Figure 1S

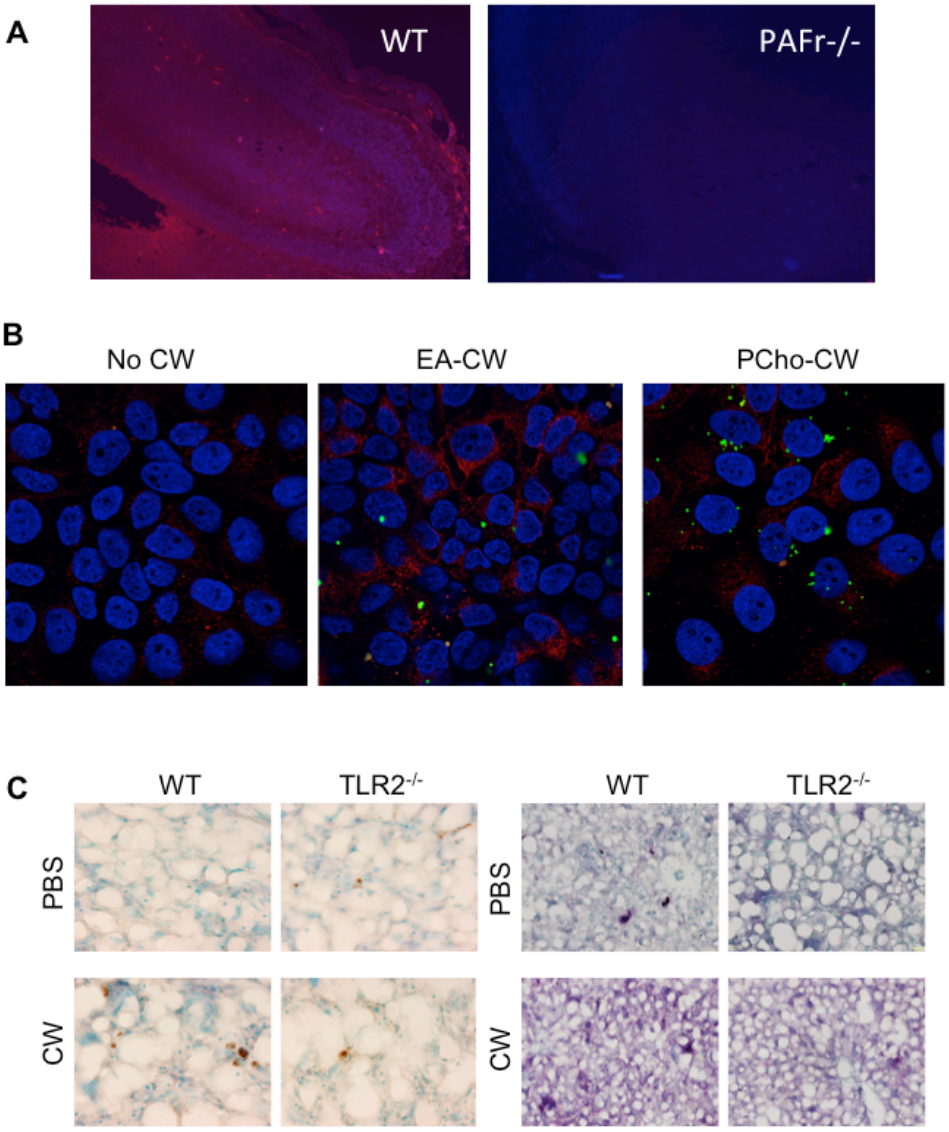

Figure 1S, related to Figure 1. *Immunohistochemical analysis of interaction of placenta and placental cells with cell wall.* A) PAFr distribution in the fetal brain was documented by treatment of fixed sections from mice overlaid with anti-PAFr antibody and counterstained with Prolong Gold (red). 20 X magnification. B) JEG-3 trophoblast cell line was exposed for 48 hours to PBS, FITC labeled wild type cell wall with choline teichoic acid (PCho-CW) or FITC labeled cell wall with ethanolamine replacing choline in the teichoic acid (EA-CW). Cells were stained with DAPI (Blue) for nuclei; anti-PAFr antibody (Red), and FITC (green) for cell wall. 63X magnification. C) WT and TLR2$^{-/-}$ mice were exposed to PBS or CW IV at E15 and the placenta was harvested 24 hours later. Sections of placenta were stained for TUNEL (left panel; brown) and caspase 3 (right panel; purple). 40 X magnification. Quantitation is presented in Figure 1 panels B and C respectively.

