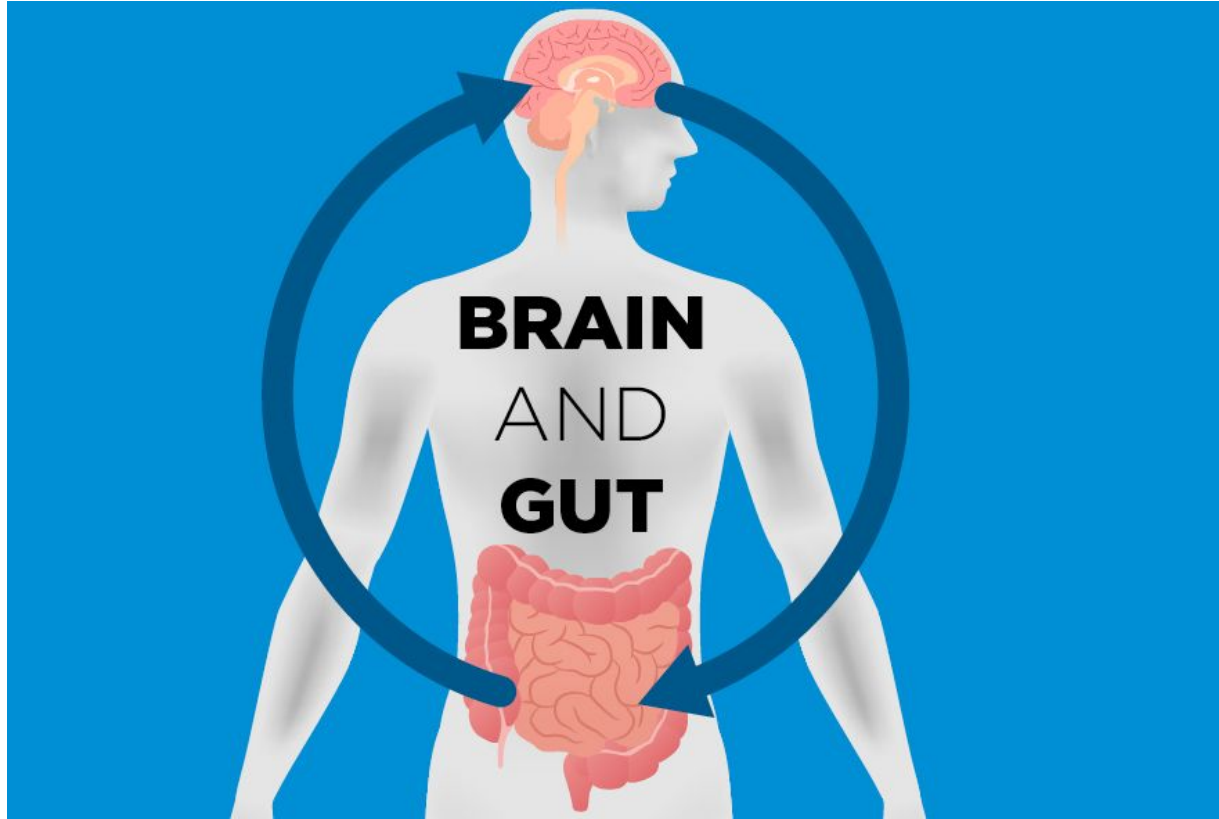


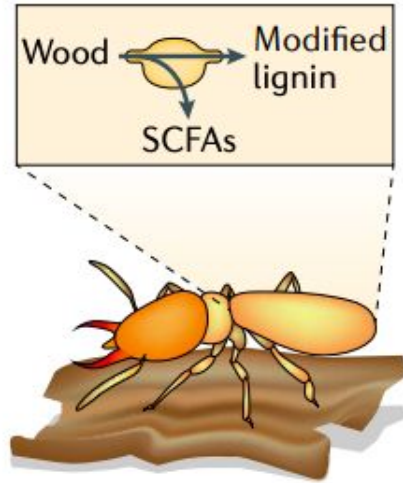
HoloFood sampling and experimental design and Q&A



