Both age and genetic predisposition to mastitis influence the digestive microbiota in ewes

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The microbiome is known to influence the immune system (Thais *et al.,* 2016). While age-related dysbiosis and inflammation have been linked in humans, there is a paucity of research in this area in livestock (Bosco & Noti, 2021). The objective of this study was to investigate the digestive tract microbiota of dairy ewes with varying ages and genetic profiles.

Does age and genetic predisposition to mastitis influence the digestive microbiota?

Materials and methods

	Resistant	Susceptible
Old	10 ewes	10 ewes
Young	10 ewes	10 ewes

A total of forty Lacaune dairy ewes from two divergent genetic lines selected for mammary inflammatory response based on milk somatic cell count (Rupp *et al.*, 2009) were equally divided into "**Old**" (≥ 5 lactations) and "**Young**" (first lactation) categories.

Ewes were managed as a single group for several months at the end of the lactation. After euthanasia, digestive content was sampled in rumen, ileum, caecum, and feces.

The bacterial DNA was sequenced on the hypervariable regions V3-V4 of the 16S rRNA with Illumina MiSeq. FROGS pipeline (Escudié *et al.*, 2018), *mixOmics* package (Lê Cao *et al.*, 2009), and *Metacoder* (Foster *et al.*, 2017) were used for bioinformatics and statistical analysis.
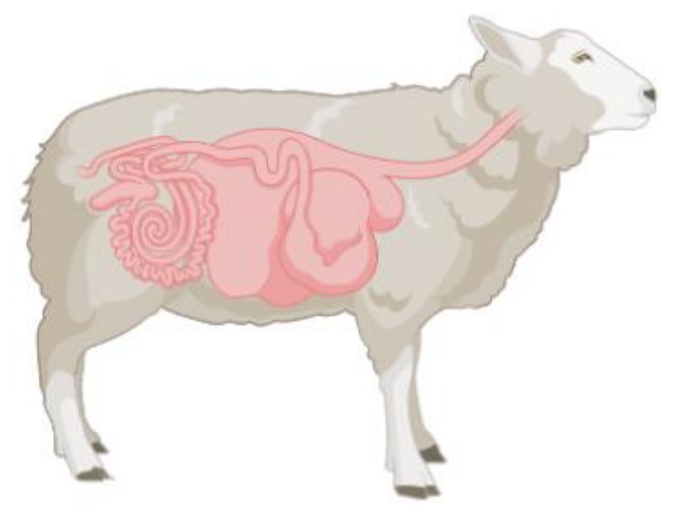
Results

A distinction between "Young" and "Old" ewes can be made based on the microbiota composition throughout the 4 compartments of the digestive tract studied using an PLS-DA (**Figure 1**). The "Young" group can be distinguished according to the genetic background for mastitis susceptibility for the ruminal microbiota, while the "Old" subgroup does not allow for this discrimination. In the caecum, the genetic lines cannot be distinguished in the "Young" group, whereas resistant and susceptible lines are identifiable in the "Old" group.

