Dynamics of rumen bacterial guilds in lambs from birth to finish are affected by the absence of dams in the suckling period

VOLAND Laurianne, DOU Paul , TOURNAYRE Jeremy , DEHORNOY Aurélie , GRAVIOU Dominique , VAZEILLE Karine, MORGAVI Diego P., POPOVA Milka

INRAE

HoloRuminant
Understanding microbiomes of the ruminant holobiont

uca
ÉCOLE DOCTORALE
DES SCIENCES DE LA VIE,
SANTÉ, AGRONOMIE
ENVIRONNEMENT

UMR1213 Herbivores
VetAgro Sup

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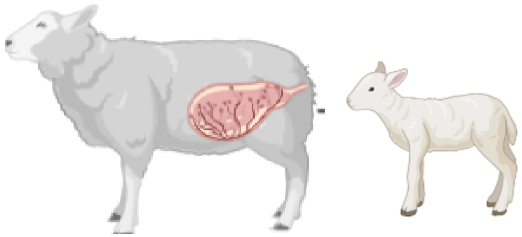
Dairy farming

MOTHER Rearing (MR)



↗ **Health**
↗ **Welfare**
↘ **Economy**

Reasons :

Young orphans

Mother does not produce enough milk

↘ risk of disease transmission

↗ cost of milk

ARTIFICIAL Rearing (AR)

+ Common in Europe

↘ **Health**
↘ **Welfare**
↗ **Economy**

Problems :

↘ Effects on growth

↗ Mortality problems

↘ Behavioral reactions

↘ Mother-young relationship

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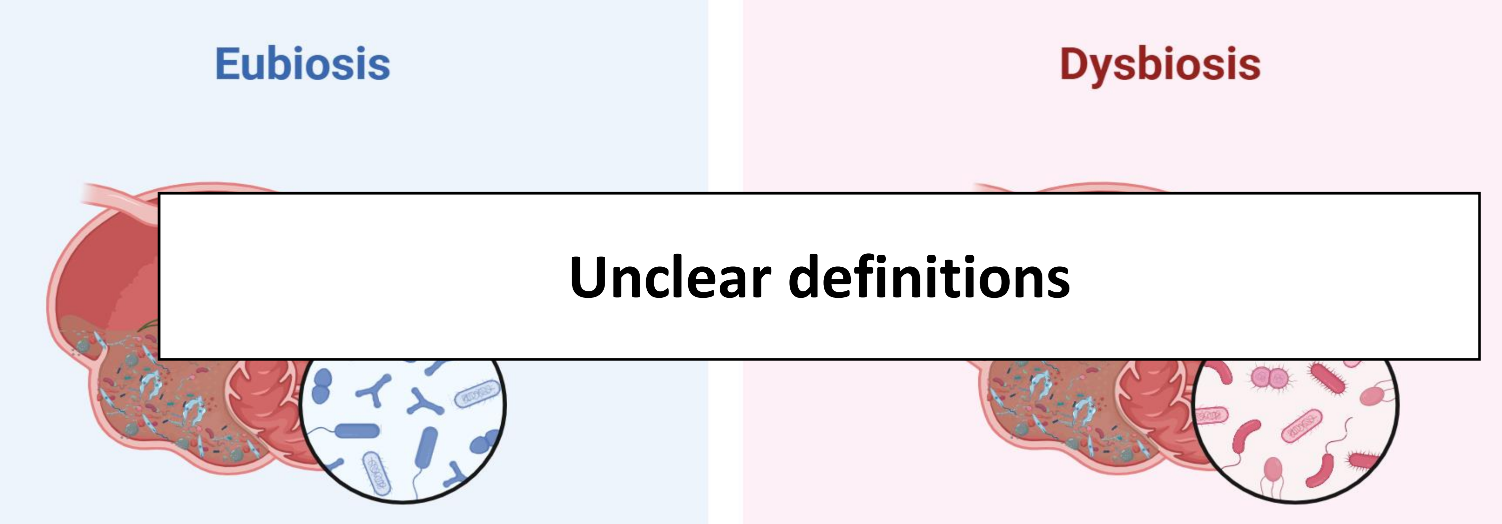
Rumen microbiota

Rumen microbial colonization

- Installation of bacteria, archaea, protozoa and fungi
- Essential for plant fiber digestion and physiological development (volatile fatty acids) : Energy source for ruminants (70%)

Microbiota : Two faces

(Di Viencezo et al., 2024; Lloyd-Price et al., 2016; Iebba et al., 2016)

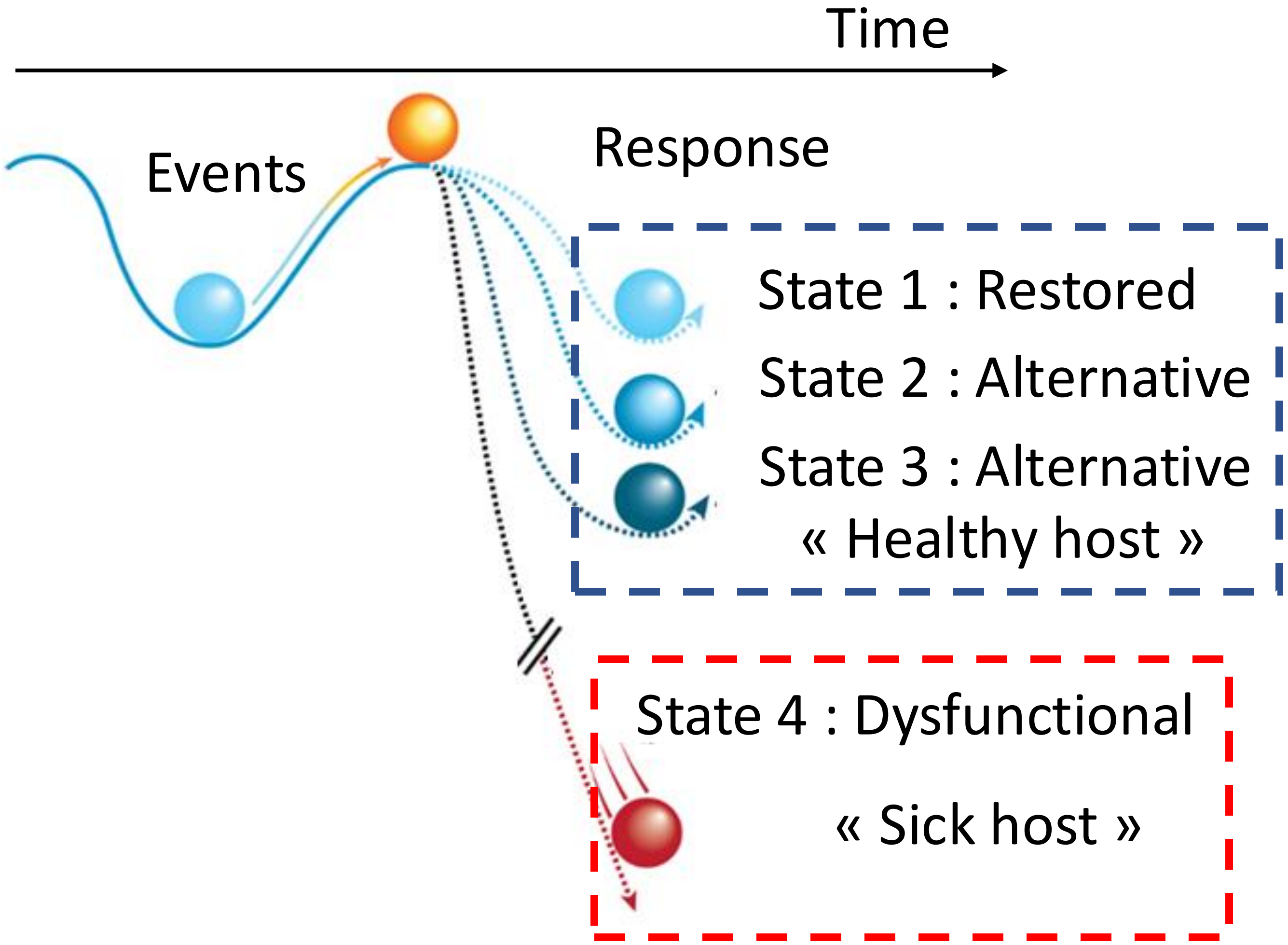


Unclear definitions

↗ Taxa diversity / microbial genes
Stable functionnal core microbiota
Tolerate invasion of pathogens
Mutualistic relationship between the microbiota and the host

Bacteria no longer life in mutual harmony
↘ Taxa diversity / microbial genes
↘ tolerate invasion of pathogens
Link to diseases

Colonization of the rumen microbiota in a healthy state



Time

Events

Response

State 1 : Restored

State 2 : Alternative

State 3 : Alternative

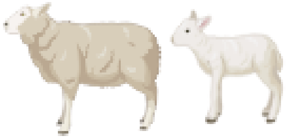
« Healthy host »

State 4 : Dysfunctional

« Sick host »

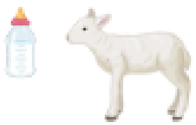
Fig : Evolution of microbiota adapted from Bornbusch et al. (2023)

Aim : Understand the colonization of the rumen microbiota in a healthy state



MR: Maternal rearing

Presence of mother → Favors colonization of key microbes



AR : Artificial rearing

Absence of the mother → Reduction of microbes or emergence of other dominant species

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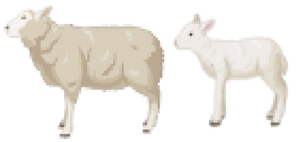
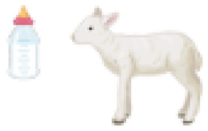
Animal study design



N= 19

MR: Maternal rearing



N= 13

AR : Artificial rearing

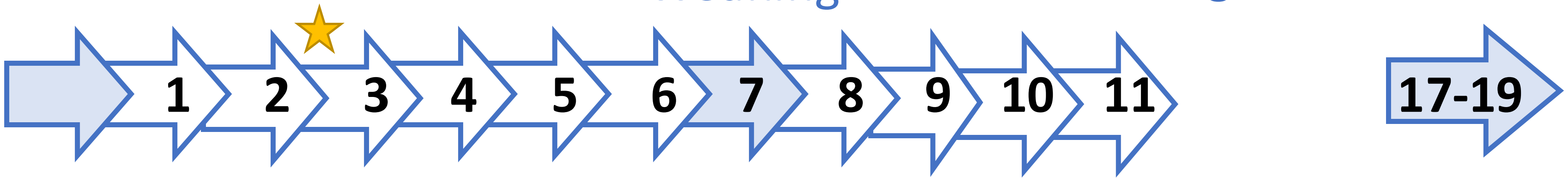
N = 13	N = 6	2022
N = 6	N = 7	2023

★ Hay and concentrate

Birth

Weaning

Slaughterhouse



MAY **JUNE** **JULY** **AUG.** **SEPT.**

Each week : Rumen fluid : Sequencing (**Bacteria**, Archaea, Fungi and protozoa)

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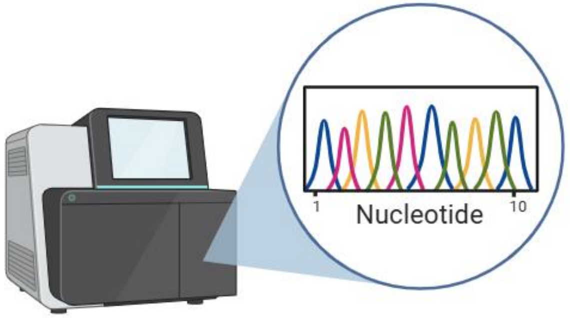
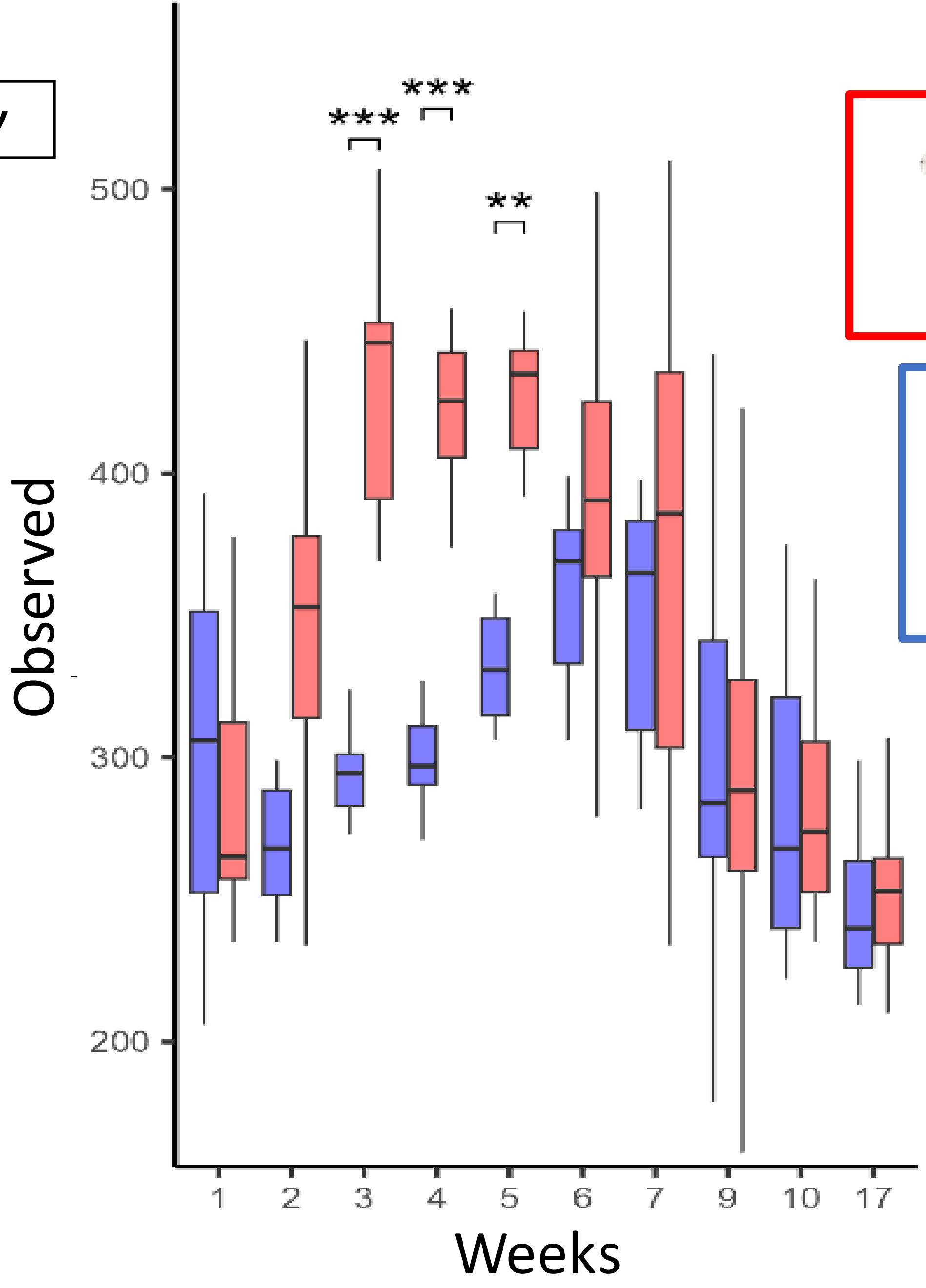
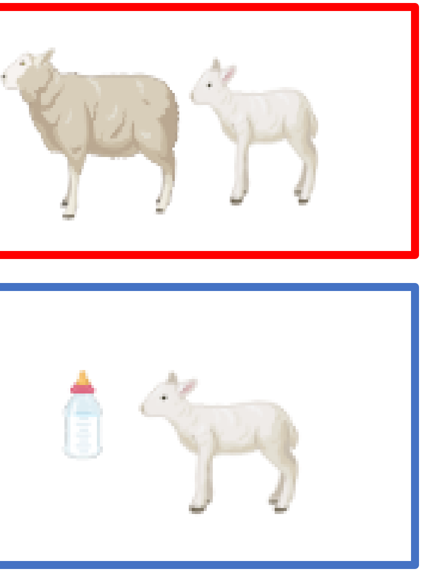
Bioinformatics analysis