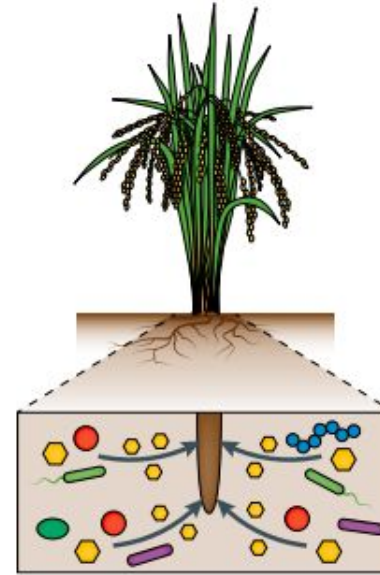
About perspectives



The host perspective



Termites and wood decay
Lignocellulose degradation

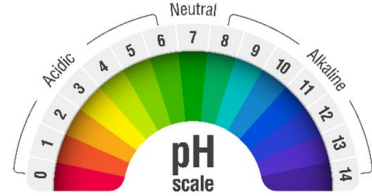
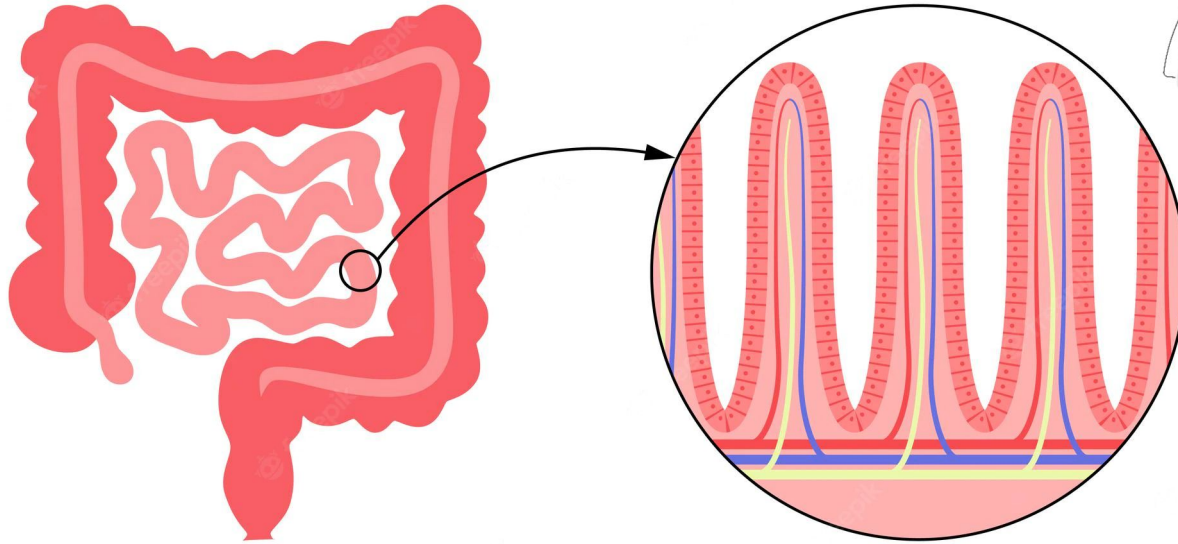


