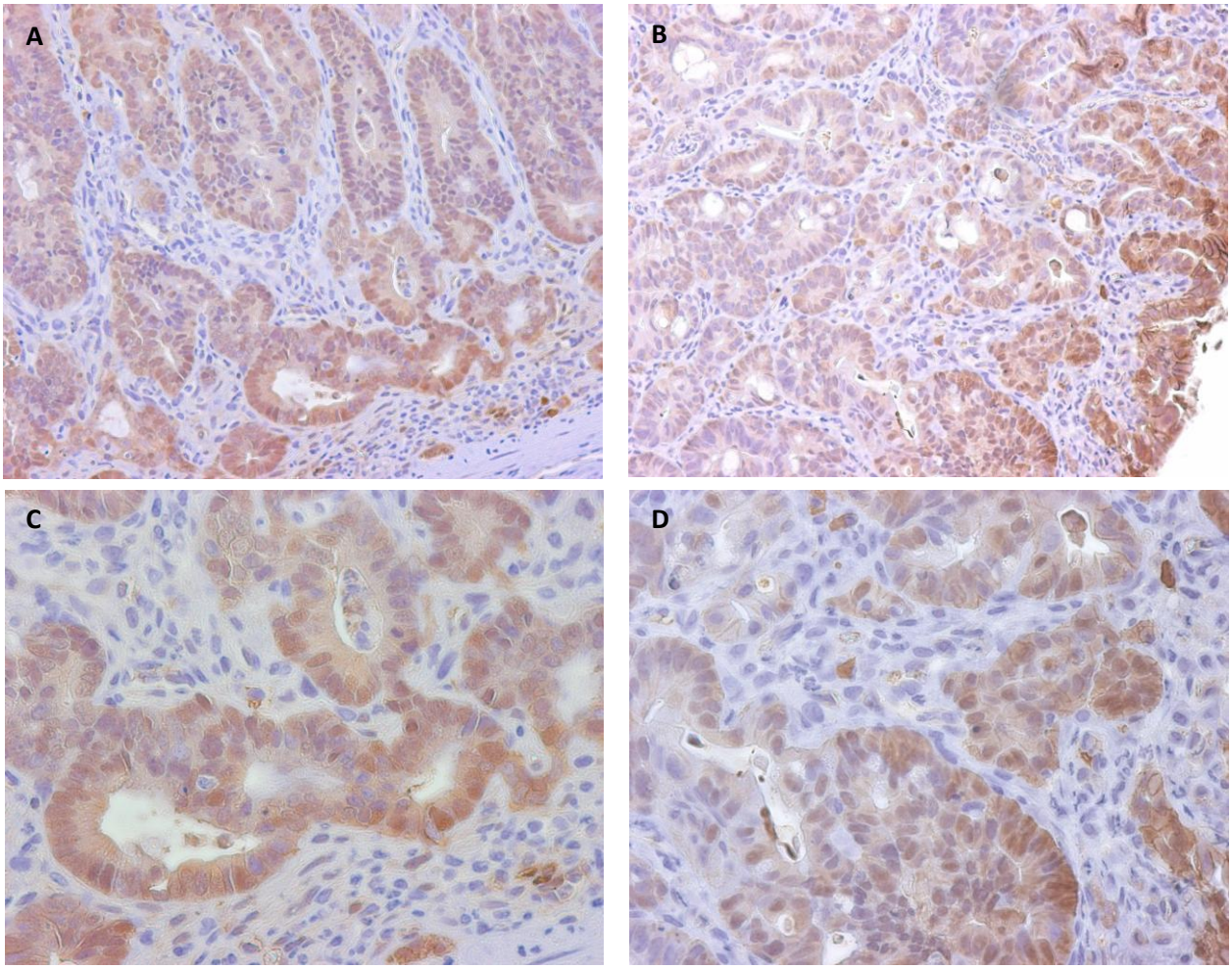
5×10^6 *epi*$^{-/-}$ and WT colon myofibroblasts were plated in T75 flasks. At 80% confluence, cells were switched to serum free media for 24 hours. Supernatants and cell lysates were harvested for measurement of BMP4 and Chordin by ELISA (R&D Systems). Results were expressed as pg of protein/ml. ELISA for TNF-α (BD Biosciences) and IL-6 (eBioscience) was performed per manufacturer's instructions. Results were expressed as pg of cytokine/ ml and pg of cytokine/μg protein.