Alpha diversity



Amplicon sequencing
515F - 806R amorce
Qiime 2
Silva





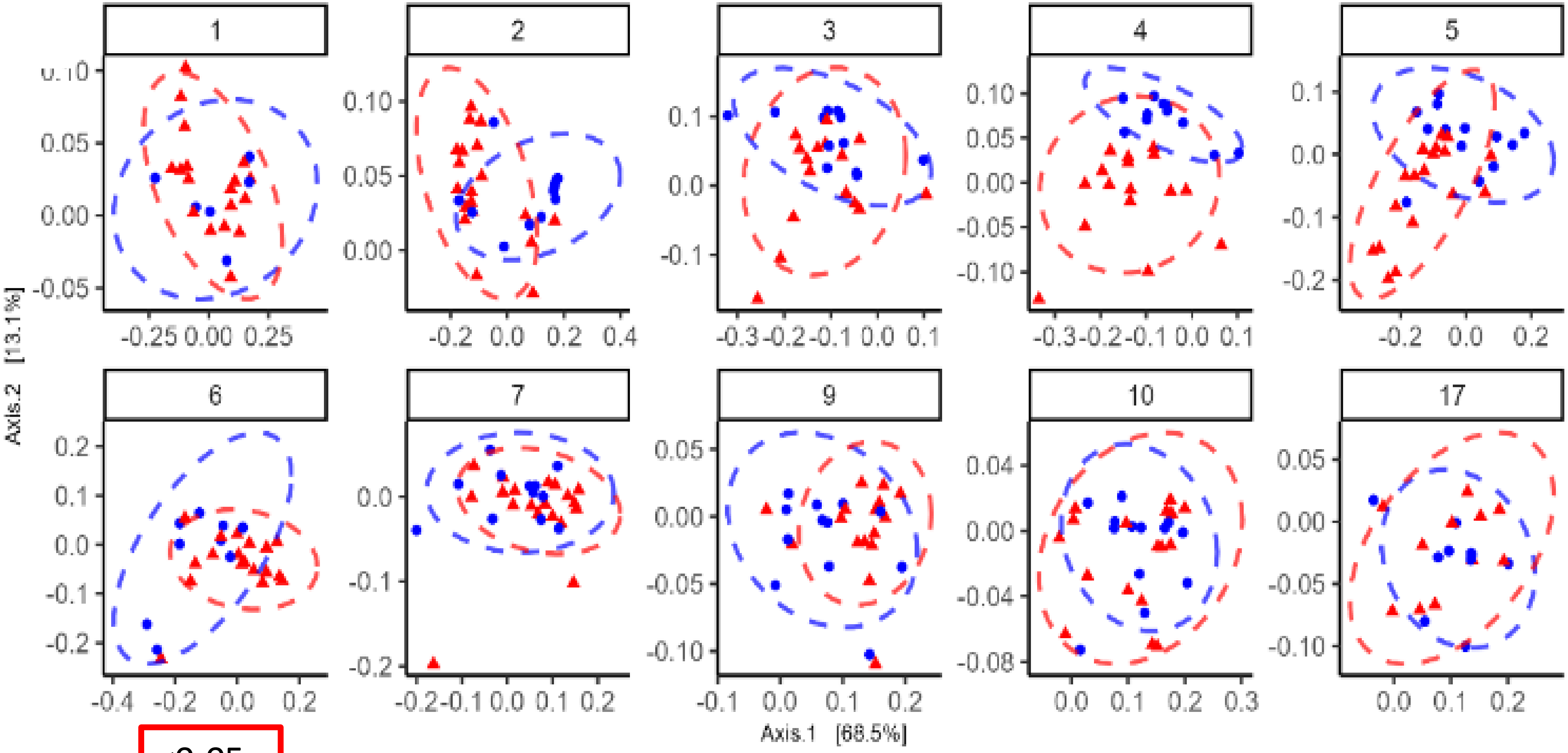
Anova test :
Time p = <0,01
Groups : p = <0,01
Interaction : p = <0,05

This project has received funding from the Eu

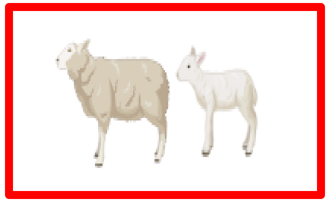
Bioinformatics analysis

Beta diversity

<0,05 <0,05 <0,05 <0,05



<0,05



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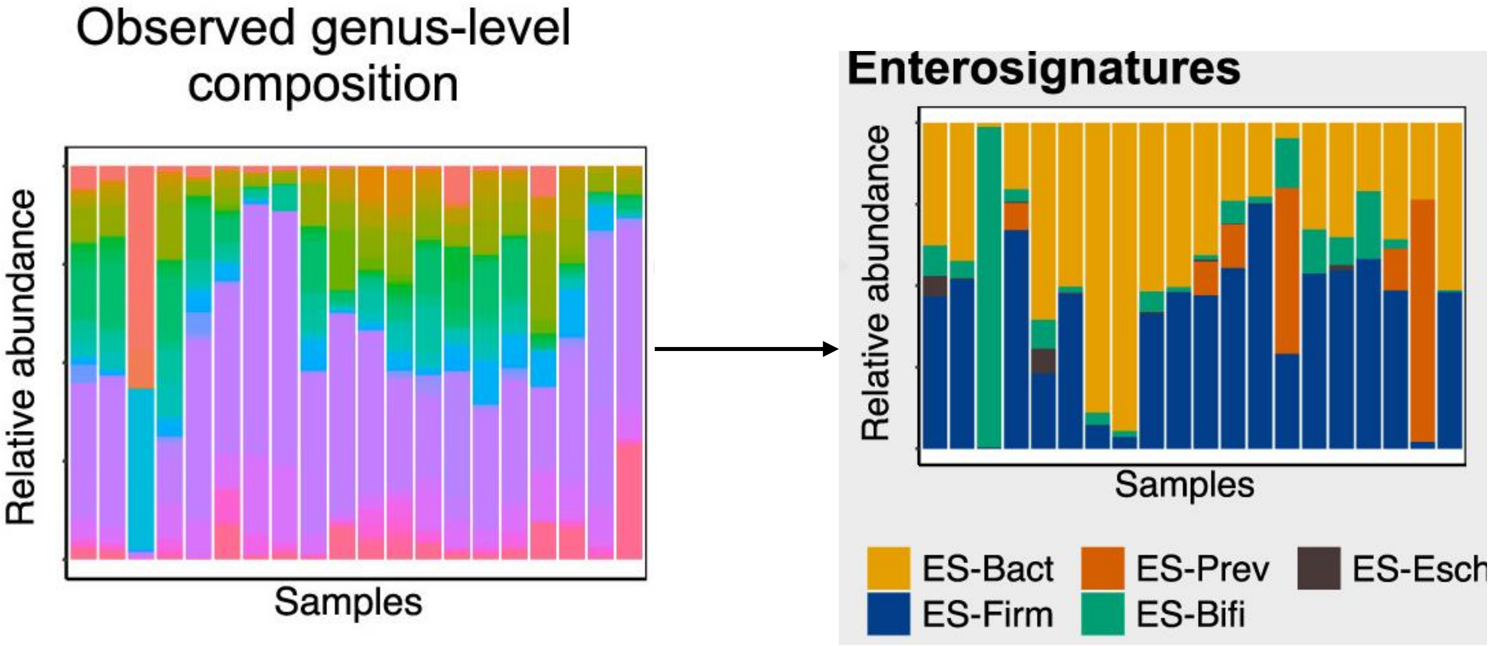
Towards Enterosignatures (Microbial guilds)

Alpha diversity :
↗ observed in MR group until weaning

Beta diversity :
Difference until weaning

How about dynamics and interaction of microbes ?



(Frioux et al., 2023)

RS to describe rumen bacteria

What combination of ASVs contributes to each RS?
(=Score of each ASV within each RS)