**Crop plants and
root microbiome**
Nutrient provision

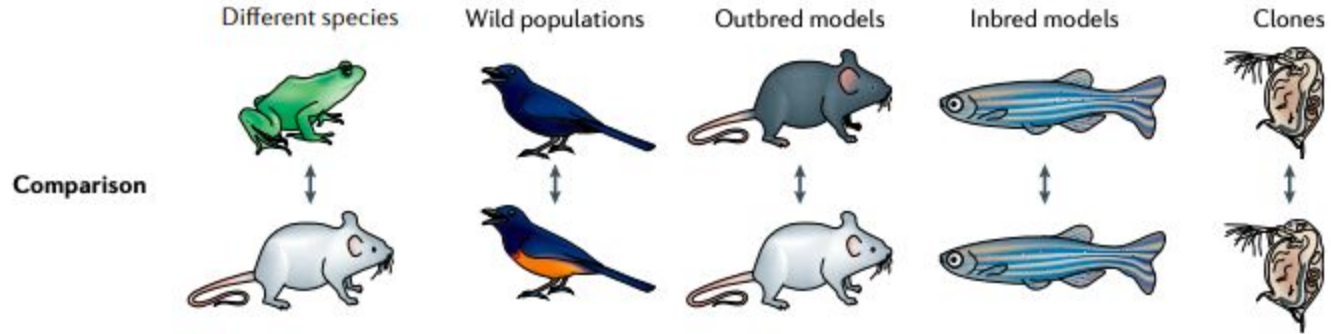


The microbiota perspective

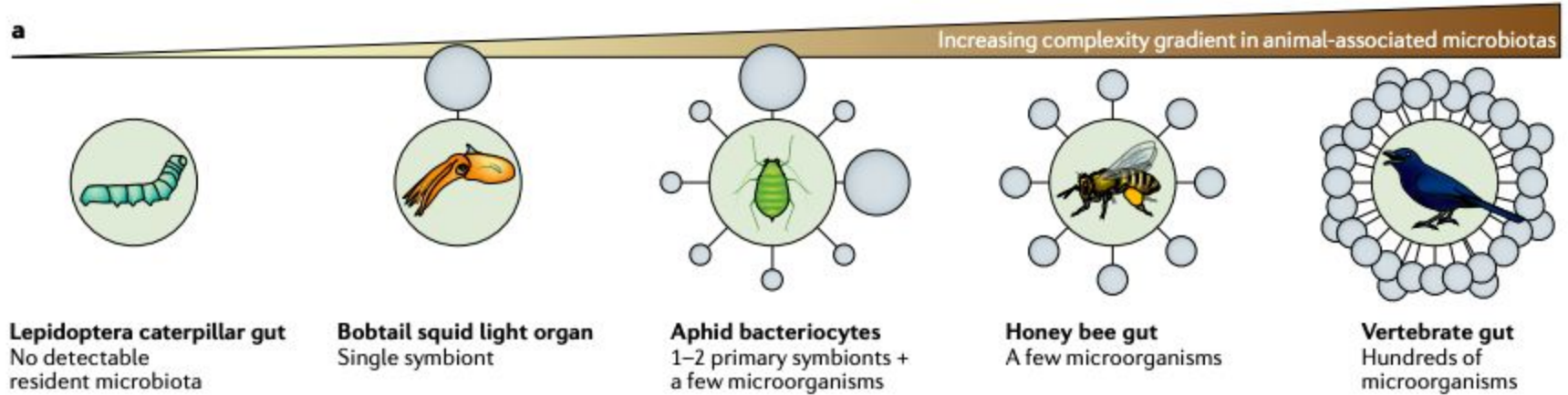
INTESTINAL VILLI



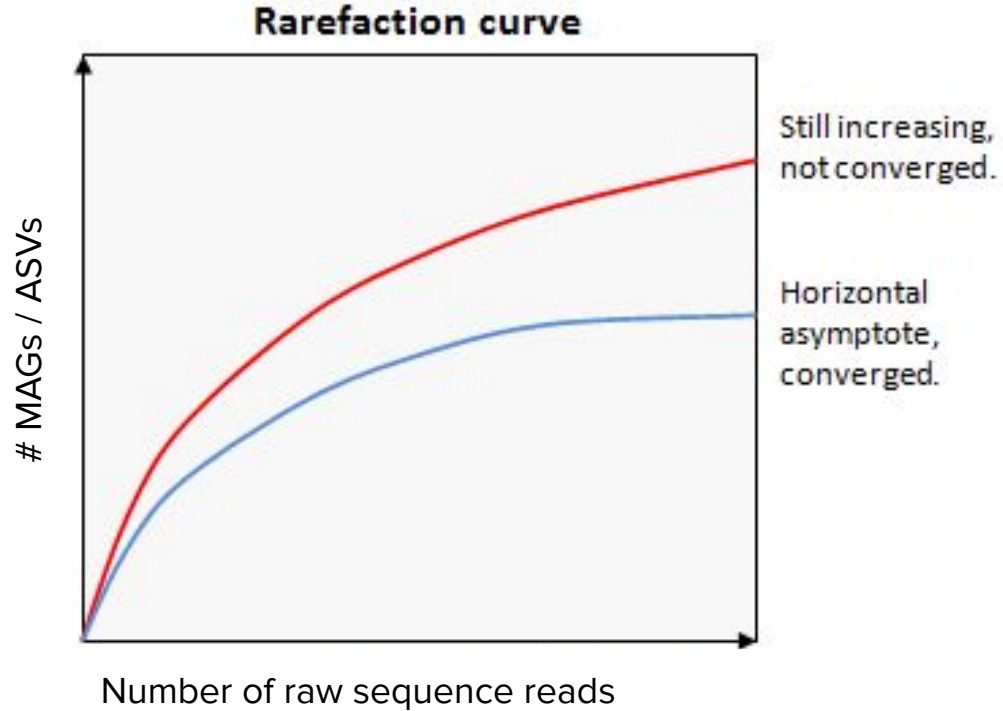
About complexity



Different levels of complexity in real world



Rarefaction curve



Rarefaction curve

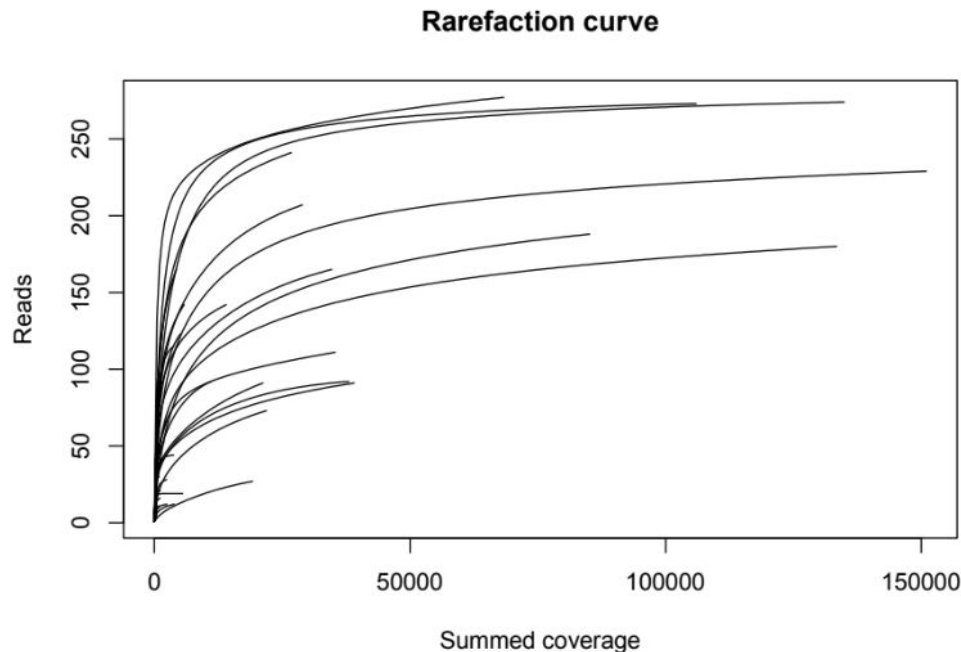
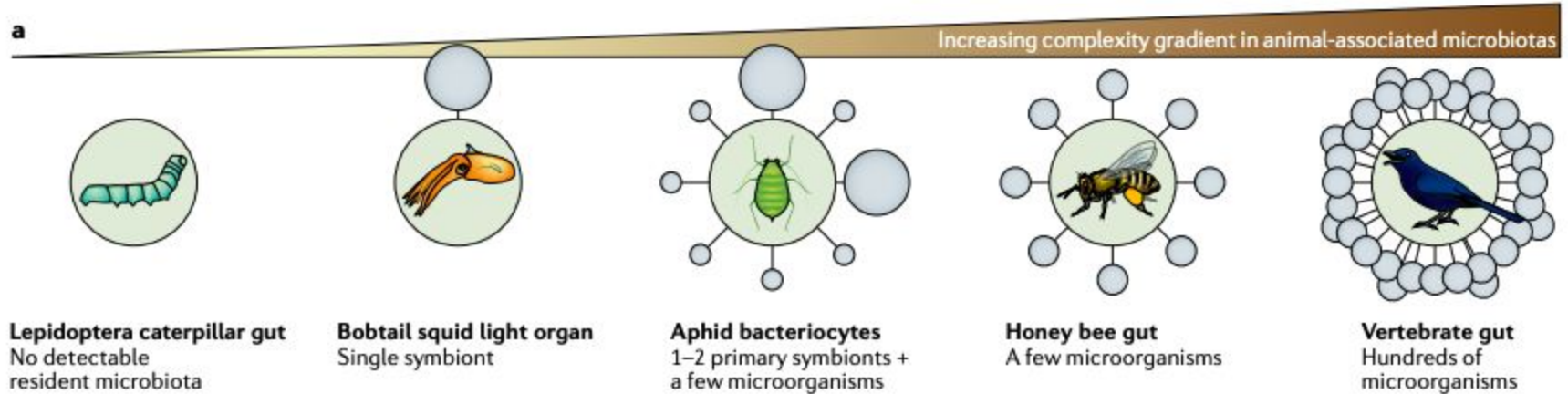