The PLS-DA based on fecal bacterial microbiota can identify four distinct groups. These groups correspond to the two ages combined with the two divergent lines.
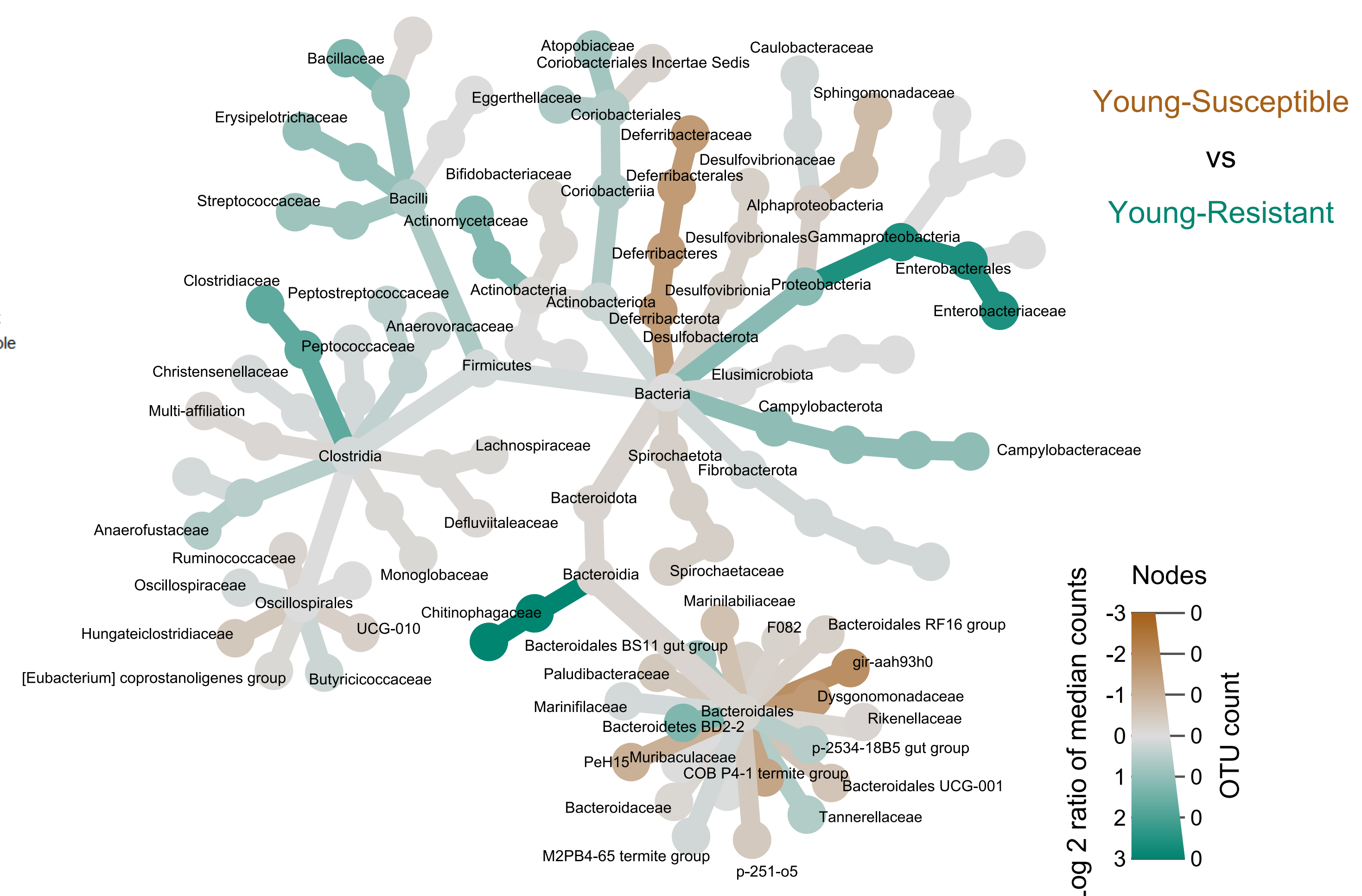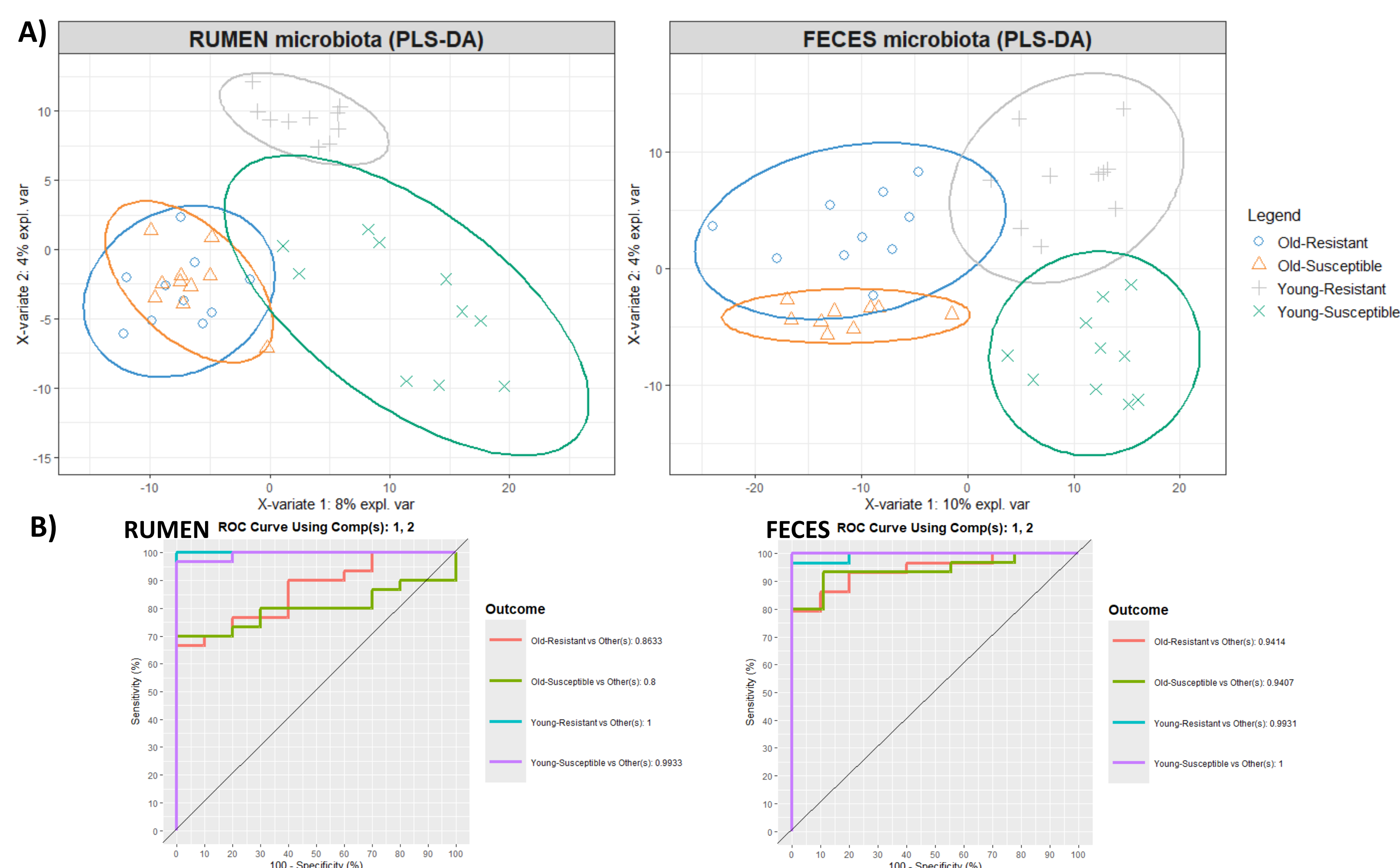