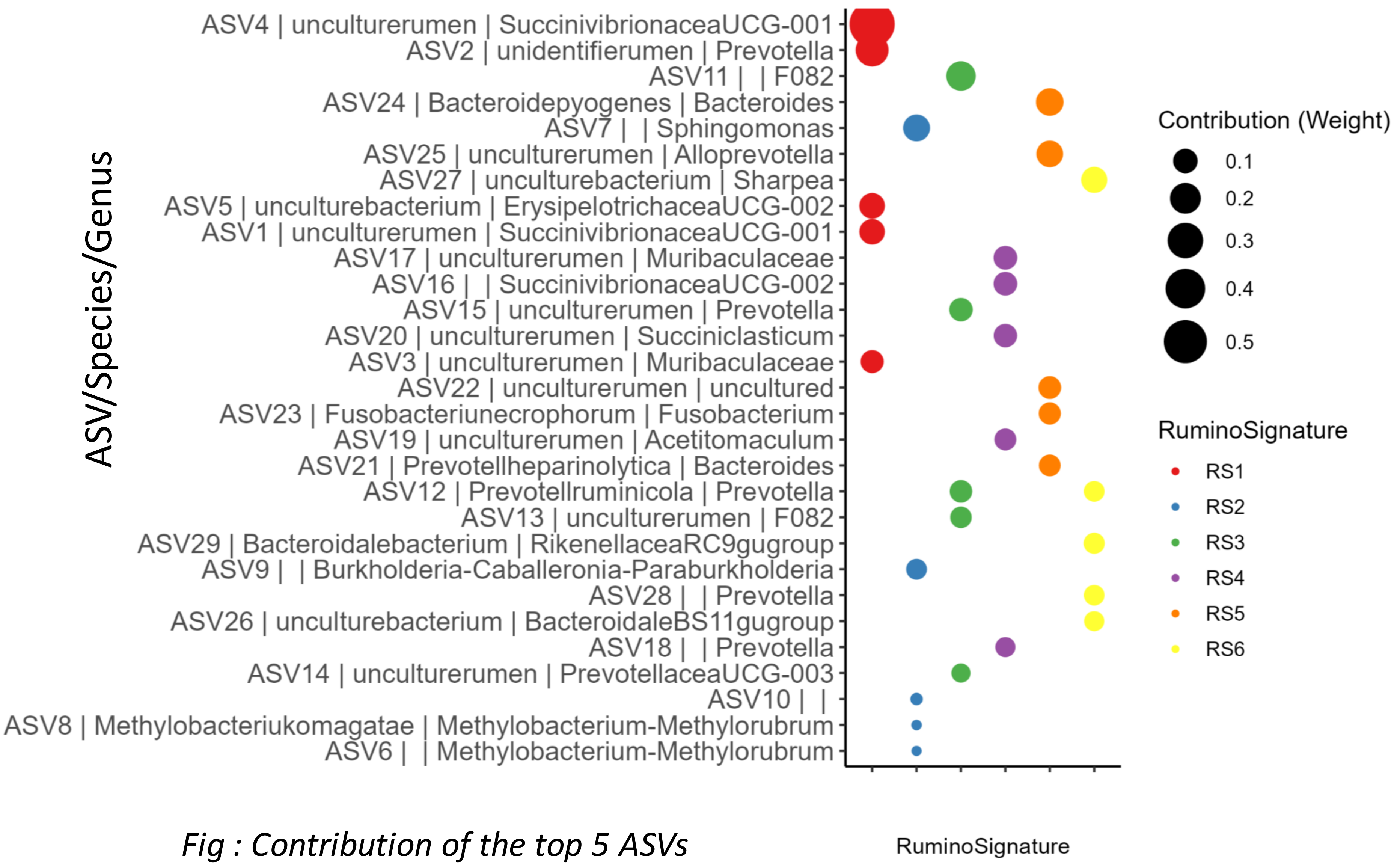


Fig : Contribution of the top 5 ASVs

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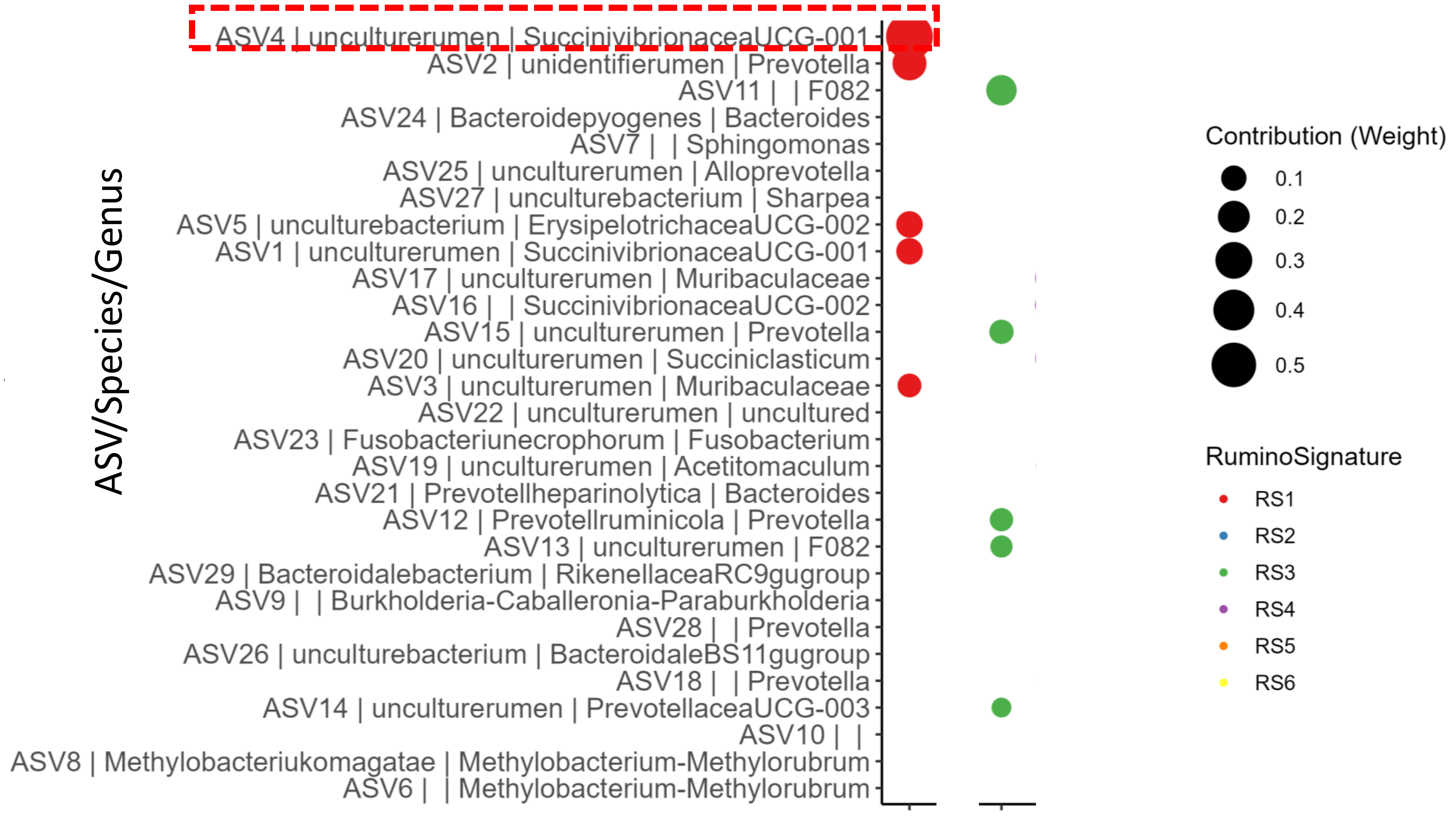

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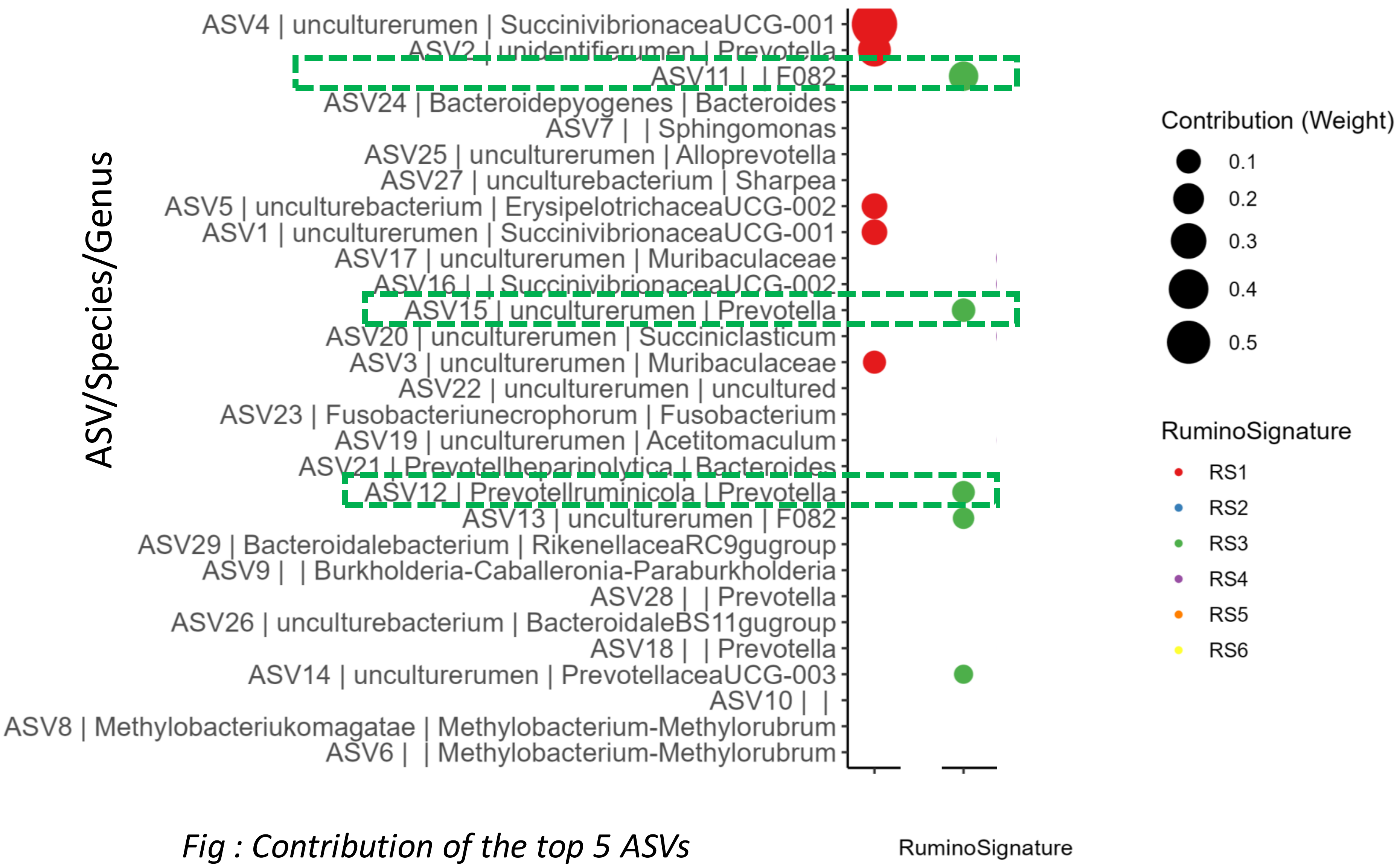

Fig : Contribution of the top 5 ASVs

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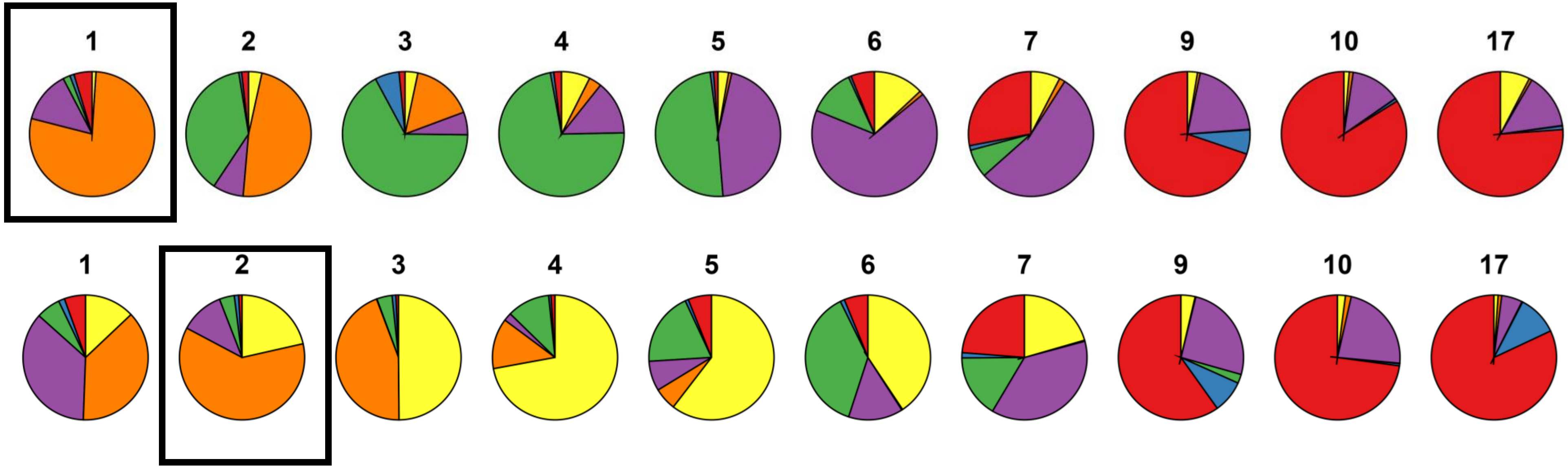
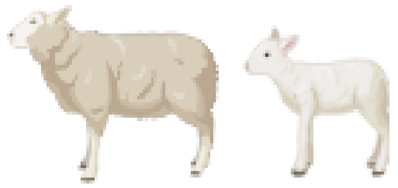
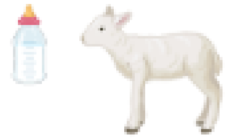
Establishment of RS in lambs

How is each sample influenced by each RS? (=Score of each SR in a sample)







● **RS5** : *Bacteroides, Alloprevotella, Neisseriaceae*

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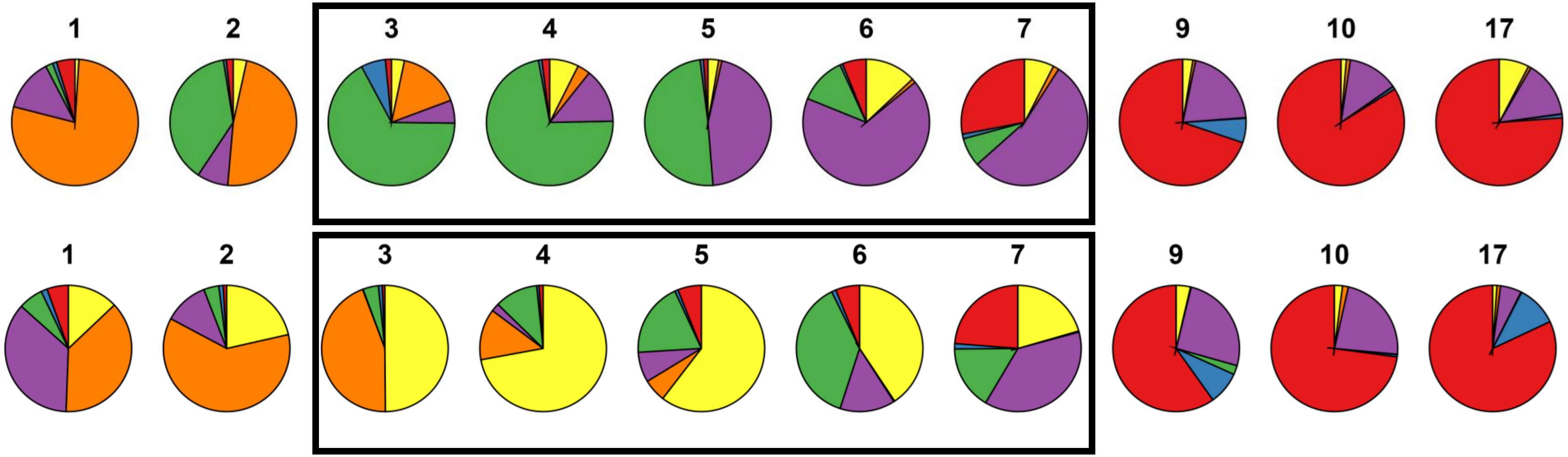

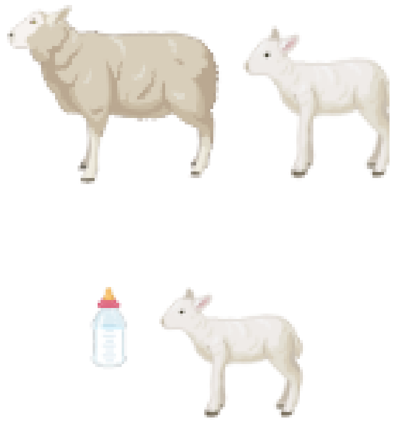

RS4 : *Muribaculaceae, Succinivibrionaceae UCG-002, Succiniticlasticum*

- **RS6** : *Sharpea, Prevotella, Rikenellaceae RC9 gut group*

- **RS3** : *F082, Prevotella, Prevotella*

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Establishment of RS in lambs