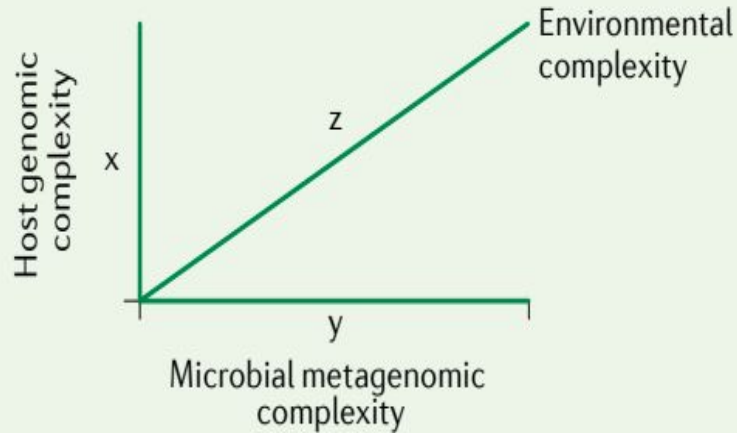
Figure S1. Rarefaction curve showing the sequencing saturation of the samples. Only a few samples reached saturation.

Where do you need the deepest sequencing?

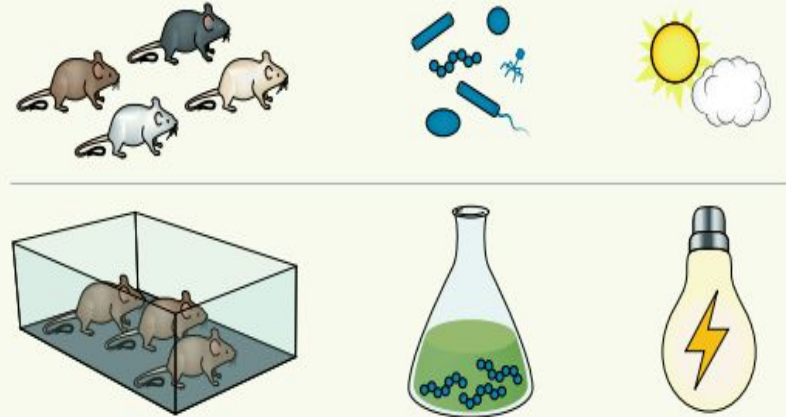


Assessing the complexity of your study system

① What is the complexity of the study system?

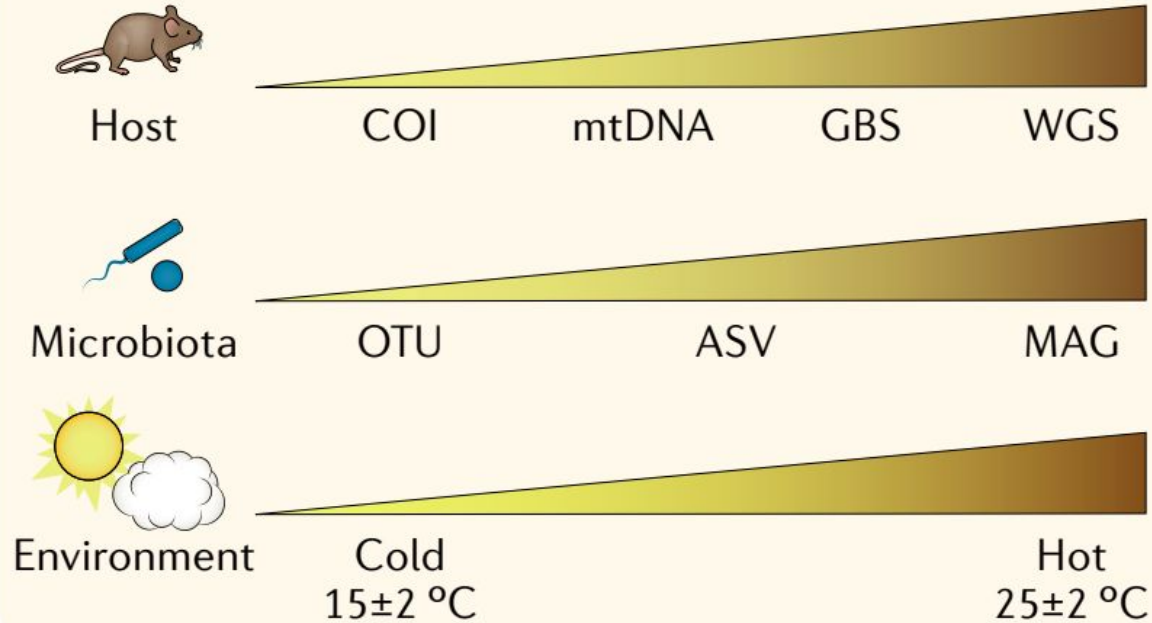


② Which variables can be controlled by the researcher?