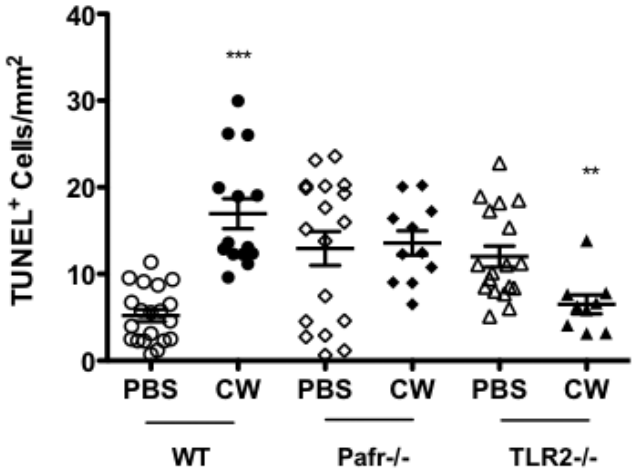

Figure 2S, related to Figure 2. *Cell wall induced death in fetal brain in WT, PAFr$^{-/-}$ and TLR2$^{-/-}$ mice.* WT, TLR2$^{-/-}$ and PAFr$^{-/-}$ pregnant mice (n = at least 4/group repeated twice; each symbol is mean of 3 slices from one embryo) were challenged with 2 x 10^7 bacterial equivalents of cell wall IV at E15. Fetal brains were harvested within 15 min (0 hours) or 24 hours post injection. Fixed sections of the cortex were stained with TUNEL for cell death.

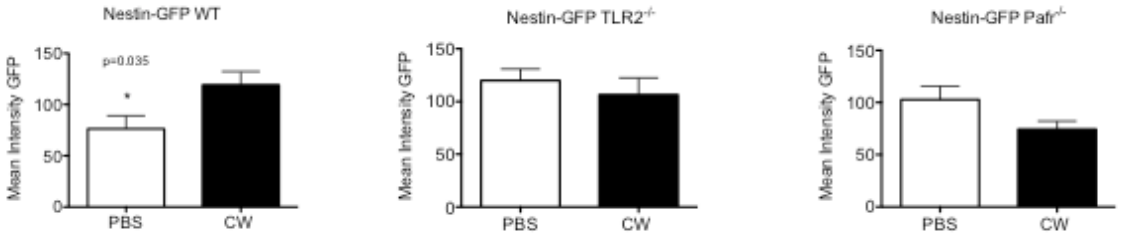

Figure 3S, related to Figure 3D. *Quantitation of GFP staining intensity for Nestin in fetal cortex.* Images shown in Figure 3D were subjected to measurement of fluorescence intensity using Image J. Mean intensity is shown for 2 images per each of at least 5 embryos/group.

