SUPPLEMENTAL FIGURES



Figure S1 – Microbiota analyses, related to Figure 1, 2 and 3. (A) Principal coordinates analyses (PCoA) of the microbiota in R-colonized and HypoR-colonized mice. PCoA of the Bray-Curtis dissimilarities between mouse fecal microbiota sampled at the ends of the experiments depicted in Figure 1A show the disparate effects of the diet supplement on the gut bacterial communities of R-colonized and HypoR-colonized mice. The first principal coordinate separates the communities by diet for the HypoR mice but the R-colonized mice do not cluster along either the first or second axes. This reflects the results of permutational multivariate analyses of variance showing that diet explains 5.3% of the variance in Rcolonized mice and 28.9% of the variance in HypoR-colonized mice. Mice from separate trials are indicated by different shapes in the scatterplots. (B) BugFACS analysis of feces obtained from Rcolonized mice in the experiment shown in Figure 1. Bubble-plot shows the degree to which particular ASVs are enriched in either the IgA- or IgA+ fractions recovered by FACS of fecal microbiota samples collected just prior to euthanasia from mice fed the indicated diets. The strength of enrichment is depicted by the size of the bubble, with red representing ASVs enriched in the IgA+ fraction and blue representing ASVs enriched in the IgA- fraction. Darker colors indicate lower P-values (Wilcoxon rank sums tests). Taxa highlighted in boldface are R community-derived invaders in the co-gavage experiment described in Figure 3A. (C) Successfully invading bacterial taxa (ASVs) across experiments. A heatmap shows ASVs that successfully invaded either the HypoR or R communities in the indicated experiments.



Figure S2 –Non-targeted LC-OTOF MS of cecal contents obtained from animals in the experiment shown in Figure 3. (A) Principal components analysis of nontargeted metabolomic profiles separates the HypoR, HypoR+5memRCC and HypoR+R treatment groups. The first two components explain 32.0% and 19.0% of the variance, respectively. (B) Heatmap showing the concentrations (centered and scaled to means of zero and unit variance) of the top 10% (n=82) of analytes with the strongest significant positive correlations with the CT-IgA ratio. Columns on the left show significant positive correlations with PC1 in red and significant negative correlations with PC2 in blue, as well as whether or not an analyte's mass and retention time matched that of an analyte observed in germ-free mice fed either the M18 or supplemented M18 diet. The percent relative abundances of ASVs assigned to the five members of the 5memRCC are shown below the heatmap. (C) Heatmap showing the standardized concentrations of analytes that differ between RCh-R, RCh-HypoR or HypoRch-R mice fed the M18 diet or the supplemented M18 diet in the cohousing experiment described in Figure 2A. (FDR-corrected Kruskal-Wallis tests followed by pairwise Wilcoxon rank-sums tests with P-values adjusted by Holm's method). All analytes included had masses and retention times that corresponded to the 346 analytes with significant, positive correlations with the CT-IgA ratio in the experiment described in Figure 3A. Columns on the left indicate whether the analytes (i) also matched compounds detected in germ-free mice fed the unsupplemented or supplemented M18 diet, or (ii) were among the 82 compounds from Panel B with Pearson's correlations ≥ 0.66 . (**D**) Heatmap showing the standardized concentrations of analytes in the supernatants of in vitro cultures of the 5memRCC and its individual members, as well as uninoculated controls. Minimum medium containing dissolved M18 diet pellets or supplemented M18 diet pellets are noted by brown or green colors respectively. The nine analytes shown matched m/z observed to have significant, positive correlations with the CT-IgA ratio in the *in vivo* experiment described in Figure 3A and were at least 3-fold more abundant in bacterial cultures compared to uninoculated media. The analyte highlighted by an arrow was identified by MS/MS as nicotinic acid riboside. The three m/z highlighted in bold face each had greater abundances in supplemented M18 cultures than in the M18 cultures of at least one bacterial strain or the 5memRCC.