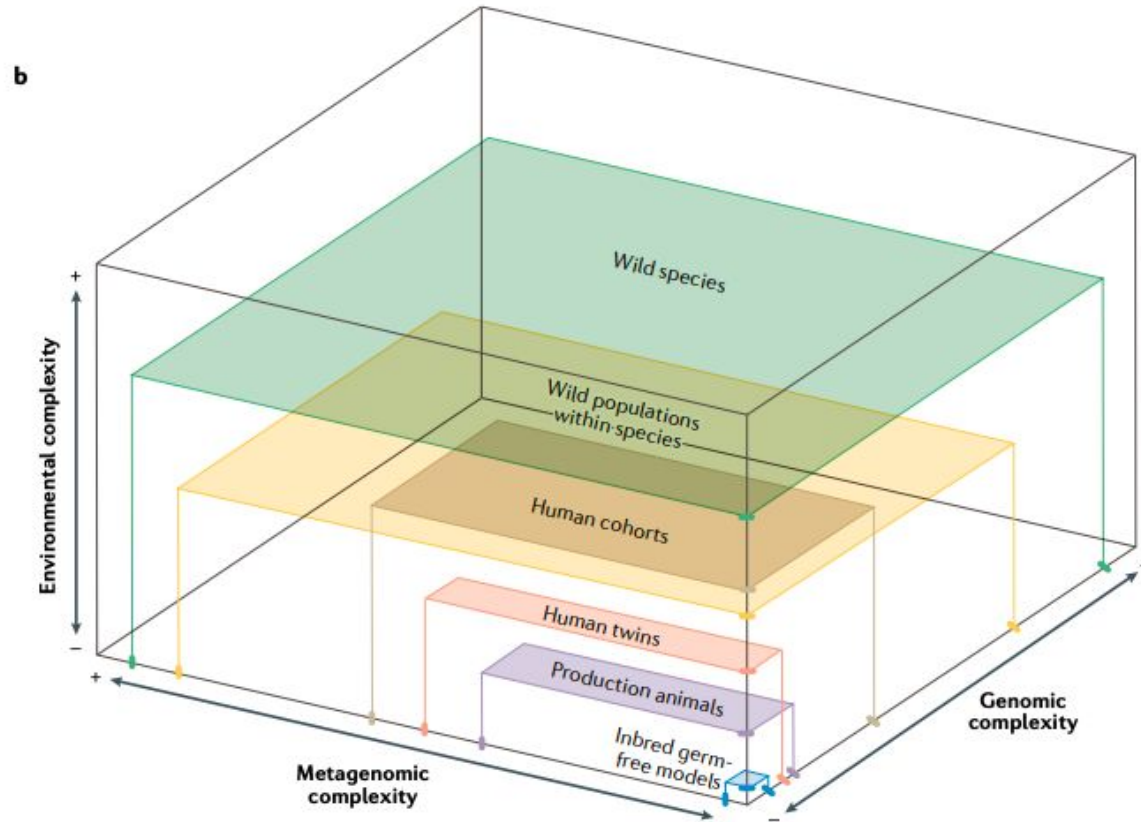


Can you control this complexity?

③ What is the genomic resolution at which hologenomic variation is studied?



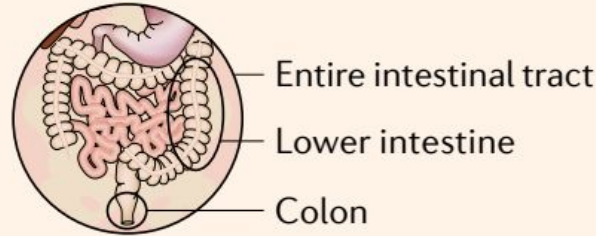
Study systems and varying complexity



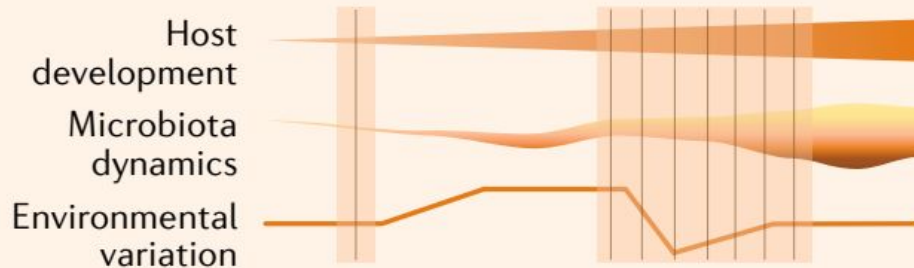
About space and time and complexity

④ What are the spatiotemporal features of the studied hologenomic variation?