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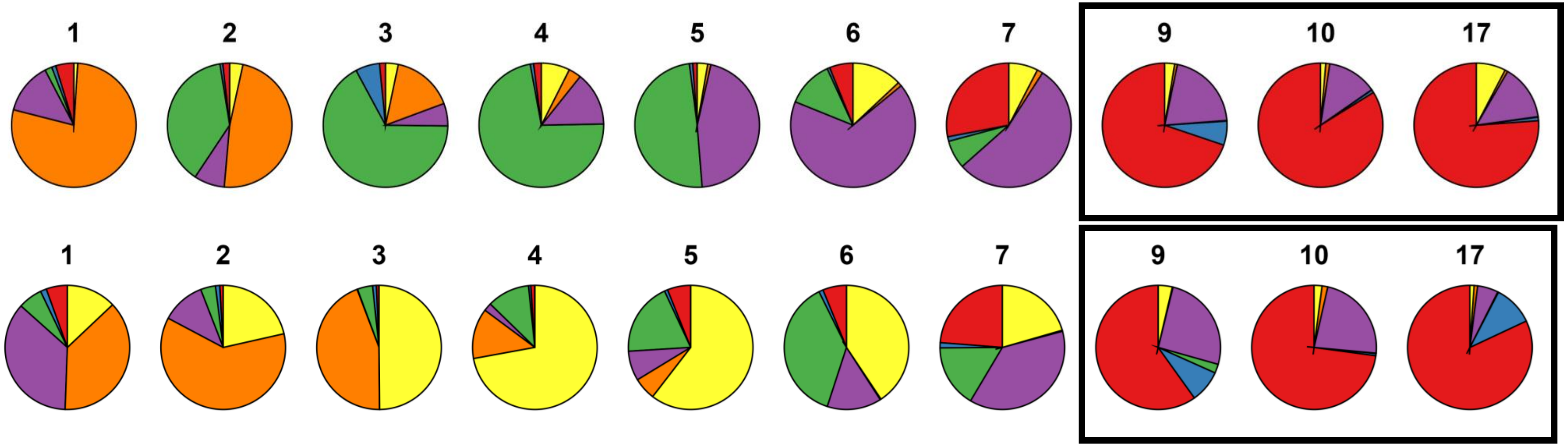

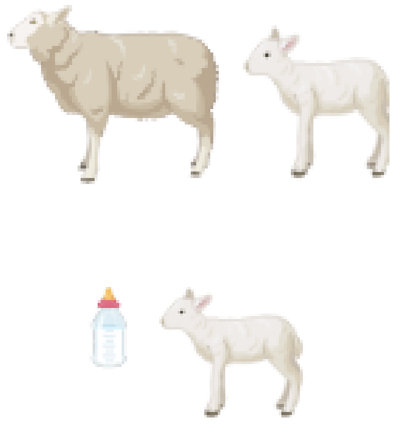

- **RS1** : *Succinivibrionaceae UCG-001, Prevotella, Erysipelotrichaceae UCG-002*

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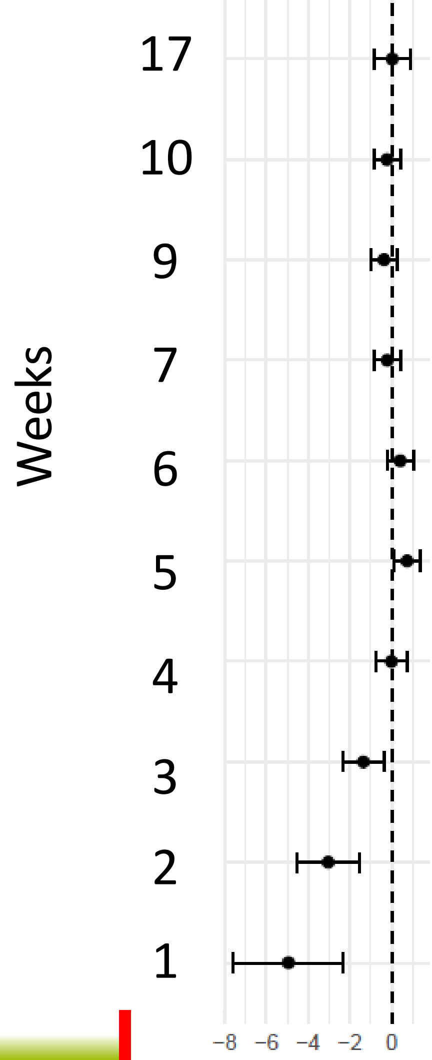
Establishment of RS in lambs

Statistical differences among RuminoSignatures

Characterizing temporal dynamics



Confidence interval at 95 %

Difference ES on mean ajusted

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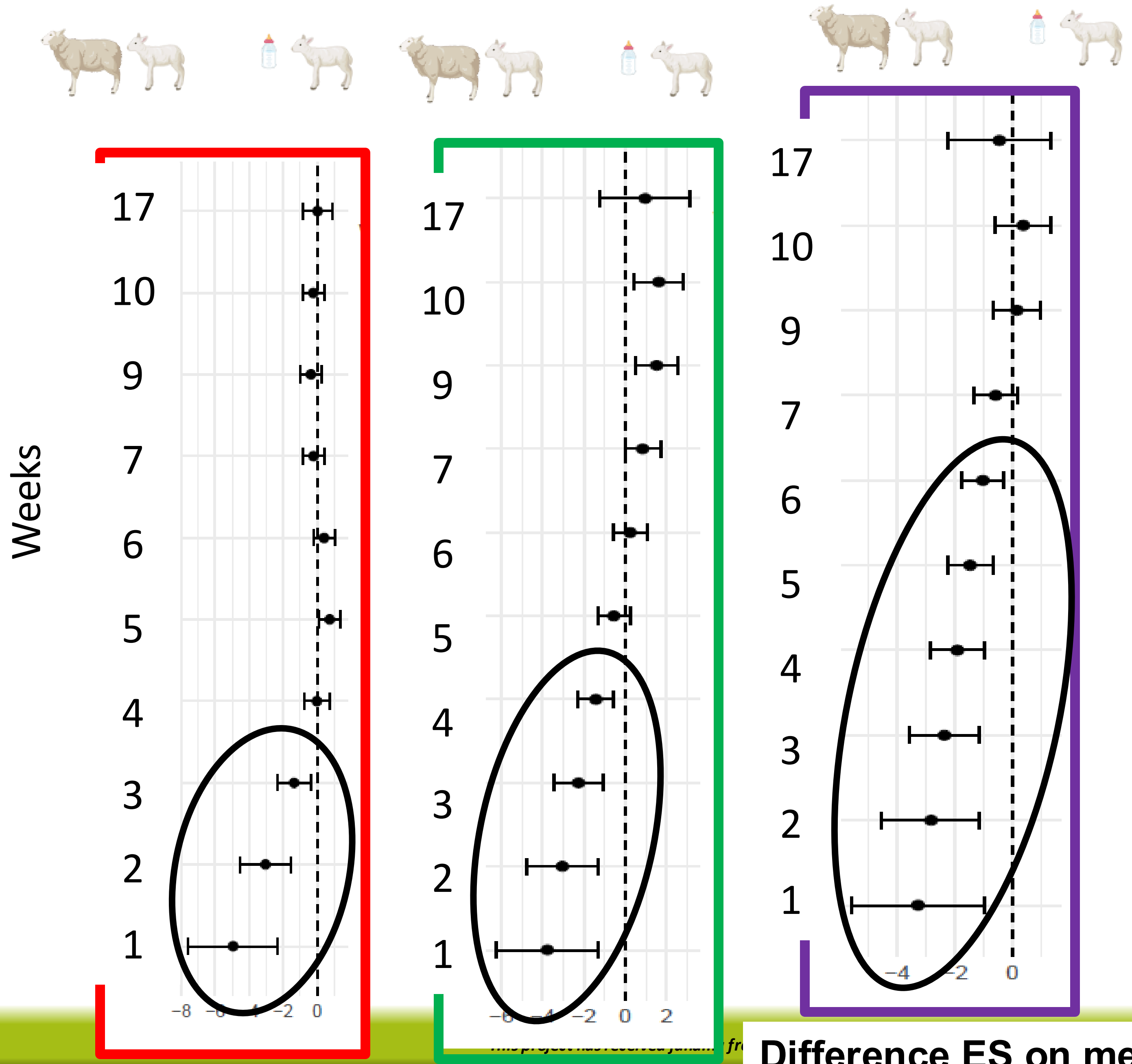
Characterizing temporal dynamics



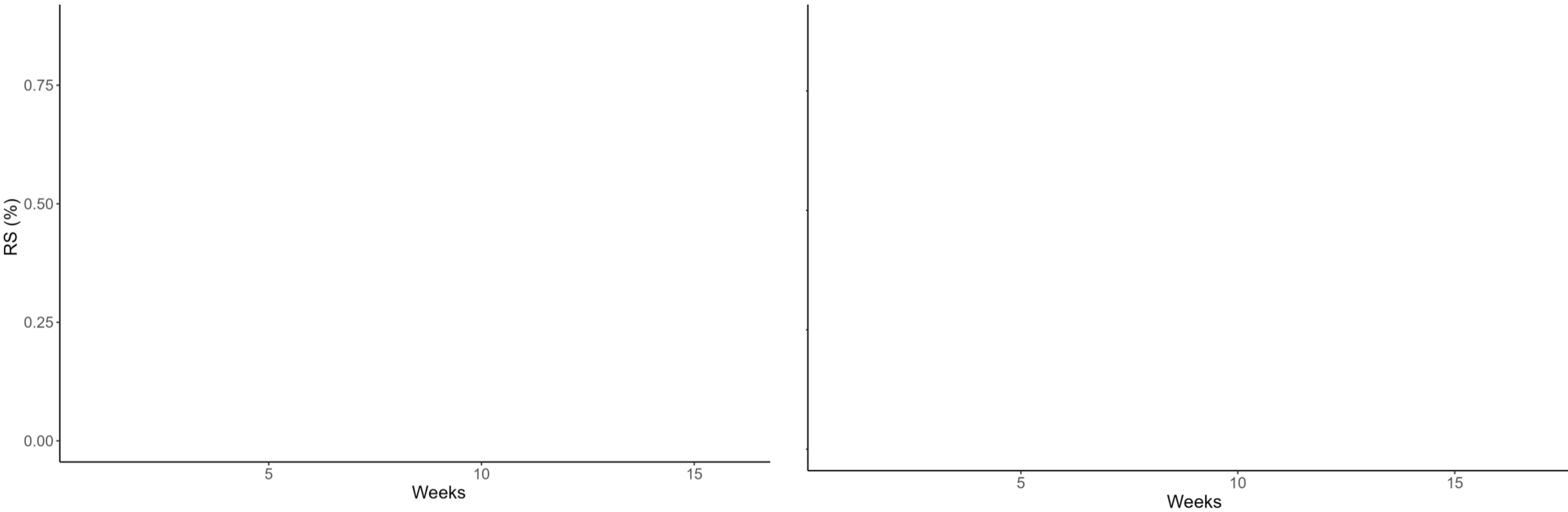
Confidence interval at 95 %

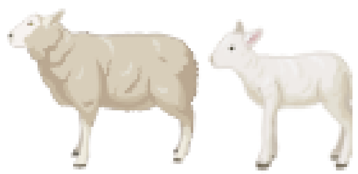
Difference ES on mean ajusted

This project has received fundin fr agreement No 101000213

Characterizing temporal dynamics

Life of lambs





●

RS5 : *Bacteroides, Alloprevotella, Neisseriaceae* : ↑ milk-using bacteria : ***Bacteroides*** *(Amin et Seifert 2021)*

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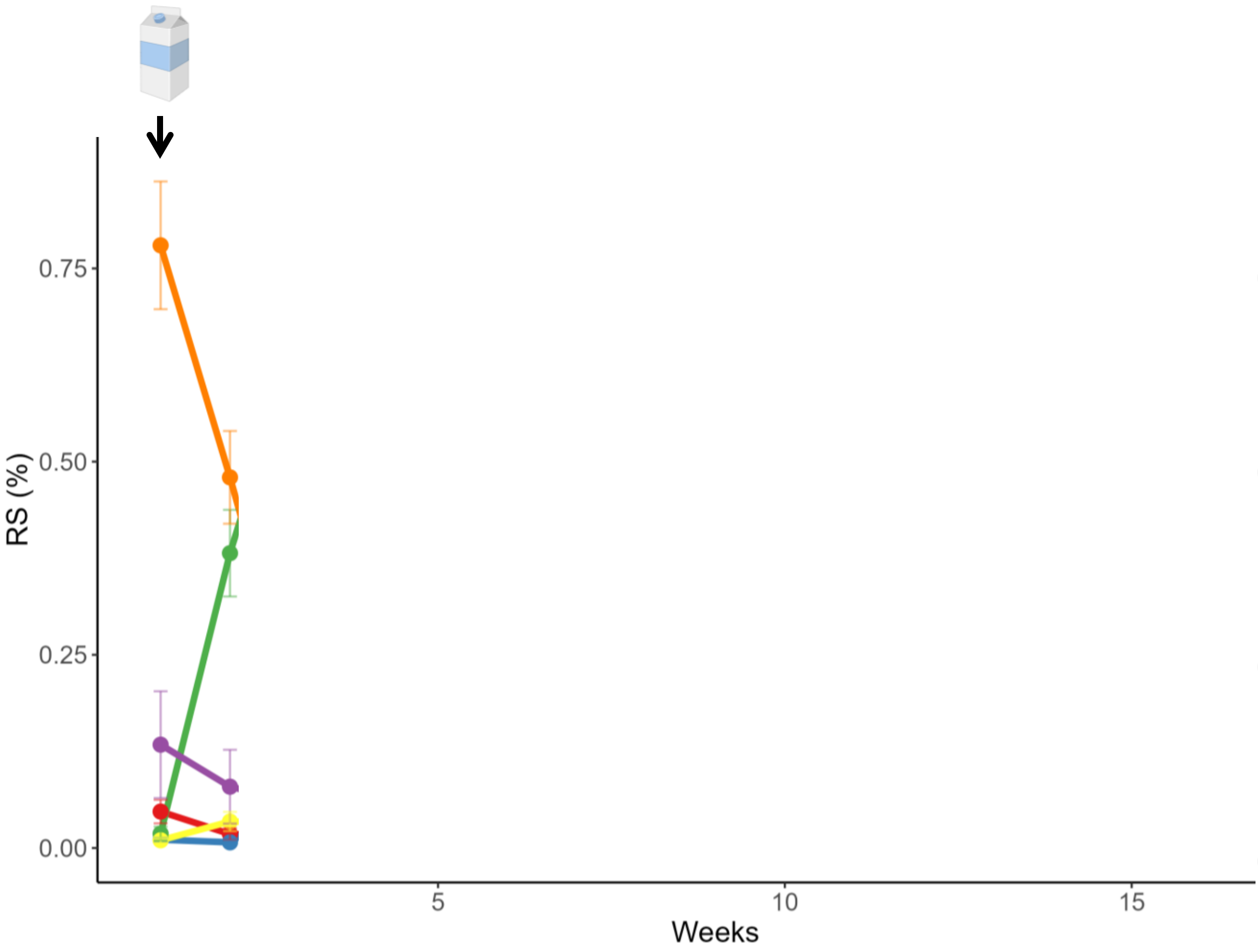