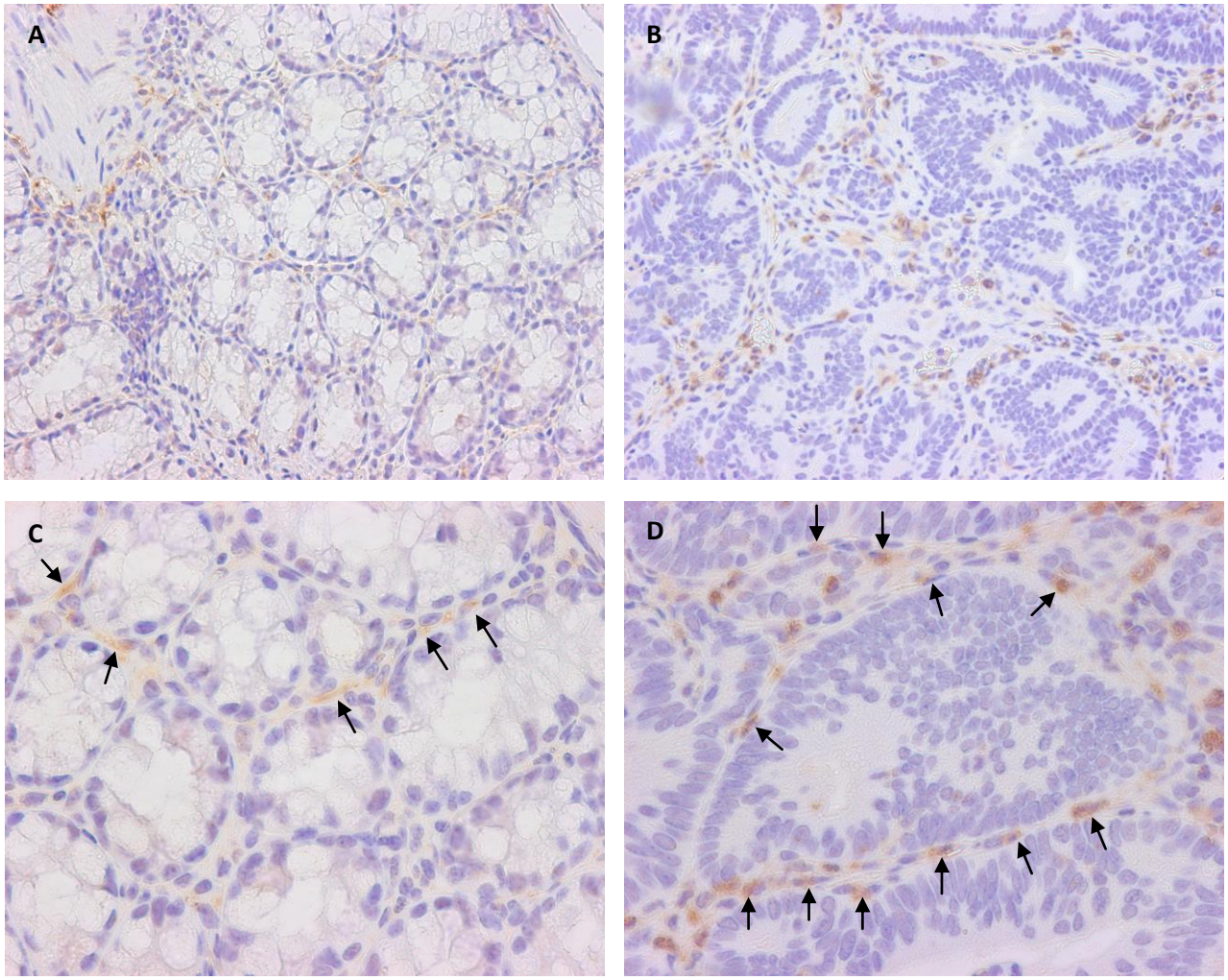
Supplemental Figure 1. Azoxymethane (AOM)-dextran sodium sulfate regimen for inflammation-induced colitis. *Epi*$^{-/-}$ and WT mice were treated with AOM (13.5 mg/kg) on day 0. Three cycles of treatment with DSS were completed prior to sacrifice of mice on day 74 of the protocol. Beginning on day 5-10, mice received 2.5% DSS in drinking water, followed by two weeks of water. Between days 27-54, mice received 3.0% DSS, followed by water for two more weeks, and then a third cycle of 3.0% DSS.



