

## Appendix 2 – additional statistical analysis

### *Parasite effects on beta diversity*

In addition, we explored the effects of parasites on beta diversity by calculating Bray-Curtis dissimilarity indices (Bray & Curtis, 1957), Yue & Clayton dissimilarity metrics (Yue & Clayton, 2005), Ochiai indices (Ochiai, 1957) and Jaccard distances (Jaccard, 1901) with software 'mothur' (Schloss et al., 2009). Bray-Curtis dissimilarity takes into account shared species proportions, Yue-Clayton distance uses species proportions from both the shared and non-shared species, Ochiai index is geometric mean of the ratio of joint occurrences of the species compared to their total occurrence, and Jaccard's index calculates the ratio of the number of shared species to the number of distinct species in two communities. We used all the factors to build models with the exception of week as it strongly correlated with presence of nematodes. We used both permutational manova with dissimilarity matrices taking into account the repeated sampling and analysis of multivariate homogeneity of group dispersions with the package 'vegan' (Oksanen et al., 2015). Permutational manova compares the effects of variables on the centroids of the groups. We used same strategy as with alpha diversity: we included all previously mentioned variables in our models and subsequently removed sequencing and amplification batch variables to simplify the models. We used analysis of group dispersions to test whether absence or presence for parasites correlates with differences in microbiota composition variation and we compared individually all our parasite species. To take into account repeated sampling with latter analysis, we used one sample per individual for analysis and iterated this 100 times.

The results differed depending on the metrics. For permutational manova, there was significant effects in Jaccard index for *Hymenolepis diminuta* presence ( $R^2 = 0.015, p = 0.010$ ) (Figure 2a), in Bray-Curtis for sequencing batch ( $R^2 = 0.034, p = 0.019$ ), ectoparasites presence ( $R^2 = 0.020, p = 0.03$ ) and *H. diminuta* presence ( $R^2 = 0.019, p = 0.025$ ) (Figure 2c) and in Yue-Clayton for ectoparasites presence ( $R^2 = 0.020, p = 0.019$ ) (Figure 2b), while Ochiai

index did not show any significant variables. None of the group dispersions depending on the parasite presence were significantly different ( $p > 0.05$ ) in over half of the iterations.

We also calculated the dissimilarity between subsequent samples from the same individual and compared with linear mixed models if change in parasite status from absent to present or other way round correlated with different level of dissimilarity. There were no significant differences in dissimilarities of microbiota communities due to gain or loss of parasites.

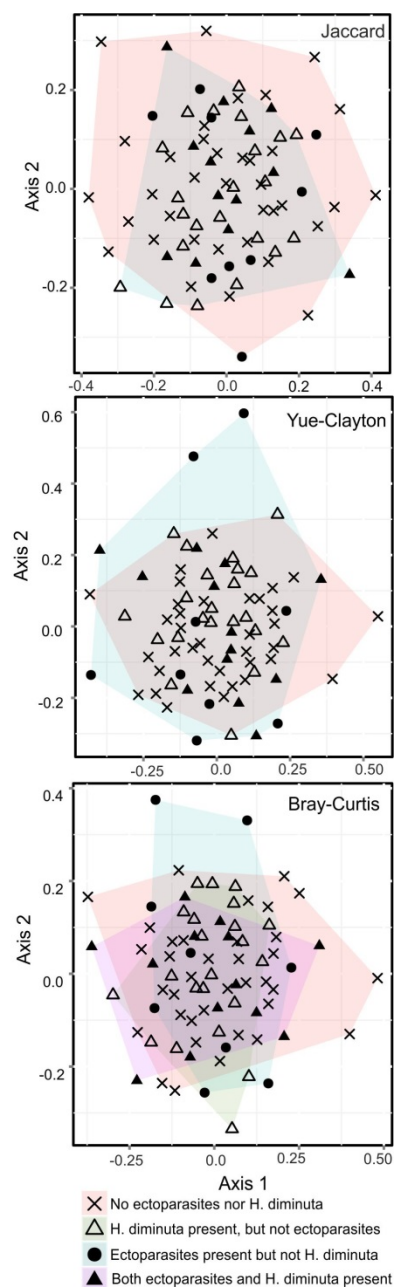


Figure A1: Non-metric multidimensional scaling (NMDS) in two dimensions for different indices. The graphs show : a) Jaccard index with shaded areas showing presences or absence of *H. diminuta*, b) Yue-Clayton index with shaded area showing presence or absence of ectoparasites for Yue-Clayton and c) Bray-Curtis index showing all four different presence-absence patterns for *H. diminuta* and ectoparasites. In all graphs, the samples with ectoparasites are denoted with filled points and samples with cestodes are marked with triangles. Samples with neither parasite are denoted by cross.

## References

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