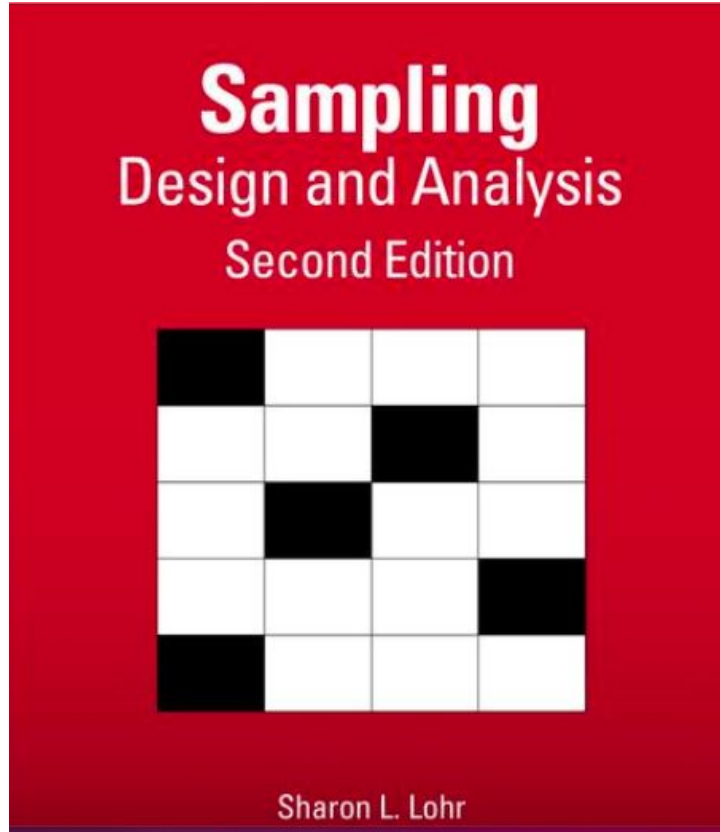
Spatial features



Temporal features: when, how often and for how long

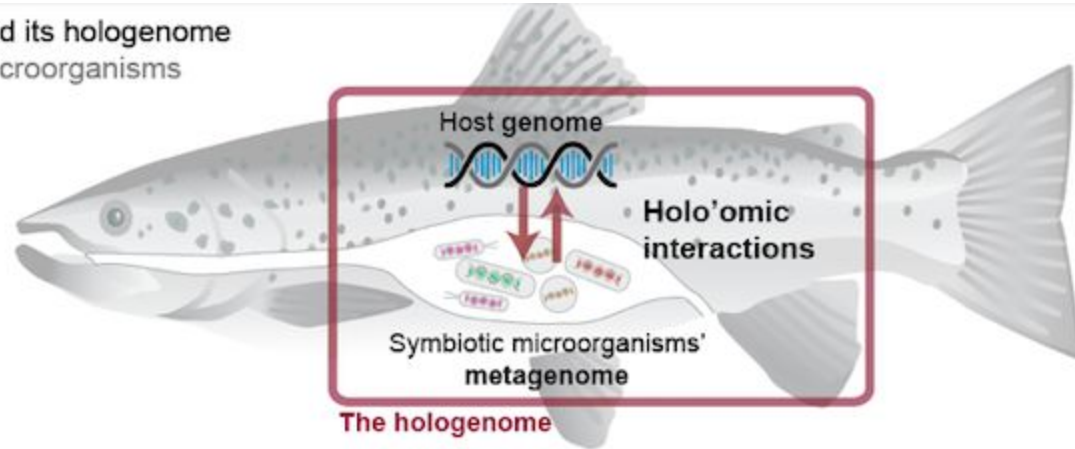


About sampling design



There are many holo-omics layers

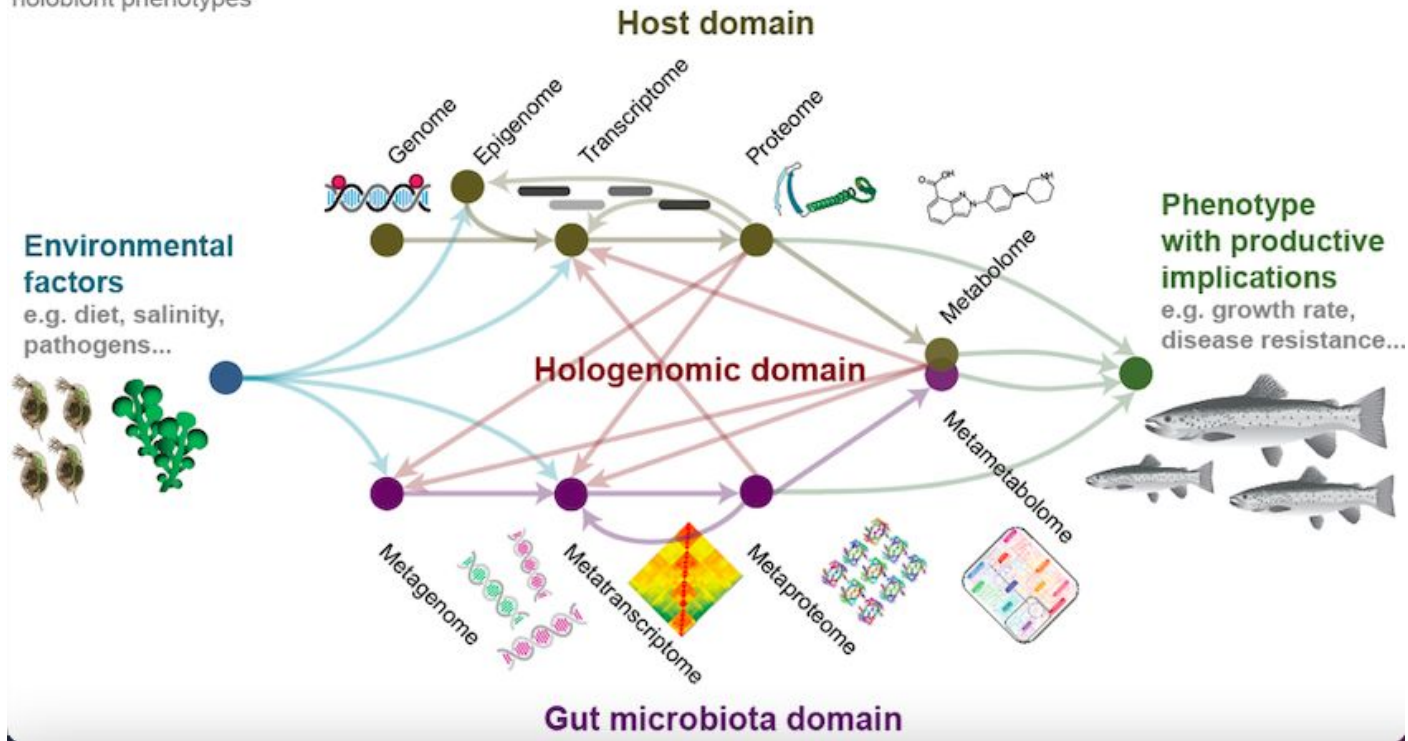
A) The holobiont and its hologenome
Host + symbiotic microorganisms



There are many holo-omics layers

B) Holo'omic interactions

Biomolecular interactions between hosts and symbiotic microorganisms triggered by environmental factors yield different holobiont phenotypes