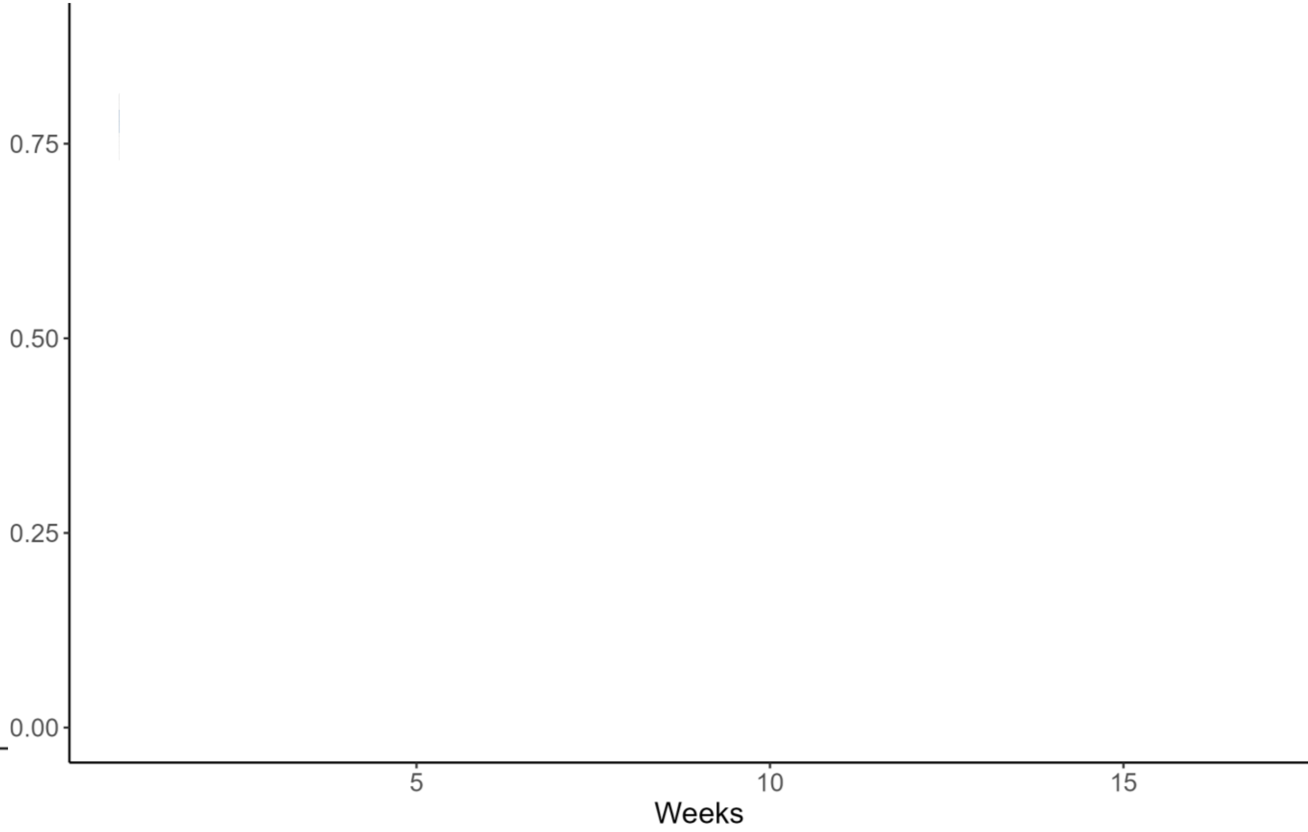

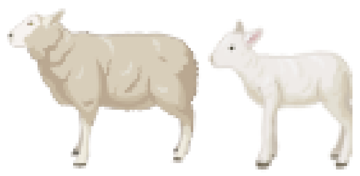

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↗ concentrate (Hartinger et al., 2024) : **genus**
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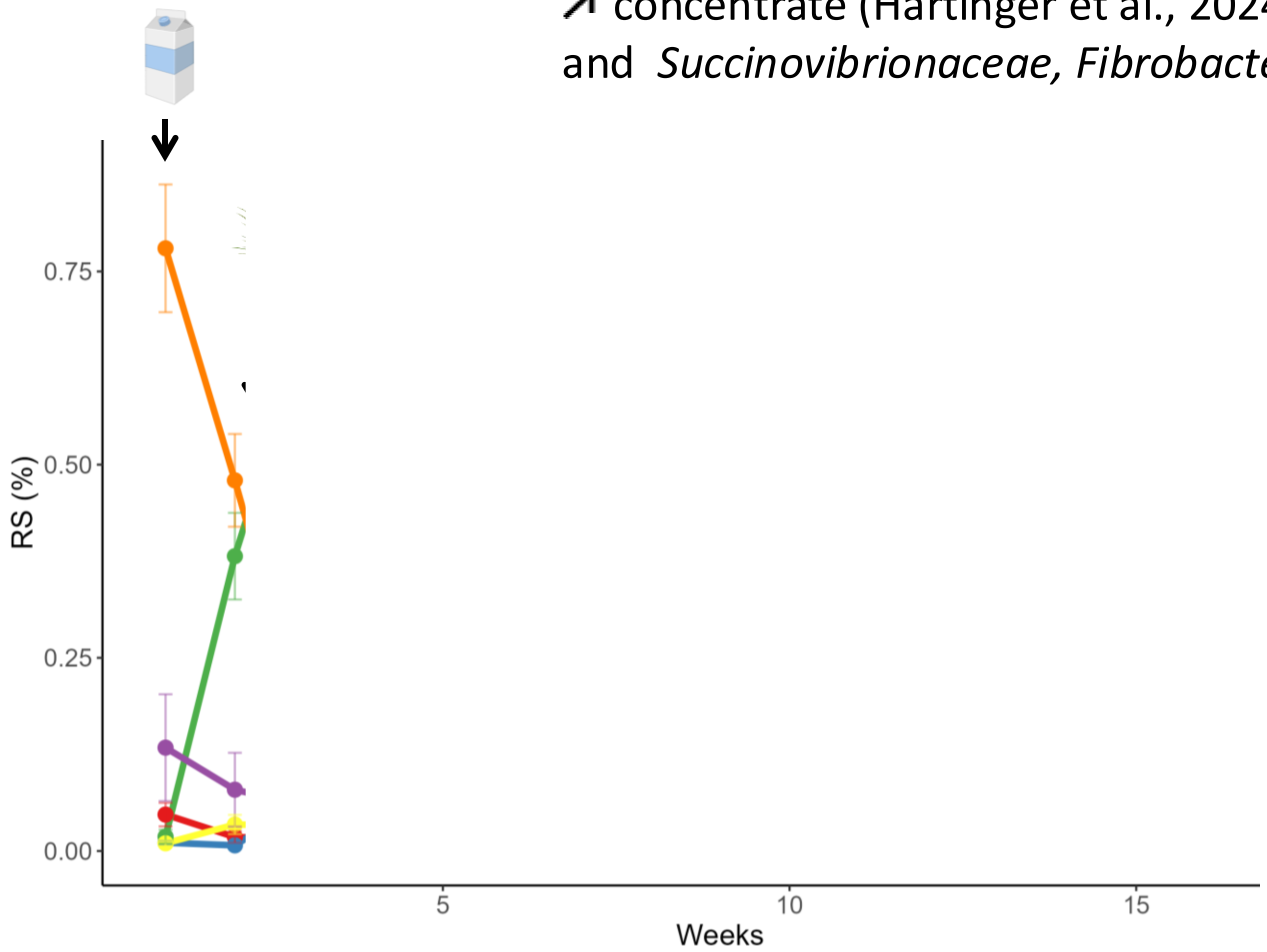

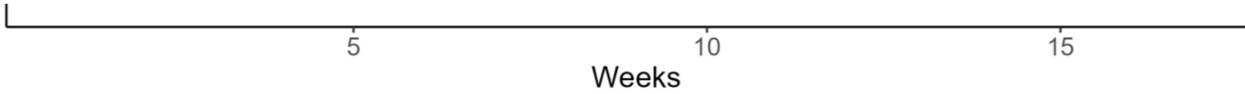

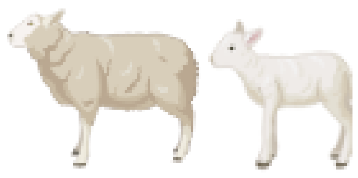

● **•RS3** : *F082*, ***Prevotella*, *Prevotella***

● **RS4** : *Muribaculaceae*, ***Succinivibrionaceae UCG-002***, *Succiniclasticum*

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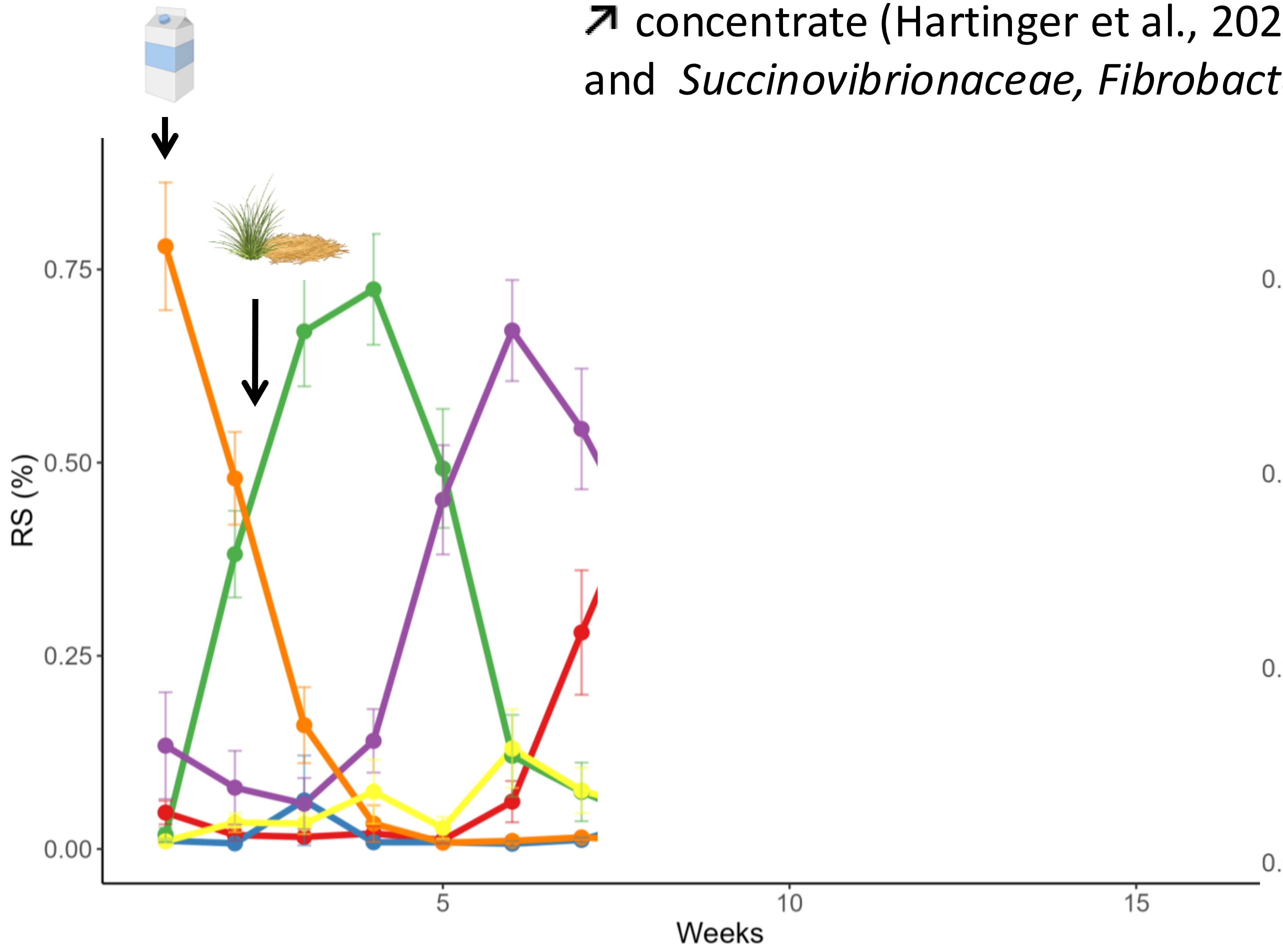

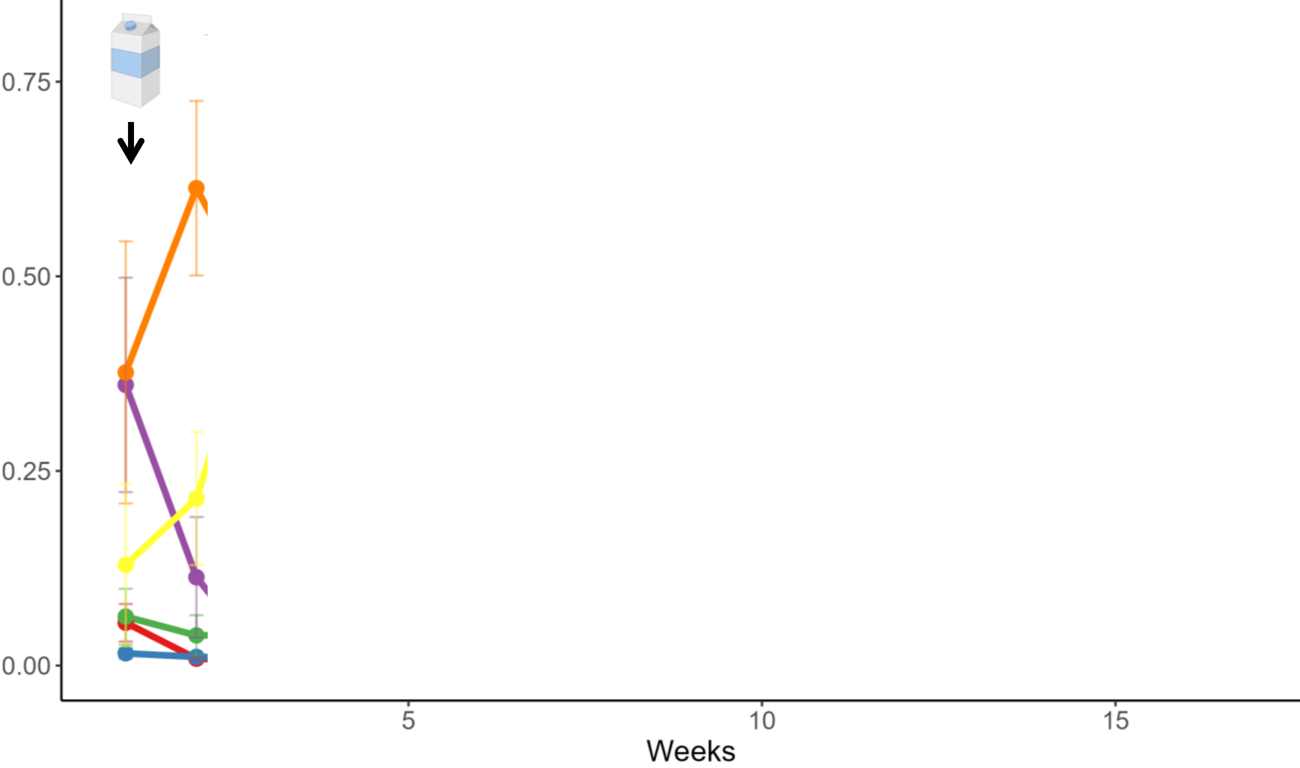

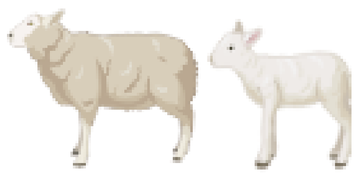

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● **•RS6** : ***Sharpea*, *Prevotella*** *Rikenellaceae RC9 gut group*

