Supplementary Material

Modulation of Human Beta-Defensin 2 Expression by Pathogenic *Neisseria meningitidis* and Commensal Lactobacilli

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Figure S1

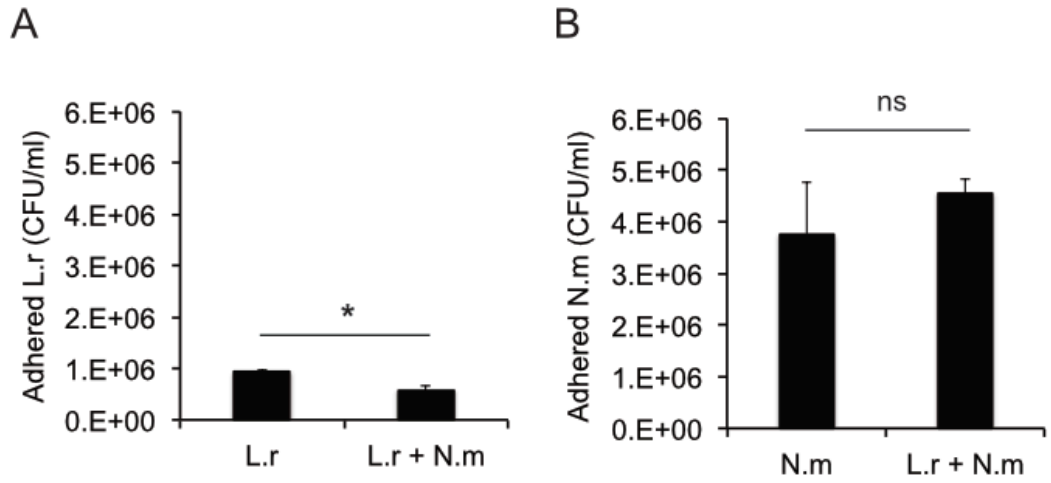

Figure S1. ***N. meningitidis*** **affects** ***L. reuteri*** **adhesion to host cells during coincubation.**

Pharyngeal epithelial cells were incubated with *L. reuteri* and *N. meningitidis*, either alone or coincubated, at an MOI of 100 for 6 h. Unbound bacteria were removed by washing. Bound bacteria were released by saponin treatment. The adhesion of *L. reuteri* (A) and *N. meningitidis* (B) to host cells was determined by viable count. Data are represented as the mean values, with error bars representing the standard deviation. Both assays were performed in triplicate at least three times. Significance was tested as indicated. **P*<0.05; ns, nonsignificant; L.r, *L. reuteri*; N.m, *N. meningitidis*.

Figure S2

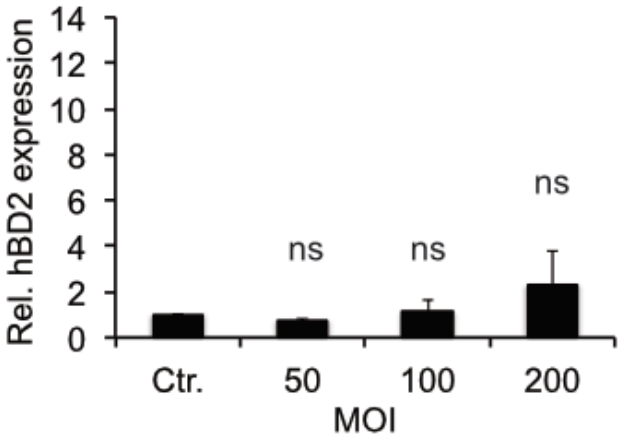

Figure S2. Expression of hBD2 after infection with *N. meningitidis*.

Gene expression of hBD2 in pharyngeal epithelial cells incubated with *N. meningitidis* at an MOI of 50, 100 or 200 for 6 h. Expression was quantified using qPCR, normalized against the housekeeping gene β-actin and expressed as the fold change compared to the control. Data are represented as the mean values, with error bars representing the standard deviation. The assays were performed in triplicate at least three times. Significance was tested against the control, or as indicated. *P<0.05; ns, nonsignificant; Rel., relative; Ctr., control without bacteria.

