Changes in alpha diversity over time in naïve vs. isoflurane-exposed mice

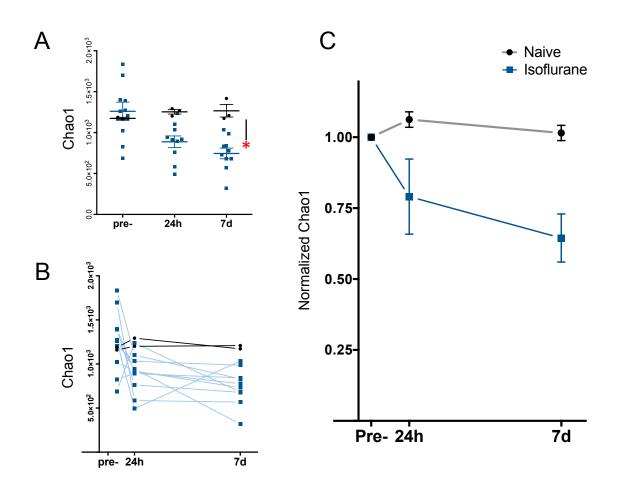


Figure S1. A) Comparison of Chao1 scores between naïve and isoflurane-exposed mice using Two-Way Anova demonstrates that isoflurane exposure, but not time, has a significant effect on microbial diversity (p=.0128). Additionally, post-hoc analysis using Bonferroni's correction shows that compared to naïve mice at the same time points, diversity in GA exposed mice is significantly reduced at 7d (Bonferroni adjusted p= .0103). Graphs represent means +/- SEM. **B)** Individual data points plotted for each mouse with data available at all 3 time points (naïve n=2, isoflurane n=10) **C)** As two-way Anova does not always accurately represent the effect of time, data from each mouse was normalized to the pre-exposure Chao1 score so that diversity at subsequent time points could be interpreted as a percentage change (fraction) from the baseline measurement. The normalized values were averaged across each group and plotted to more clearly illustrate this change (Mean +/- SEM).

Relative abundances of bacterial species (phyla) in naïve mice

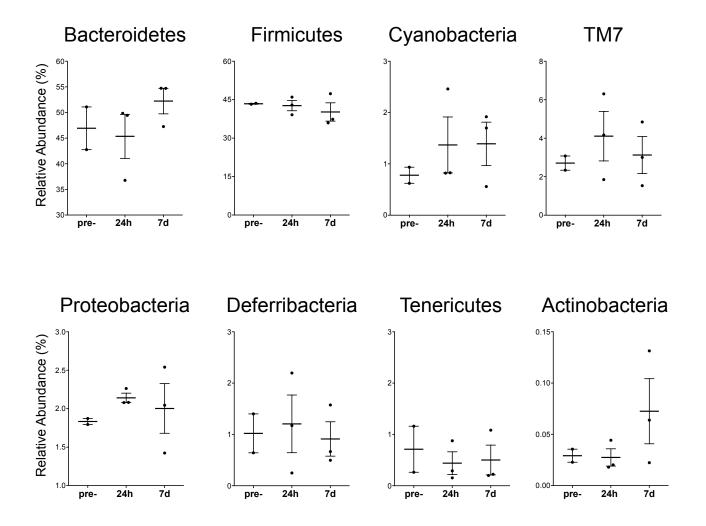


Figure S2: Relative abundances of taxa belonging to 8 most prevalent phyla do not change over time period studied in naïve cagemates (n=3, samples from 2 naïve mice were collected at time "pre", and from all 3 naïve mice at "24h" and "7d"). Analysis by ordinary one-way Anova with Bonferroni's post-hoc analysis to correct for multiple comparisons. Mean +/- SEM shown.

Relative abundance of *Firmicutes* and *Clostridiales* in naïve vs. isoflurane-exposed mice

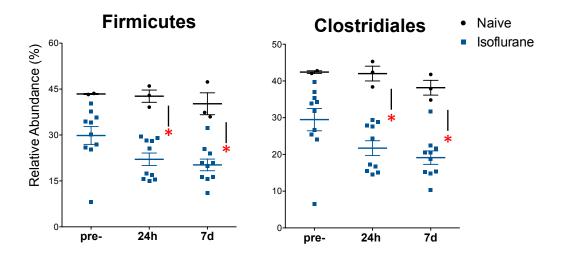


Figure S3. Two-way Anova for comparison of naïve and GA exposed mice demonstrates that isoflurane significantly affects relative abundance of *Firmicutes* and *Clostridiales* (p<. 001 for both). Post-hoc analysis with Bonferroni's correction for multiple comparisons demonstrates significantly reduced populations of both at 24h and 7d (adjusted p-values all <.005).) Mean +/- SEM shown.