

Supporting Information

Mirakaj et al. 10.1073/pnas.1015605108

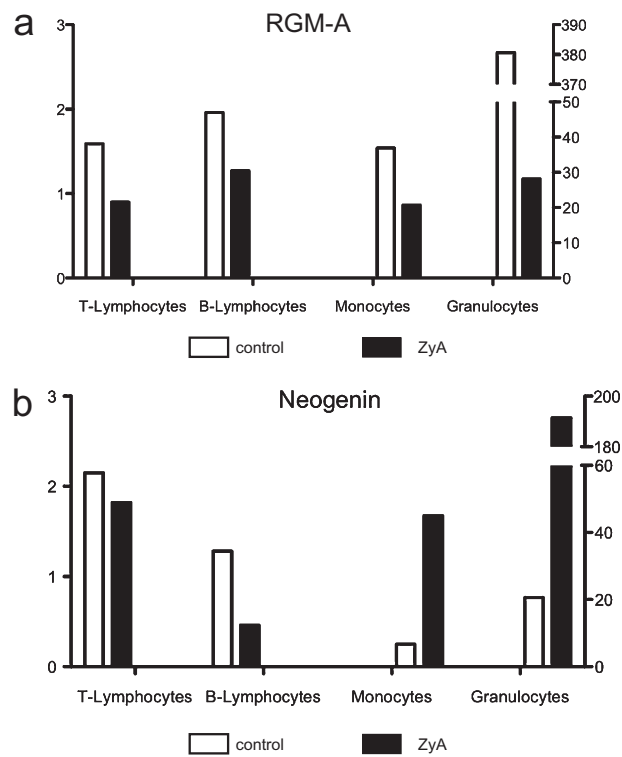


Fig. S1. Stimulus induced regulation of RGM-A (A) and neogenin (B) expression. Splenocytes were stimulated with 10 $\mu\text{g}/\text{mL}$ ZyA for 24 h and then differentially analyzed for T and B lymphocytes (left axis) and monocyte and granulocyte subsets (right axis) expressing RGM-A (A) or neogenin (B), presented as mean fluorescent intensity after subtraction of isotype control levels. In response to perceived inflammatory stimulation, leukocytes down-regulate RGM-A expression, contrasted with induced expression of neogenin by PMNs and monocytes. Data are representative of two experiments.

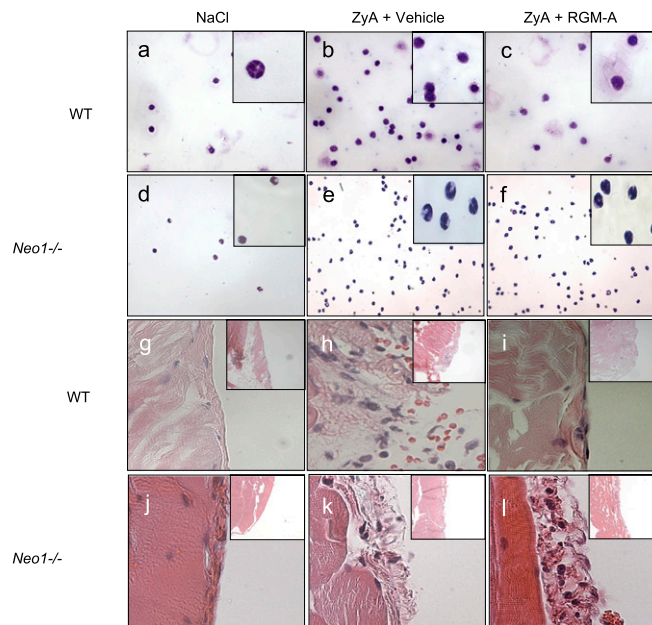


Fig. S2. RGM-A does not attenuate morphological signs of inflammation in *neo1*^{-/-} mice. Exudate cellularity and morphology evacuated from the inflamed peritoneum at 4 h in WT mice and *neo1*^{-/-} mice after vehicle (Left), ZyA (Middle), or combined ZyA and RGM-A injections (Right). Representative histological analysis of the peritoneal cavity in WT mice compared with *neo1*^{-/-} mice after vehicle (Left), ZyA (Middle), or ZyA injection with concomitant RGM-A application (Right). (A–F) No difference in leukocyte accumulation between WT and *neo1*^{-/-} mice was detected after either vehicle (NaCl) (A and D) or ZyA (B and E) injection, showing that *neo1*^{-/-} mice have a similar susceptibility to ZyA peritonitis as WT mice. The application of RGM-A led to a robust decline in infiltrating PMNs accumulating at the inflammatory lesion site in WT mice (C). In contrast, this robust biological effect was absent in the *neo1*^{-/-} mice (C–F). A similar effect was detected in the peritoneal cavity (G–I). Whereas RGM-A reduced intraparenchymal leukocyte accumulation and spongiotic edema formation in WT mice (H and I), *neo1*^{-/-} mice developed a full-blown course of inflammation irrespective of concomitant RGM-A applications (K and L). All slides were prepared and stained with H&E. RGM-A–induced suppression of ZyA-elicited peritonitis was detected in WT mice, but not in *neo1*^{-/-} mice (original magnification 400×; Inserts, 1,000×).