Figure 1: Bacterial microbiota of ewes, grouped according to their age and genetic line (resistant or susceptible to mastitis), represented via PLS-DA (A) and the subsequent Area Under the Curve (AUC) and Receiver Operating Characteristic (ROC) curves.

Figure 2: *Metacoder* representation of the taxonomic composition of the fecal microbiota of ewes, according to age and genetic line ("Young-resistant" or "Young-susceptible" to mastitis).
Nodes and branches are colored by the Log 2 ratio of median OTU counts: if brown, higher in "Young-susceptible" compared to "Young-resistant" and inverse if green.

Conclusion

Our study indicates that both age- and genetic background for disease susceptibility influences the microbiota composition. **Fecal samples** might be more appropriate to identify changes in the microbiota that are associated with **increased susceptibility to infection and inflammatory response**, compared to the rumen.

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Foster ZSL, Sharpton TJ, Grünwald NJ (2017) Metacoder: An R package for visualization and manipulation of community taxonomic diversity data. PLOS Computational Biology 13(2): e1005404

Lê Cao, K-A., González I. and Déjean S. (2009) integrOmics: an R package to unravel relationships between two omics data sets. Bioinformatics. 25(21):2855-2856 NOTE: the package 'integrOmics' has been renamed to 'mixOmics'

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