



**Supplementary information, Fig. S5** The two enzymes iGb3S and Hexb are involved in the generation of iGb3 in macrophages after TLR activation. **a** Q-PCR analysis of *iGb3S* and *Hexb* mRNA expression in peritoneal macrophages stimulated with LPS for the indicated times. **b** Q-PCR analysis of *iGb3S* and *Hexb* mRNA expression in peritoneal macrophages pretreated with inhibitors of ERK, JNK, p38 or NF- $\kappa$ B for 30 min and then stimulated with LPS for the indicated times. **c** ELISA of cytokines in supernatants of macrophages transfected with control siRNA, iGb3S siRNA or Hexb siRNA, and 48 h later, stimulated with LPS, Poly(I:C) or CpG for 6 h. **d** ELISA of TNF and IL-6 in supernatants of peritoneal macrophages stimulated with different GSLs for 6 h. **e** ELISA of cytokines in supernatants of macrophages transfected with control siRNA, Gb3S siRNA or Gb4S siRNA, and 48 h later, stimulated with LPS, Poly(I:C) or CpG for 6 h. Data are shown as mean  $\pm$  SEM and from three independent experiments. \* $P$  < 0.05; \*\* $P$  < 0.01 (Student's  $t$ -test or ANOVA).