Figure S3

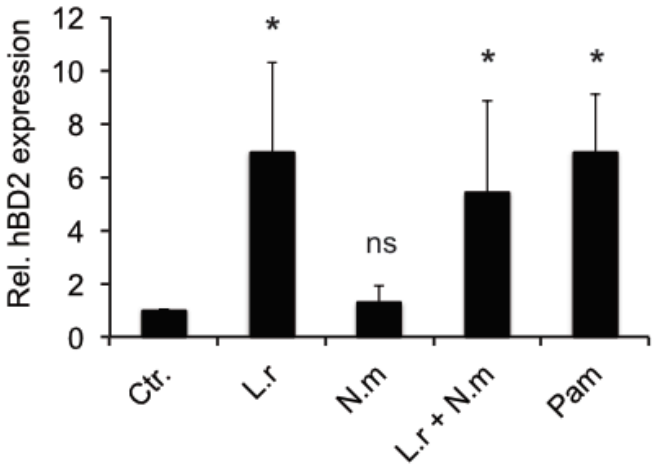

Figure S3. Validation of hBD2 expression in pNiFty-transfected epithelial cells.

Pharyngeal epithelial cells were transfected with the inducible NF-κB reporter plasmid pNiFty for 6 h. The cells were maintained for a further 48 h before incubation with *L. reuteri* and *N. meningitidis*, either alone or coincubated. Pam3CSK4 stimulation was used a positive control. Gene expression of hBD2 was quantified using qPCR. Expression was normalized against the housekeeping gene β-actin and expressed as the fold change compared to the control. Data are represented as the mean values, with error bars representing the standard deviation. The assay was performed in duplicate at least three times. Significance was tested against the control. Rel., relative; Ctr., control without bacteria; L.r, *L. reuteri*; N.m, *N. meningitidis*; Pam, Pam3CSK4.

Figure S4

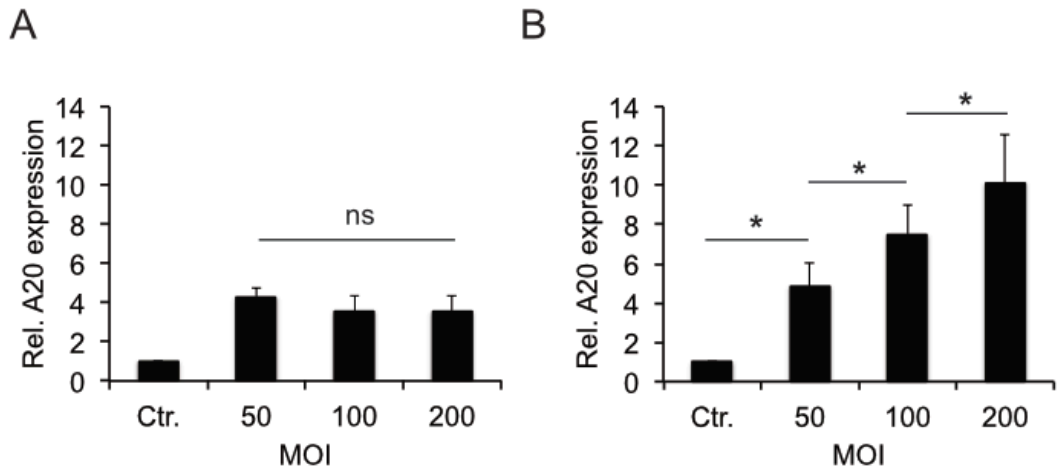

Figure S4. A20 expression in response to *L. reuteri* and *N. meningitidis* at different MOIs.

Pharyngeal epithelial cells were incubated with *L. reuteri* (A) or *N. meningitidis* (B) at an MOI of 50, 100 or 200 for 6 h. Gene expression of A20 was quantified using qPCR. Expression was normalized against the housekeeping gene β-actin and expressed as the fold change compared to the control. The assays were performed in triplicate at least three times. Data are represented as the mean values, with error bars representing the standard deviation. Significance was tested as indicated. Rel., relative; Ctr., control without bacteria.