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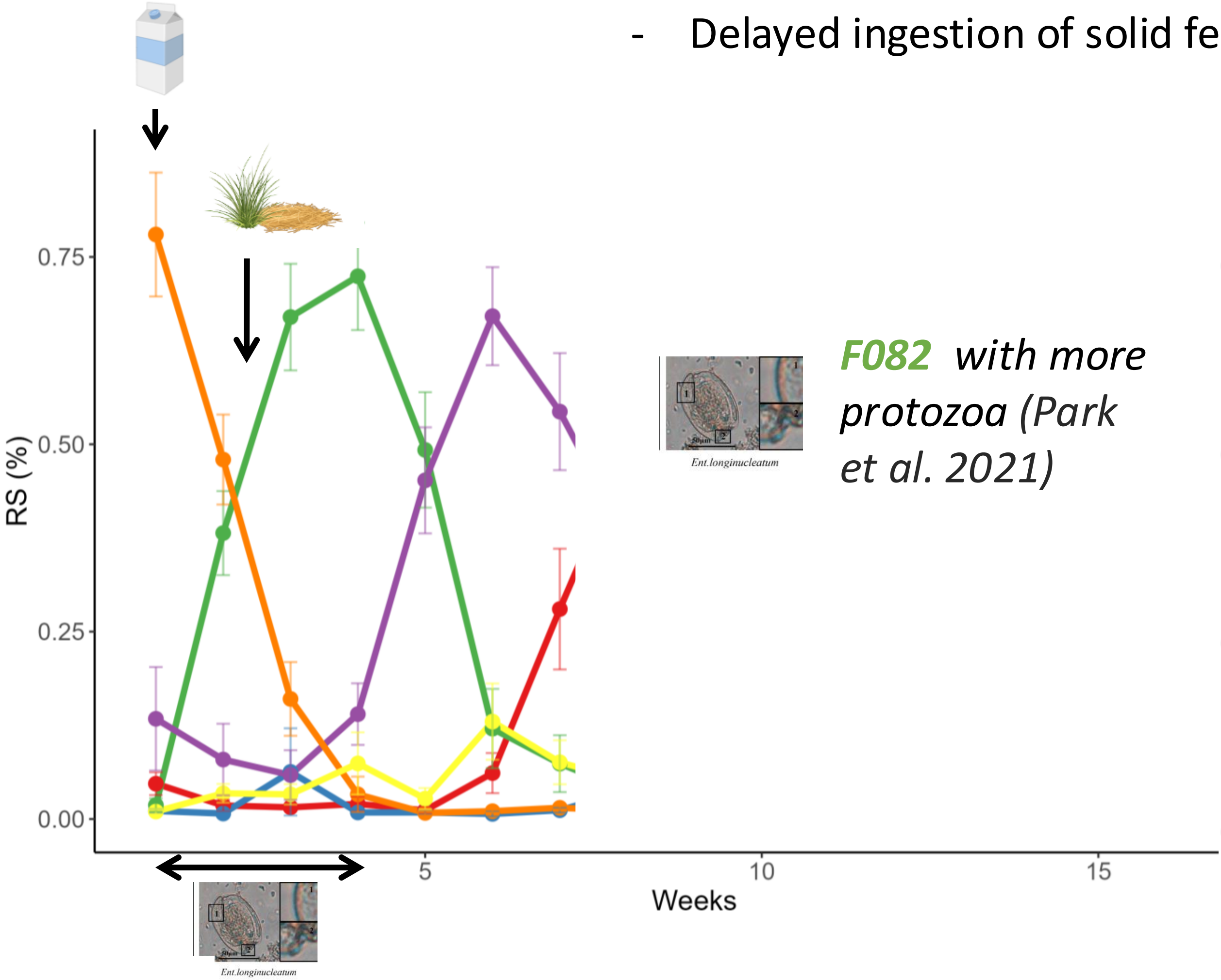
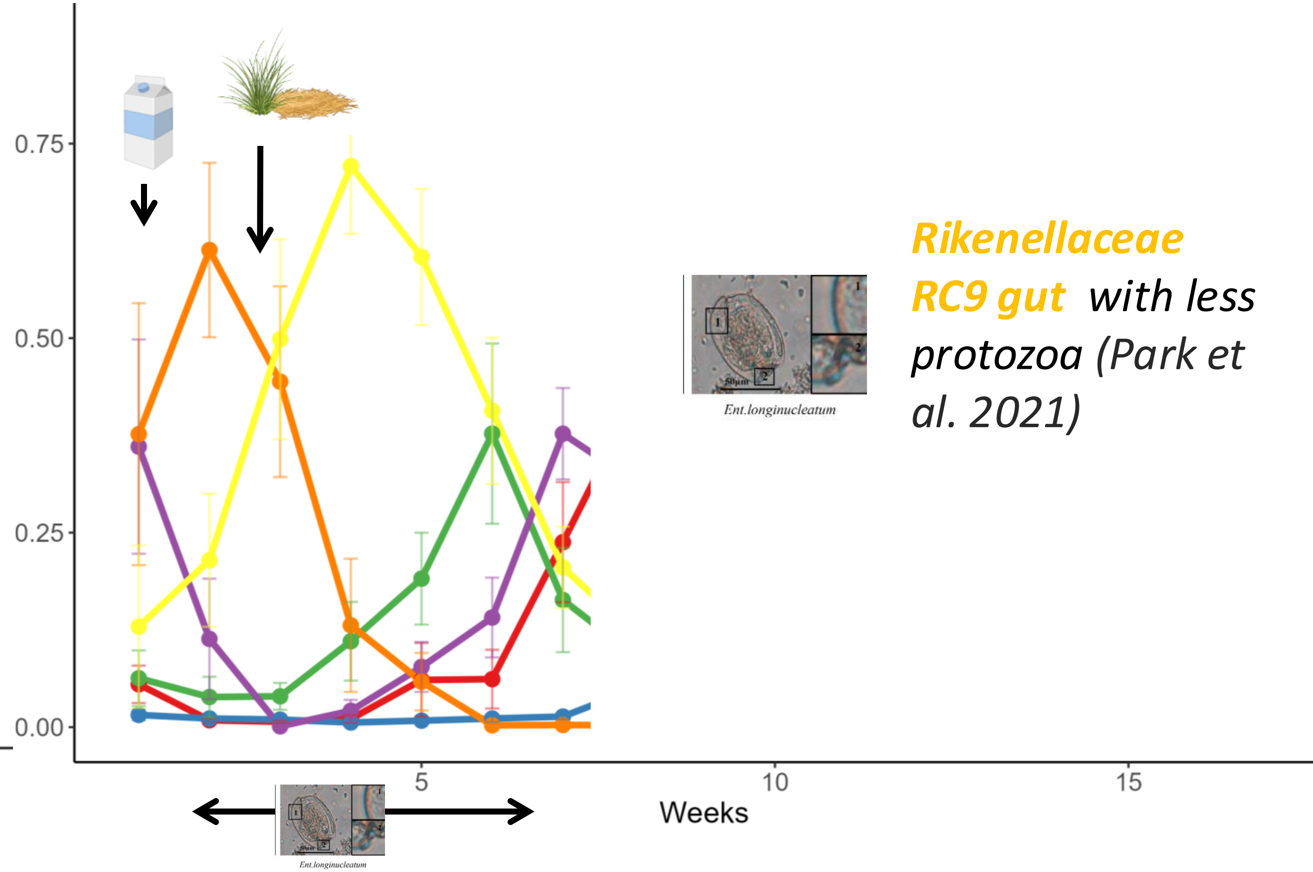
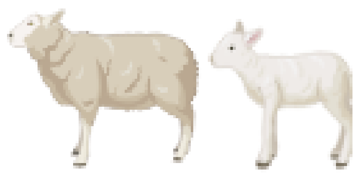
- Protozoa implantation ?
- Delayed ingestion of solid feeds ?



F082 *with more protozoa (Park et al. 2021)*



Rikenellaceae RC9 gut *with less protozoa (Park et al. 2021)*



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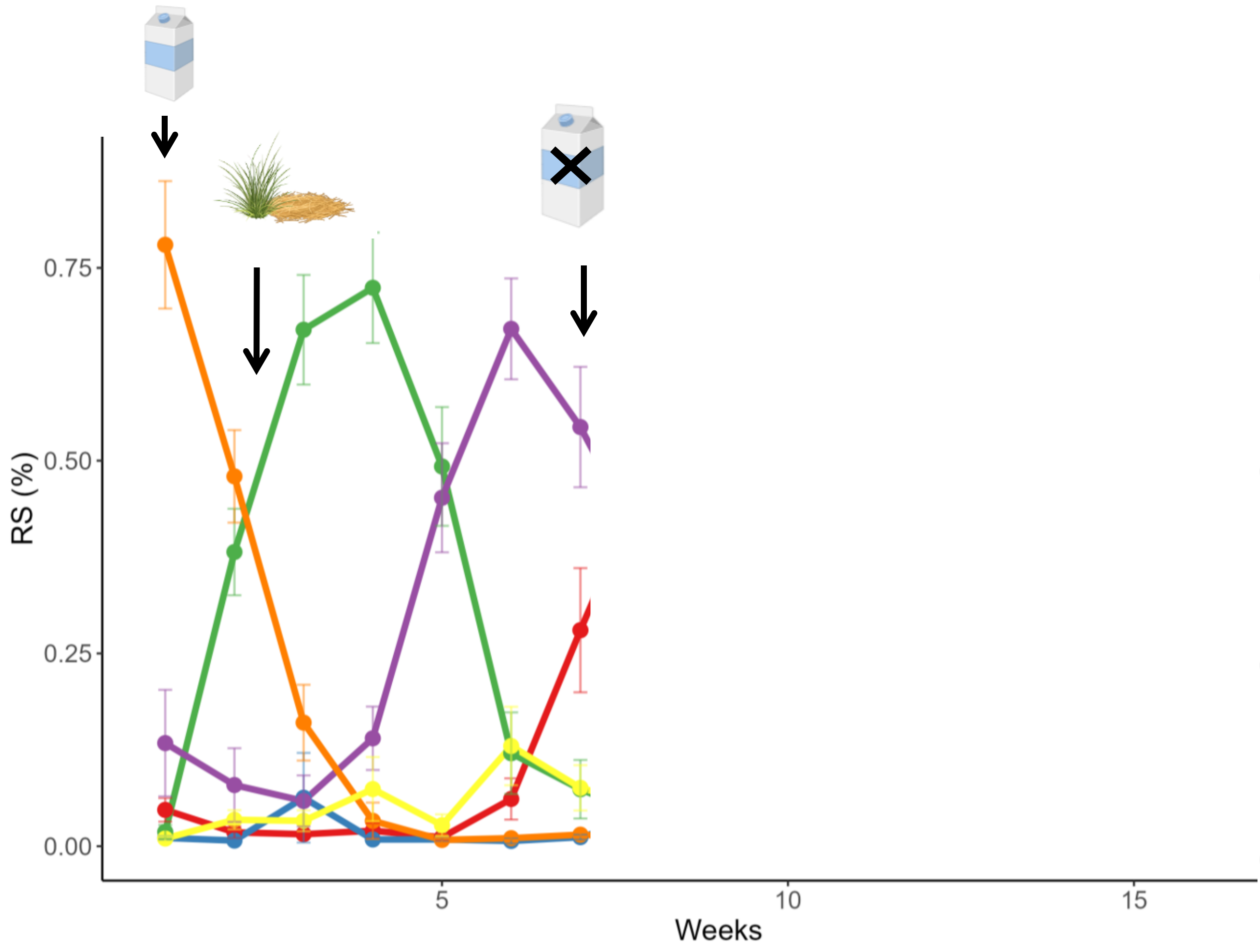