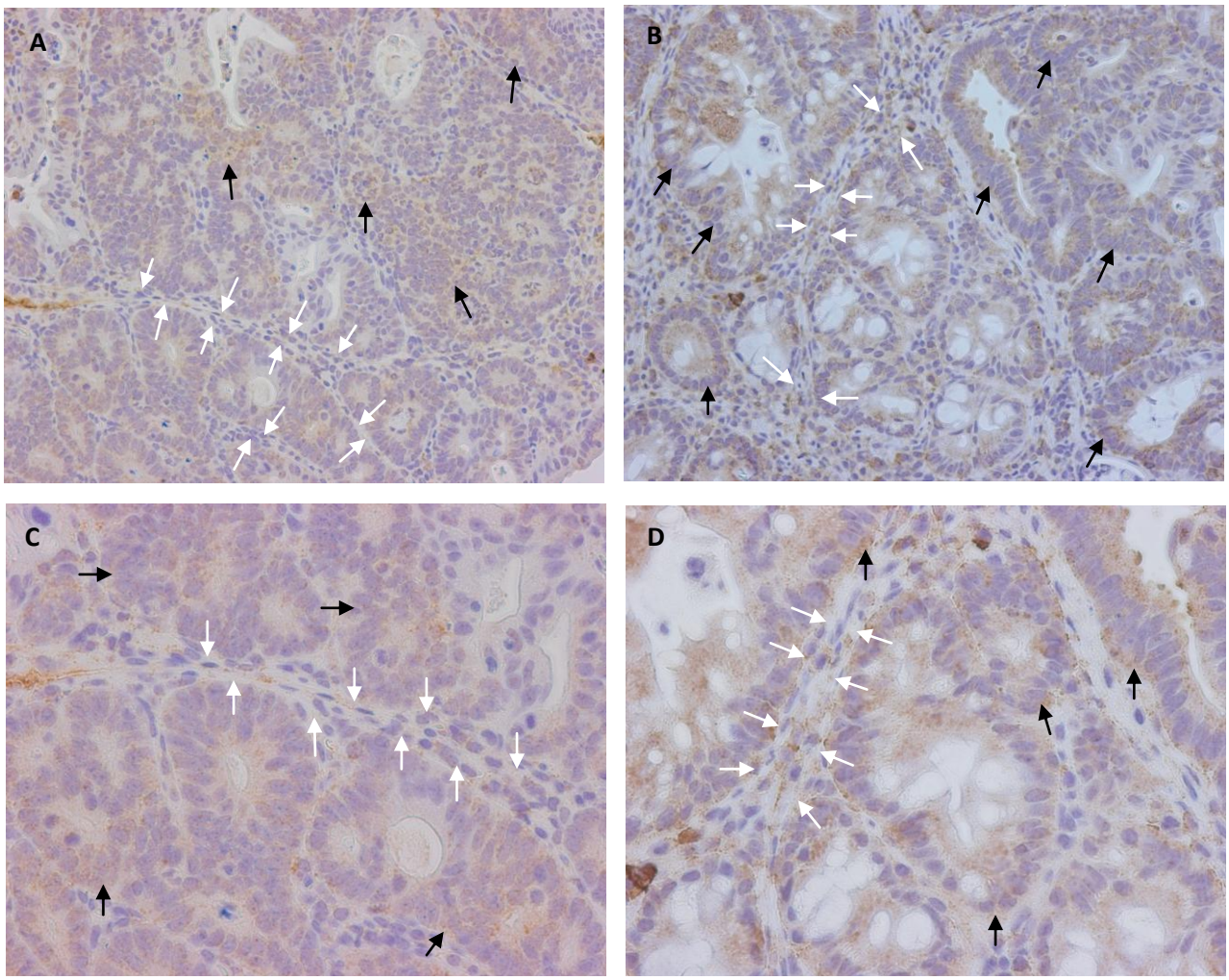
Supplemental Figure 2. Weight loss resulting from AOM/DSS treatment is ameliorated in *epi*$^{-/-}$ mice. The percentage body weight change was measured after each cycle of DSS. Data are expressed as mean percentage body weight loss for *epi*$^{-/-}$ (n=11) compared to WT (n=11) mice. *p<0.05.



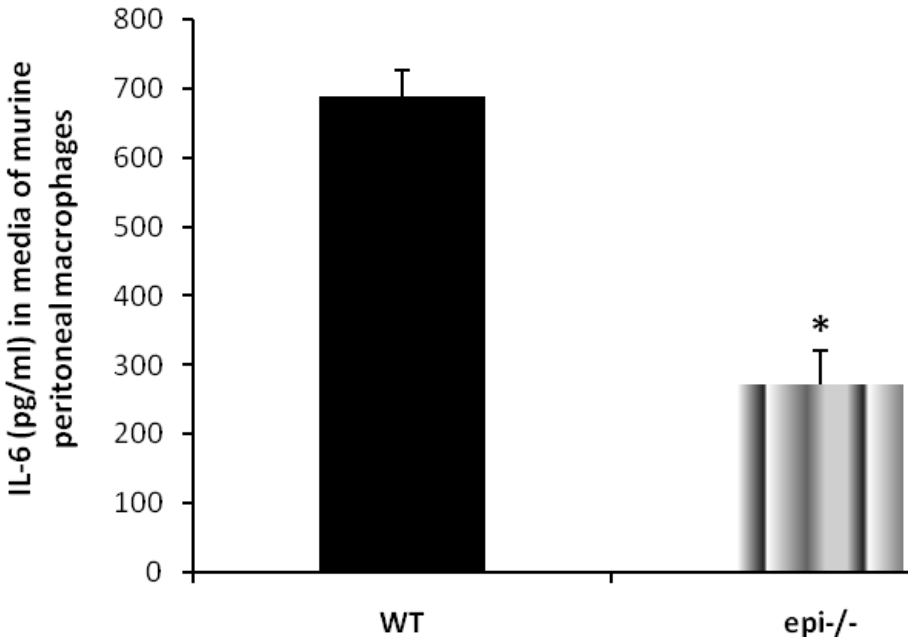
Supplemental Figure 3. **Nuclear β-catenin expression in AOM/DSS treated dysplastic WT and *epi*$^{-/-}$ colon.** Nuclear β-catenin expression in dysplastic WT (**A, C**) and *epi*$^{-/-}$ (**B, D**) colon was detected using a polyclonal rabbit anti-β-catenin antibody. The pattern of nuclear β-catenin expression is similar in WT and *epi*$^{-/-}$mice dysplasia (**A, B** X 200; **C, D** X 400).