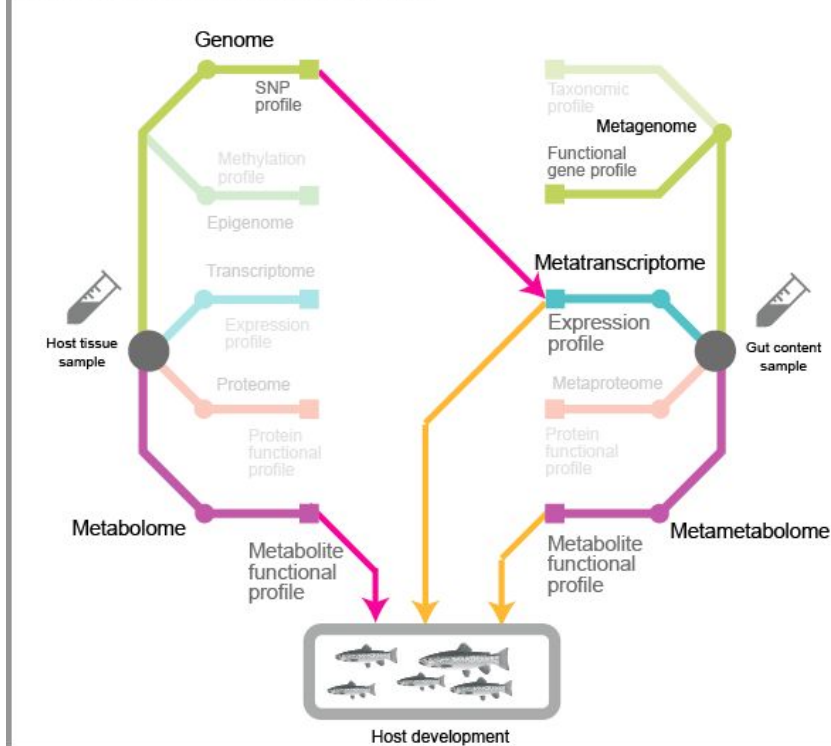


Shall we generate data for all layers?

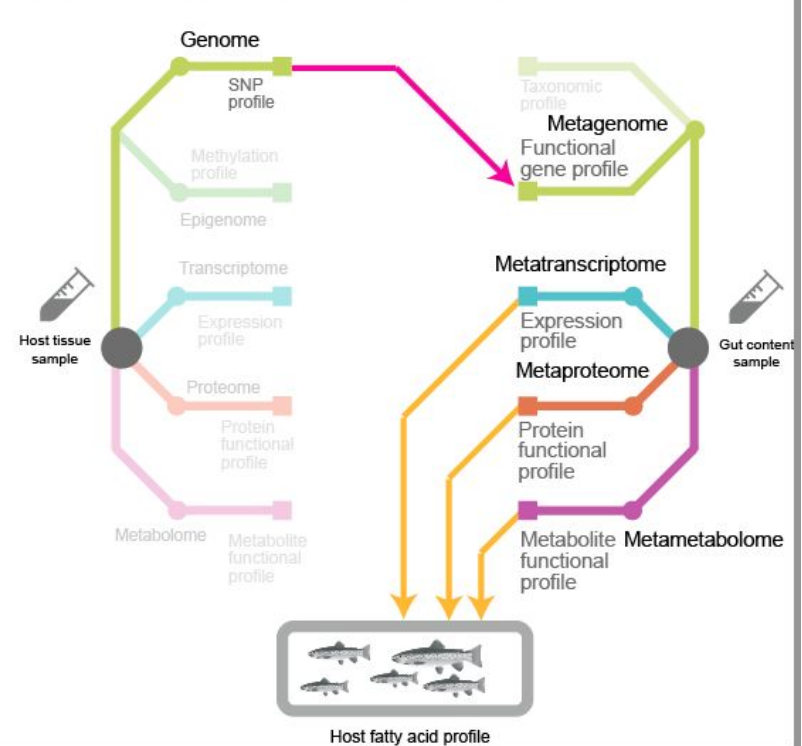


Shall we generate data for all layers?

A) Example 1. Replacing fish meal



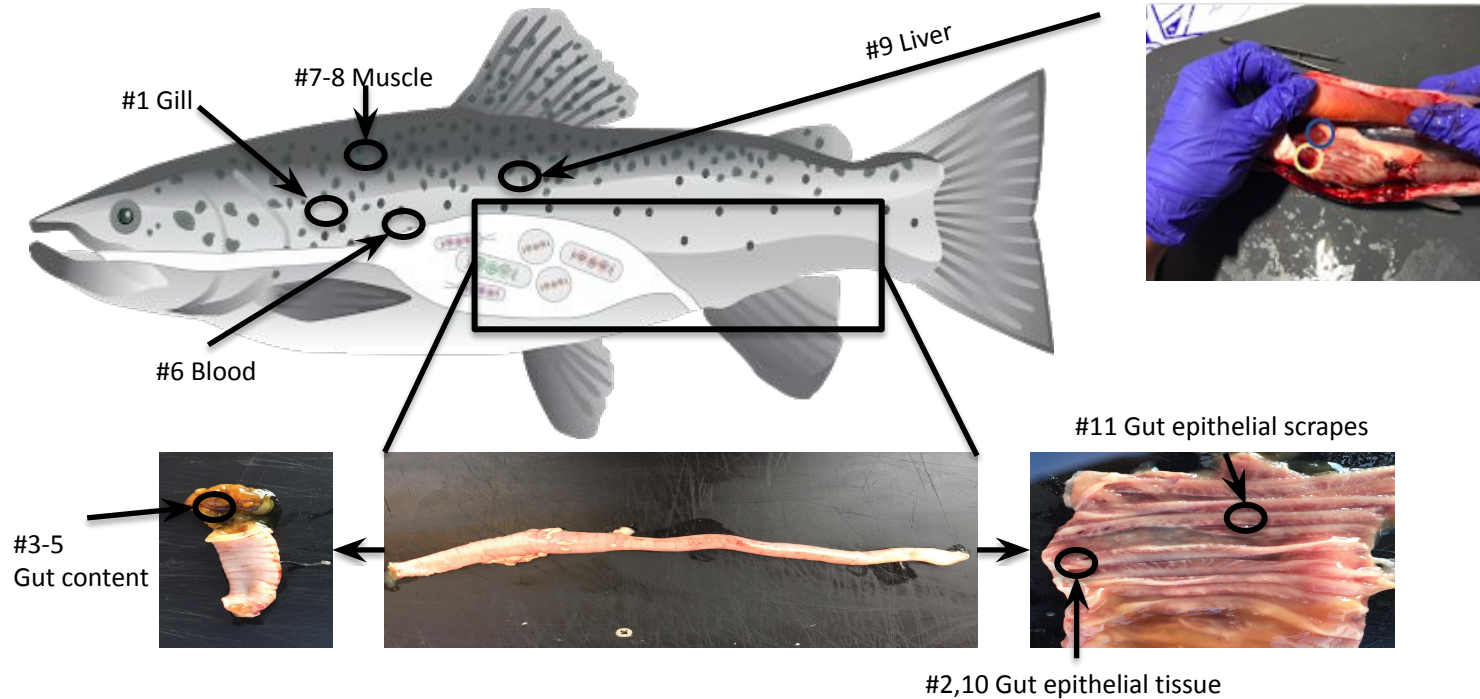
B) Example 2. Controlling fatty acid composition