Figure 4S

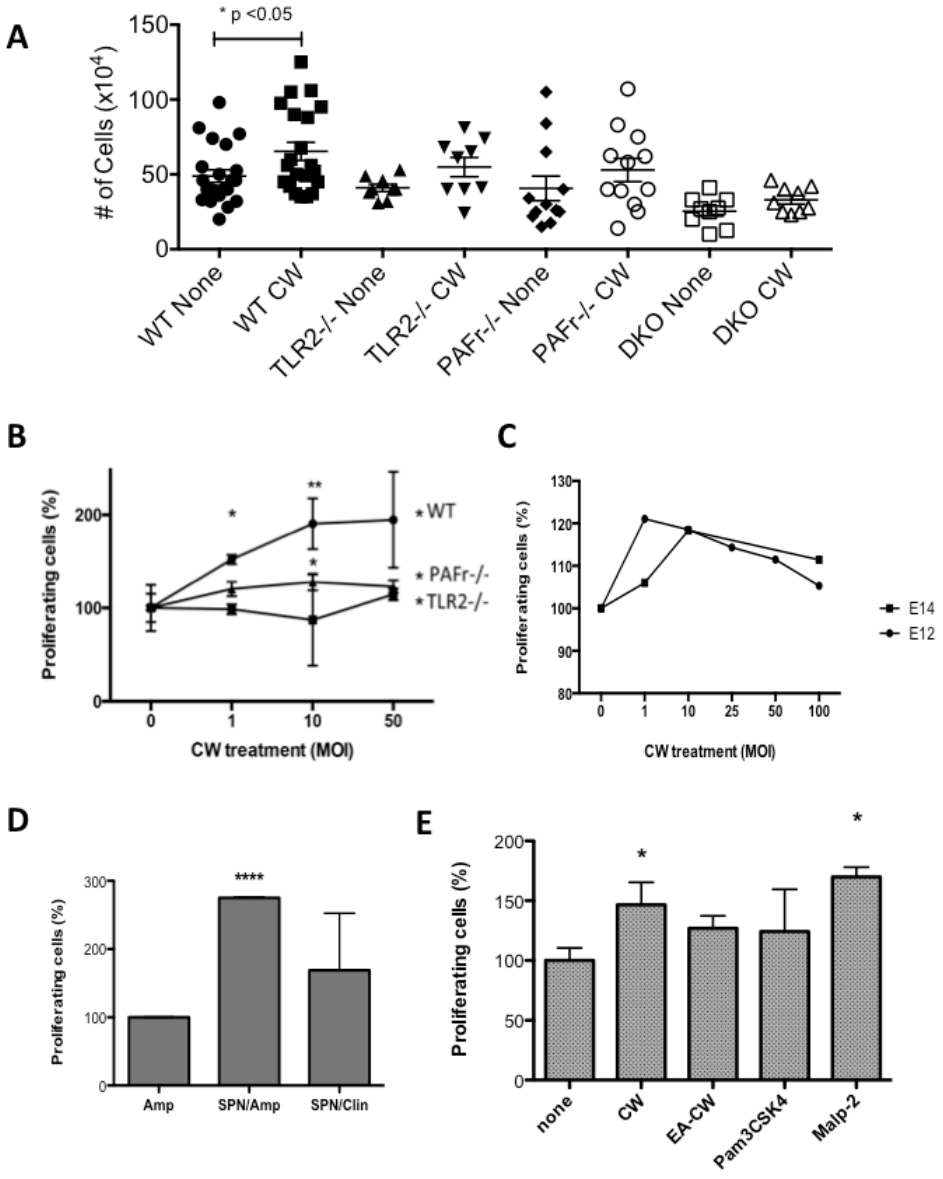

Figure 4S, related to Figure 4. *Quantitation of proliferation in vitro of NPCs of varying genotype and cell wall preparation.* A) NPCs were prepared from E16 fetal brains and at 24 hrs, they were challenge with CW or PBS (none). At day 3, cells were harvested and counted visually by microscopy (each symbol is mean of NPCs from one embryo tested in triplicate wells; n= at least 8 embryos/group combined from 2 experiments). B) NPCs as in A were labeled with CFSE, challenged with different doses of CW and proliferation quantified at 24 hours by FACS. C) NPCs were harvested from E12 or E14 and proliferation was measured by incorporation of BrDU. (% proliferation over time is charted since NPC harvest on different days of embryogenesis yields different cell number). D) NPCs prepared as in A were exposed to living *S. pneumoniae* and immediately treated with ampicillin or clindamycin. Proliferation was measured by CSFE FACS analysis at 3 days. E) Quantitative analysis of FACS data from Fig 4A,B, C.