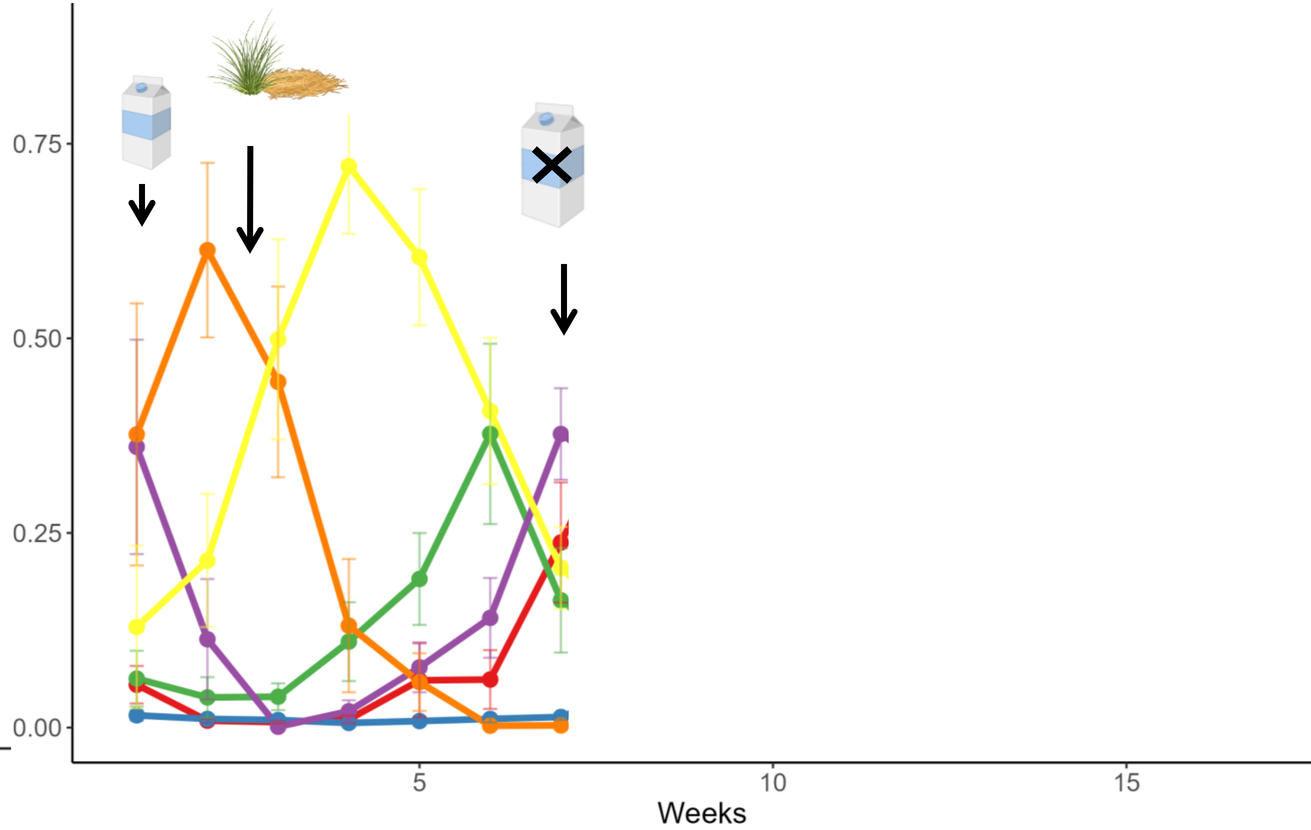

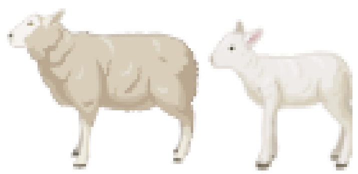

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Messages and outlook

Enterosignature is a good way to follow dynamic of microbes' colonization

Two signatures are common to both systems: orange at the beginning of life and red after weaning.

Signatures are modulated by the rearing system before weaning

Next step :

- link to protozoa implantation
- With pertubation
- Go deeper to understand variability among artificial group

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Thank you for your attention

Dinamic team

HoloRuminants partners

HoloRuminant
Understanding microbiomes of the ruminant holobiont

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ÉCOLE DOCTORALE
DES SCIENCES DE LA VIE,
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UMR1213 Herbivores
VetAgro Sup

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	Groups	ENM	EM
Sex	Female	7	8
	Male	6	11
Birth weight (mean ± standard deviation)		4.02 ± 1.03	4.02 ± 0.76
Number of lambs		13	19

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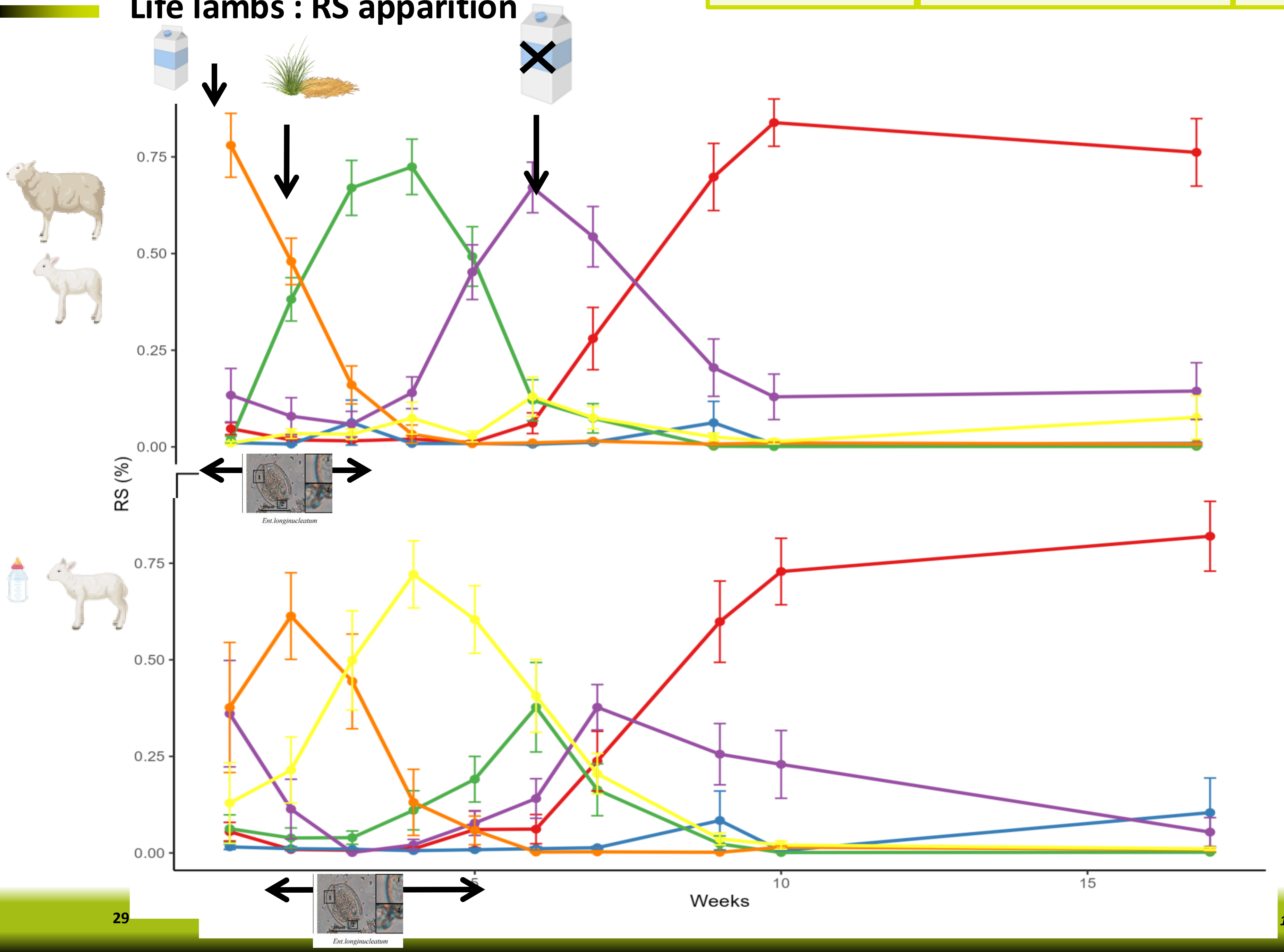
Life lambs : RS apparition



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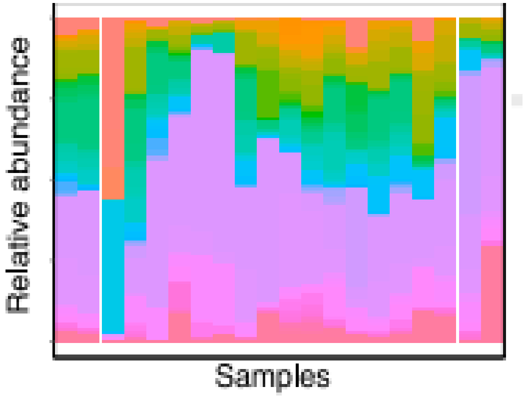
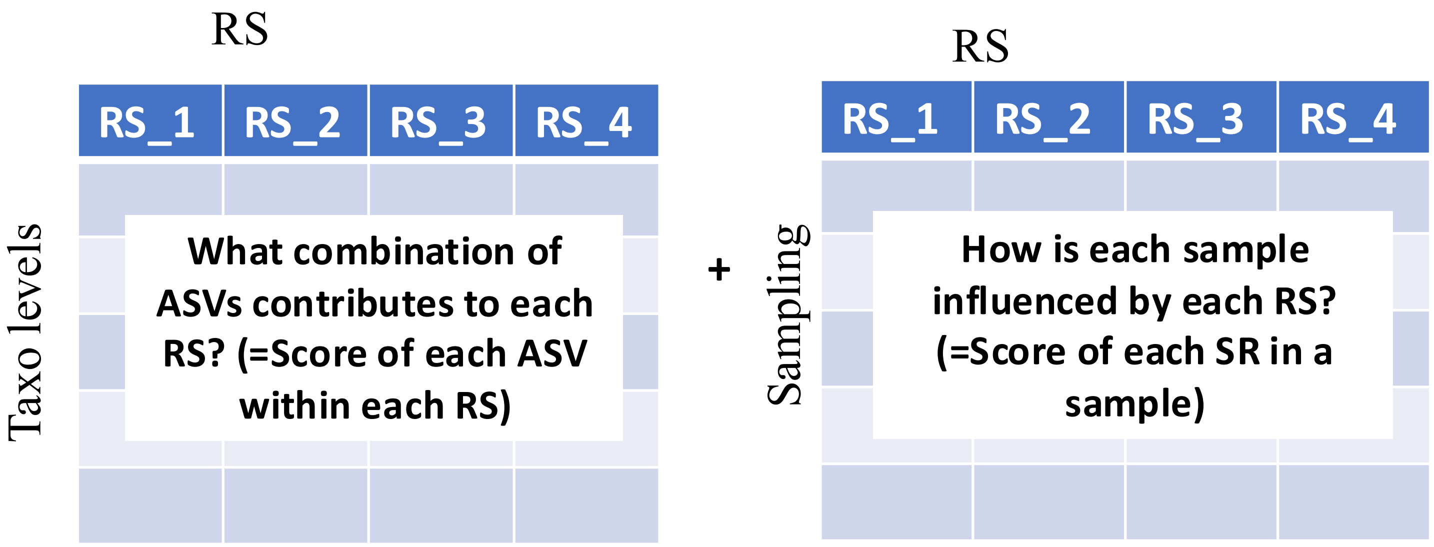
101000213

RuminoSignatures, Non-matrix factoring (NMF)

ASV composition observed in each sample

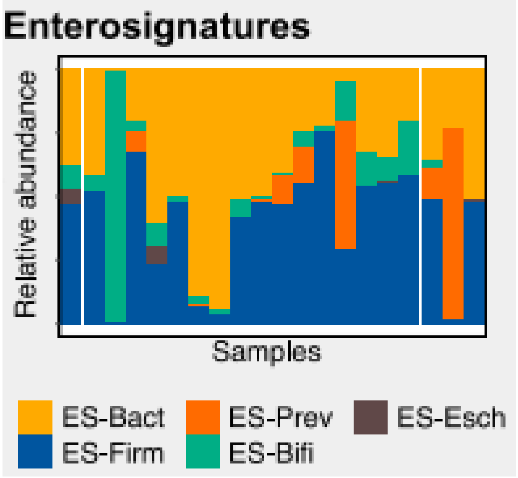


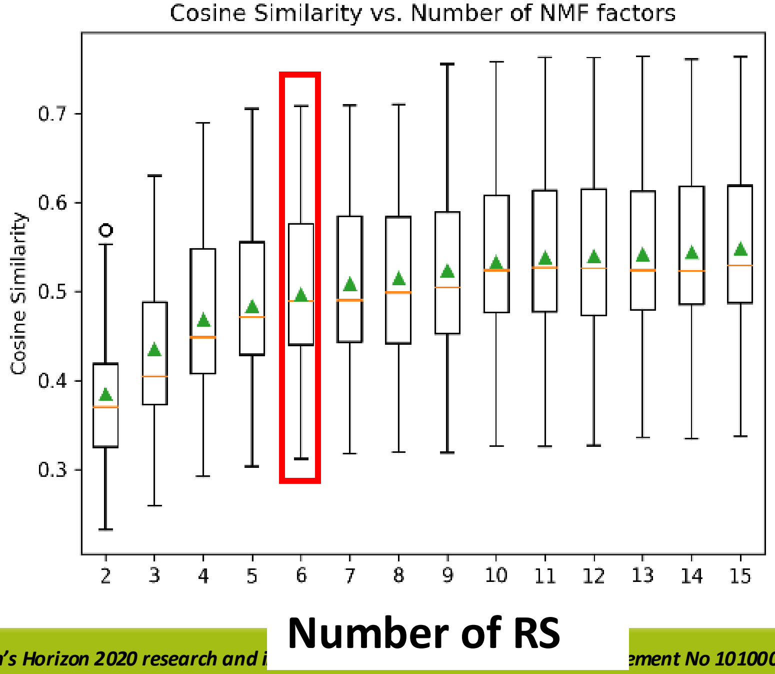
NMF/ RuminoSignatures (RS)



What combination of ASVs contributes to each RS? (=Score of each ASV within each RS)

How is each sample influenced by each RS? (=Score of each SR in a sample)