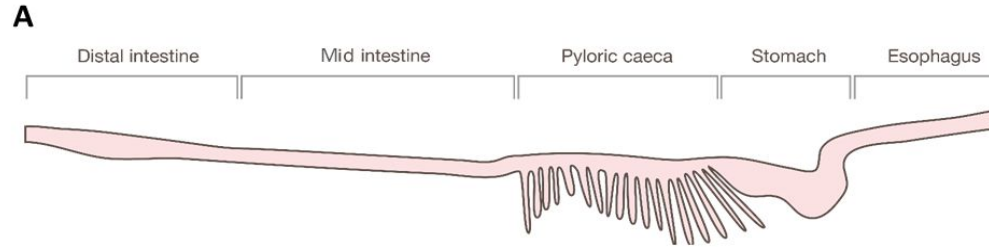
About sampling protocols



Multiple potential sites to sample



Gut content vs gut mucosa sampling



About Preservation methods

Preservation methods





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A comparison of storage methods for gut microbiome studies in teleosts: Insights from rainbow trout (*Oncorhynchus mykiss*)

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National History Museum of Denmark, University of Copenhagen, DK-1350 Copenhagen, Denmark

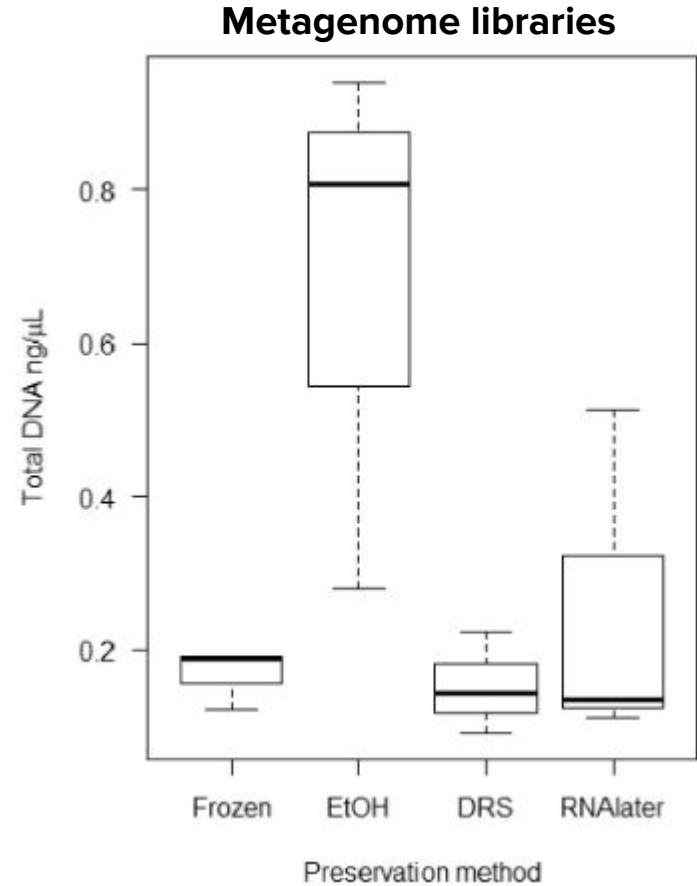
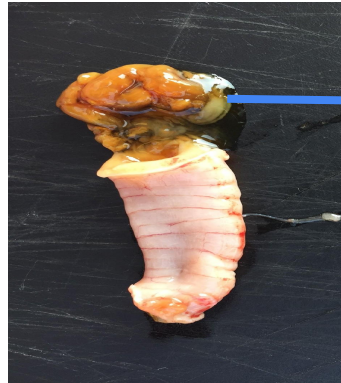




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Ok - that's enough background



Group exercise - take 2



1. Re-visit exercise from lecture 1
2. Now design a sampling protocol for the hologenomic study you proposed in exercise 1
3. Consider for example:
 - What data do you want?
 - Types of samples from host
 - Types of samples from microbiota
 - Tissues?
 - Buffer or no buffer?
 - *More steps of the protocol to obtain your samples ?*
4. Speed presentations by all + discussions

HoloFood sampling and experimental design and Q&A

The END