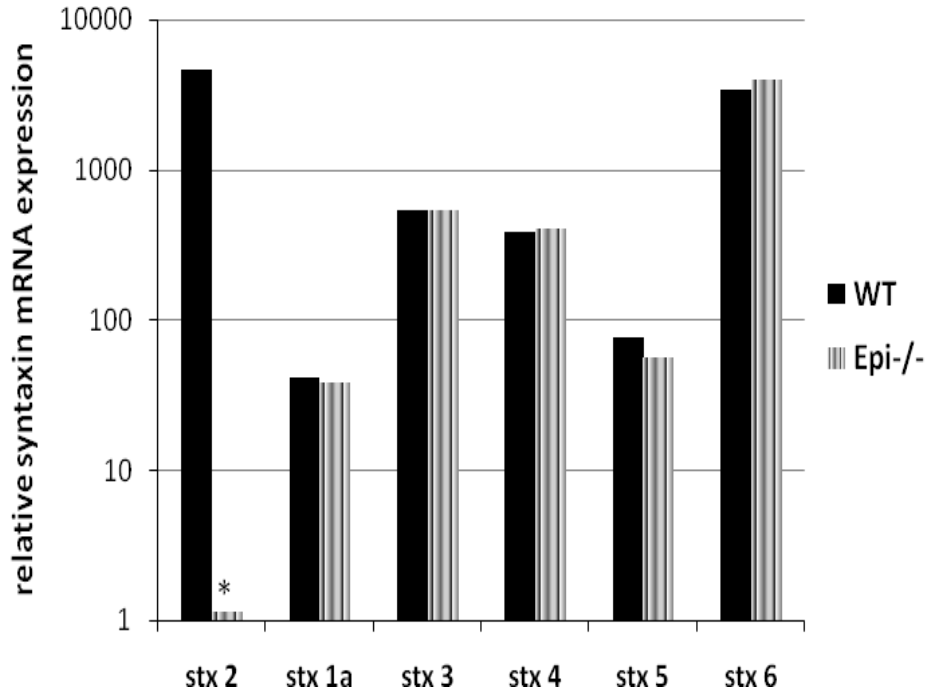
Supplemental Figure 4. **Epimorphin/Syntaxin-2 expression in normal and AOM/DSS treated WT colon**. Epimorphin expression in sections of untreated WT descending colon (**A, C**) and AOM/DSS treated WT descending colon (**B**, **D**) was analyzed using a polyclonal rabbit anti-syntaxin 2 antibody (1:500) (**A, B** X 200; **C, D** X 400).



Supplemental Figure 5. BMP expression in WT and *epi*$^{-/-}$ AOM/DSS treated colon. Sections of descending colon were incubated with polyclonal goat anti-recombinant human BMP2/4 antibody (1:300) that cross reacts with mouse. Antigen-antibody complexes were detected with biotinylated rabbit anti-goat IgG and streptavidin-horseradish peroxidase. Representative sections of WT (**A, C**) and *epi* $^{-/-}$ (**B, D**) dysplastic colon are shown. Epithelial (**black arrow**) and stromal (**white arrow**) BMP expression is more pronounced in *epi*$^{-/-}$ compared to WT colon (**A, B** X 200; **C, D** X 400).



Supplemental Figure 6. IL-6 secretion from WT and *epi*$^{-/-}$ peritoneal macrophages. IL-6 secretion from peritoneal macrophages isolated from *epi*$^{-/-}$ mice is diminished compared to IL-6 secretion from peritoneal macrophages from WT mice. Murine peritoneal macrophages were isolated from WT and *epi*$^{-/-}$ age-matched mice as described in Methods. IL-6 secretion was measured after 24 h in culture by ELISA. IL-6 secretion is higher from WT compared to *epi*$^{-/-}$ peritoneal macrophages (689 pg/ml vs. 272 pg/ml, *$p<0.05$).



Supplemental Figure 7. **Syntaxin expression in WT and *epi*$^{-/-}$ colon myofibroblasts.** Syntaxin-2 (stx2)/epimorphin is not expressed in *epi*$^{-/-}$ myofibroblasts (*$p < 0.05$; n=3 independent experiments) compared to WT myofibroblasts. Deletion of epimorphin does not affect the expression of several other syntaxins.