S3. Plantain NSP inhibits adhesion to, and invasion of Caco2-cl1 cells by adherent, invasive E. coli (AIEC).

Three adherent, invasive Crohn’s disease E. coli isolates (HM605, HM615 and LF82) and the non-pathogenic reference strain E. coli K12 were selected to examine the effect of plantain NSP on the ability of E. coli to adhere to, and invade the Caco2-cl1 cell-line which was to be used to generate M-cells. Briefly, Caco2-cl1 cells were seeded (5 x 10^4 cells per 1 ml well) to 24-well tissue culture plates (Corning-Costar; High Wycombe, UK) and maintained in culture. Following pre-treatment of confluent cells for 30 min, with or without plantain NSP (0 to 50 mg/mL, in DMEM without antibiotics), each monolayer was infected for 4 h with approximately 1 x 10^6 bacteria per well (MOI 10:1). Adherence and invasion was assessed using a gentamicin protection assay as previously described [Martín HM et al. Gastroenterology 2004;127:80-93.]

Caco2-cl1 epithelial cell invasion by all three colonic and ileal AIEC isolates investigated was inhibited by the presence of plantain NSP both at 5 and 50 mg/mL. For E. coli K12, significant inhibition of invasion was only observed at plantain NSP levels of 50 mg/mL. Similarly, bacterial adhesion to Caco2-cl1 cells by all E. coli isolates assessed was also significantly blocked by the presence of plantain NSP at either 5 or 50 mg/mL. *, P<.05; **, P<.01; and ***, P<.001; ANOVA (n=3).