Figure 5S

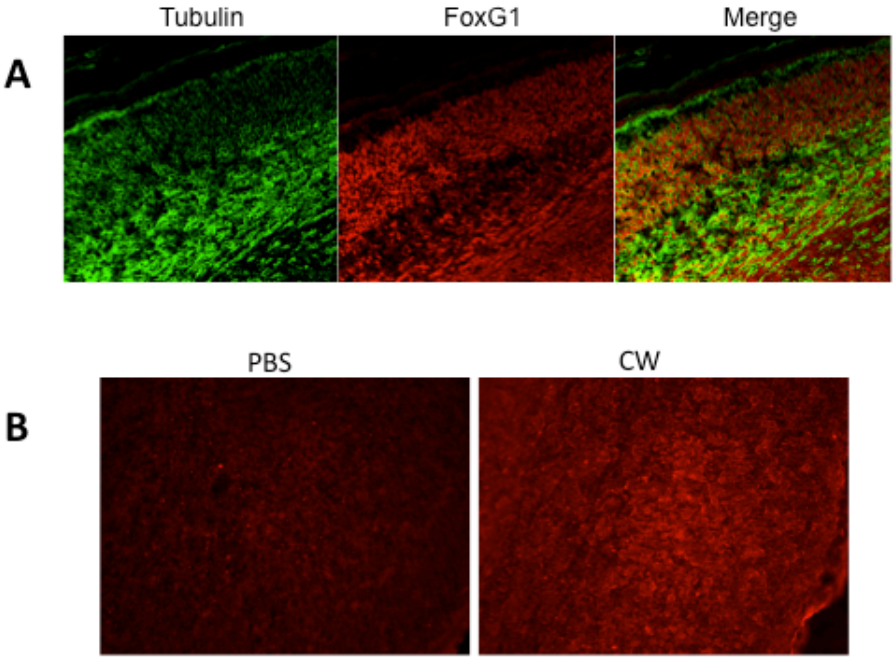

Figure 5S, related to Figure 5. *FoxG1 colocalization with neuronal marker and induction of PI3kinase*. A) WT embryos were challenged with CW and assessed by confocal imaging as in Figure 5B. FoxG1 (red) localized to the same cells expressing the neuronal marker Tubulin (green; 40X magnification) but did not overlap in the same cellular compartment consistent with FoxG1 in the nucleus and Tubulin in the cytoplasm. Images are representative of at least 2 independent experiments. B) WT embryos were treated as in A and brain sections were stained by anti-PI3kinase antibody (red). Representative of 4.

Figure 6S

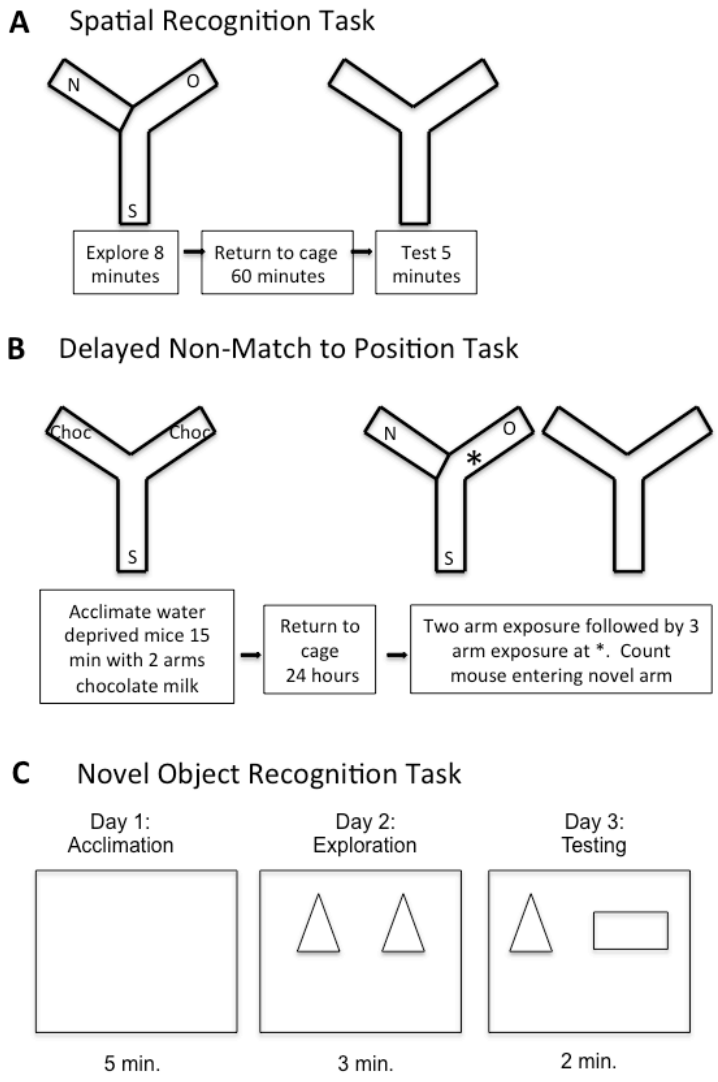

Figure 6S, related for Figure 6. *Models for effect on postnatal behavior of early embryonic exposure to CW*. (A) Spatial Recognition Task: Mothers were challenged at E10 and pups were tested at 5 months of age by placing them in a Y maze (S=start) for 8 min with one arm blocked (N) and one arm open (O) as indicated. One hour later, they were again placed in the maze with both arms open and the time spent exploring the novel (N) vs old (O) arm was measured as a % of total exploration time (5 minutes). (B) Delayed Non-match to Position Task: Using the same Y maze as (A), water-deprived mice were acclimated to explore the maze with chocolate milk in both arms (Choc). 24 hours later, they were placed in the maze at S without chocolate milk. Upon entry into the open arm (*), they were quickly replaced at the start S and the novel arm (N) was opened. Number of entries into the novel arm was measured as a % of total exploration time. (C) Novel Object Recognition: Mothers were challenged at E10 or E15 as indicated and pups were tested at 3 months of age for memory and anxiety behavior. All mice underwent acclimation to the testing chamber on Day 1. On Day 2 all mice were allowed to explore the chamber with 2 identical objects. On Day 3 all mice were tested for the time exploring the chamber containing one old object and one new object. Time was counted in seconds starting when the mice turned their nose toward the object.