Optimum number of signatures

Evolution according to RS:
Model learning platform =
No significant improvement

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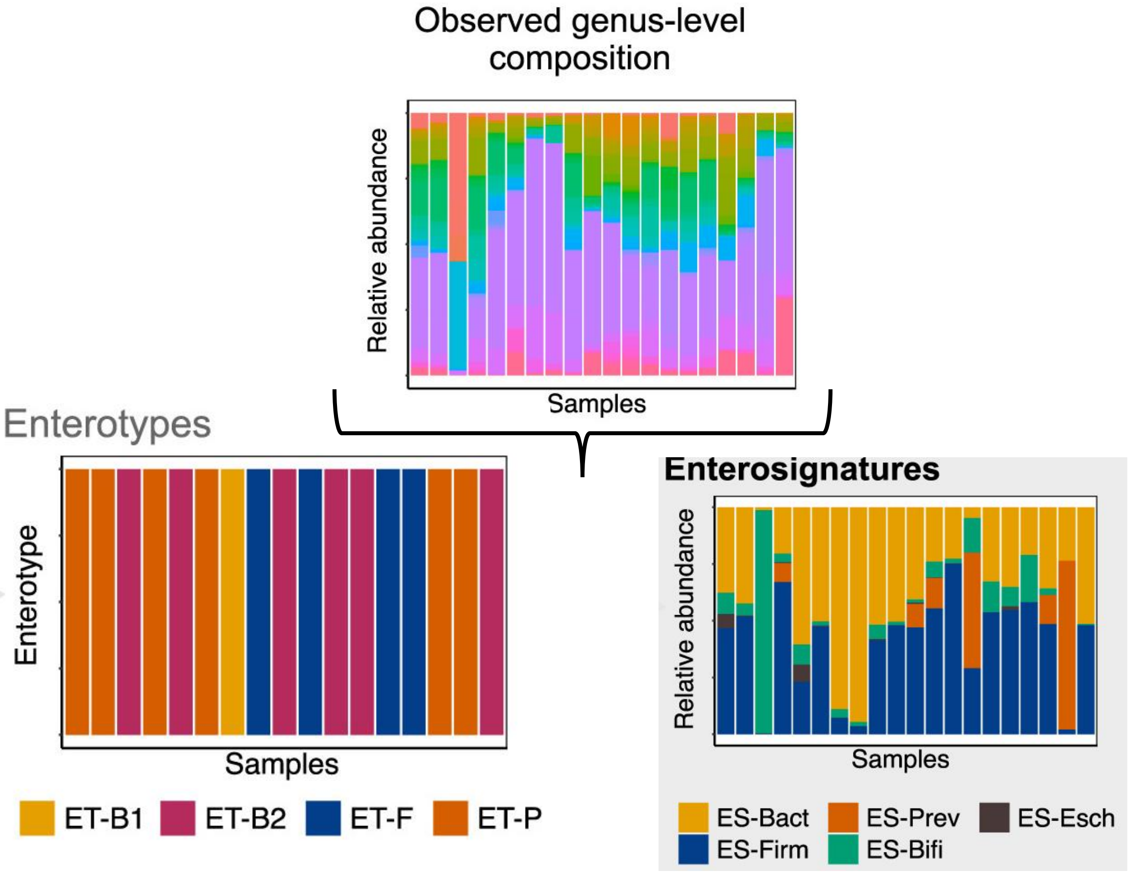
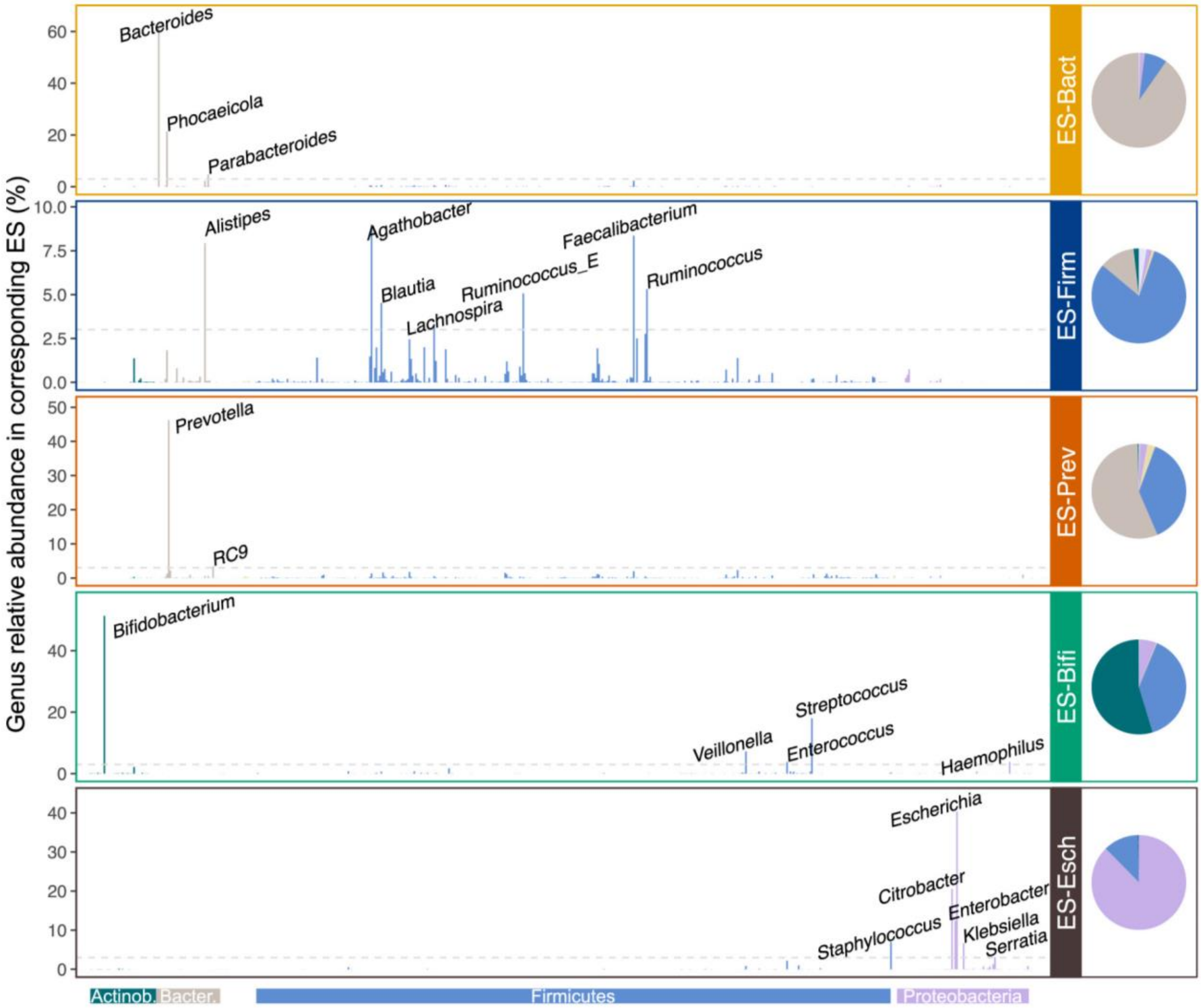
Enterotypes vs Enterosignatures (Microbial guilds)



GENERA CONTRIBUTING TO ESs



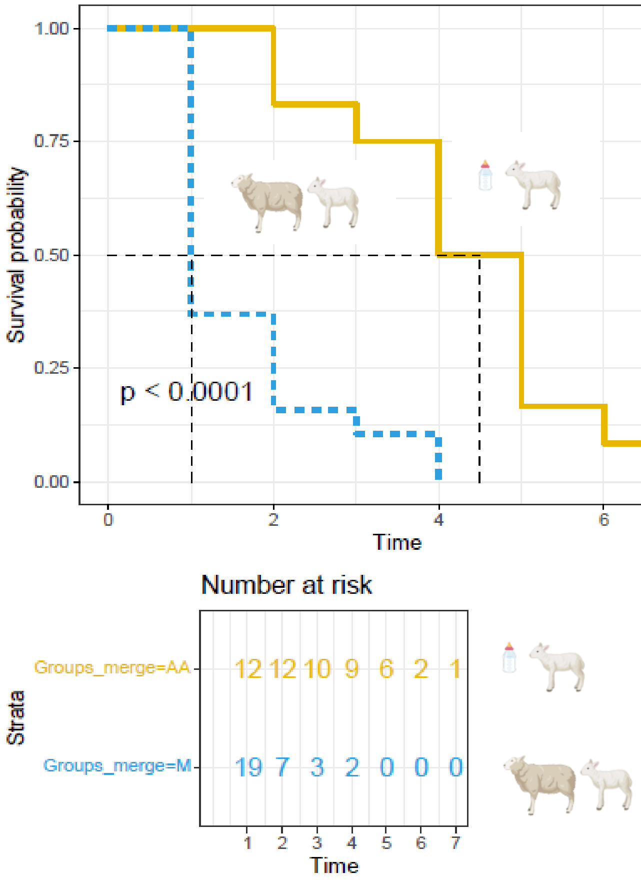
This project has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 101000213

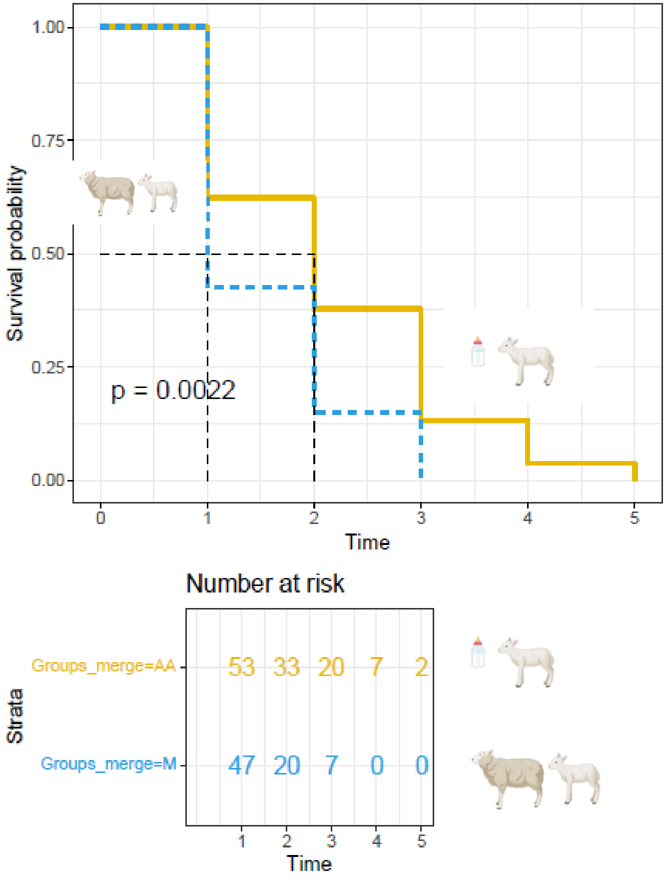
What we do after

Eukaryotic implantation contributes to the establishment of bacterial RS ?

RuminoSignature : ASV

Implantation of eukaryotes





How can we demonstrate that these eukaryotes *drive* some of the observed rumen signatures?

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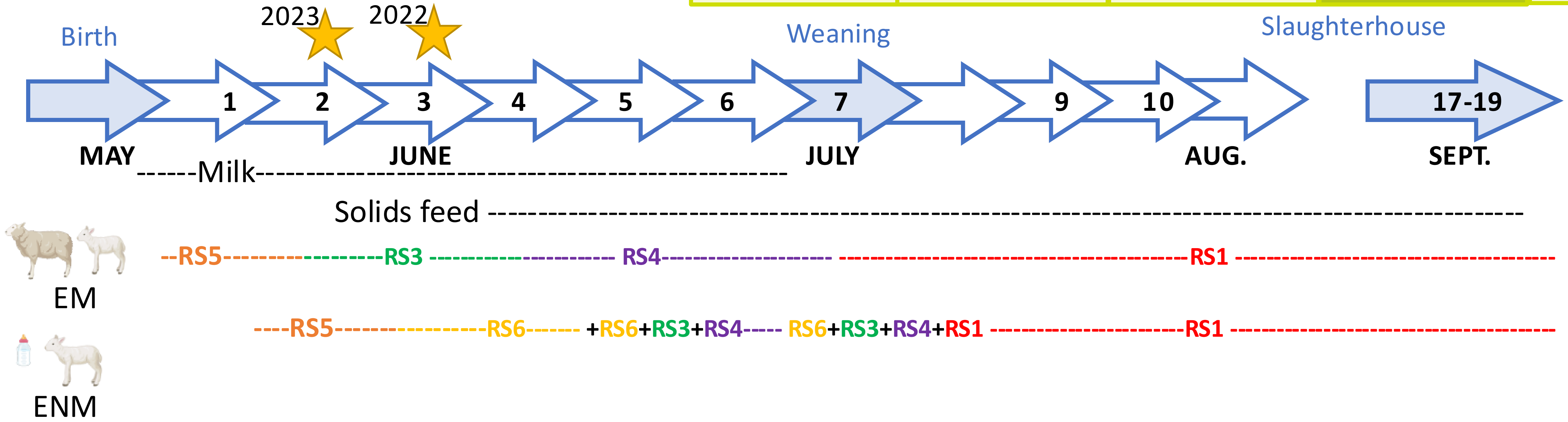
Characterizing temporal dynamics



Birth

2023 ★

2022 ★

Weaning

Slaughterhouse

1 2 3 4 5 6 7 9 10 17-19

MAY JUNE JULY AUG. SEPT.

------Milk---

Solids feed --

EM

--RS5------------------RS3 ----------------------- RS4-------------------- --RS1 --

ENM

----RS5-----------------RS6------- +RS6+RS3+RS4----- RS6+RS3+RS4+RS1 ----------------------RS1 --

- **RS1** : *Succinivibrionaceae UCG-001* ✖, *Prevotella* ✖, *Erysipelotrichaceae UCG-002*
- **RS3** : *F082* ❎, *Prevotella* ✖, *Prevotella* ✖
- **RS4** : *Muribaculaceae*, *Succinivibrionaceae UCG-002* ✖, *Succiniticlasticum*
- **RS5** : *Bacteroides*, *Alloprevotella*, *Neisseriaceae* : ↑ milk-using bacteria : ***Bacteroides*** *(Amin et Seifert 2021)*
- **RS6** : *Sharpea* ✖, *Rikenellaceae RC9 gut group* ❎, *Prevotella* ✖

✖ Added concentrate: increases aundance (Hartinger et al., 2024)
b
↑ abundance of amylolytic and fibrolytic bacteria :
Succinovibrionaceae, Fibrobacteraceae,Prevotellaceae
(Amin et Seifert 2021)

❎ ***Rikenellaceae RC9 gut found with rumen not enriched with protozoa*** *Park et al. 2021)*
F082 discovered with more protozoa *(Park et al. 2021)*

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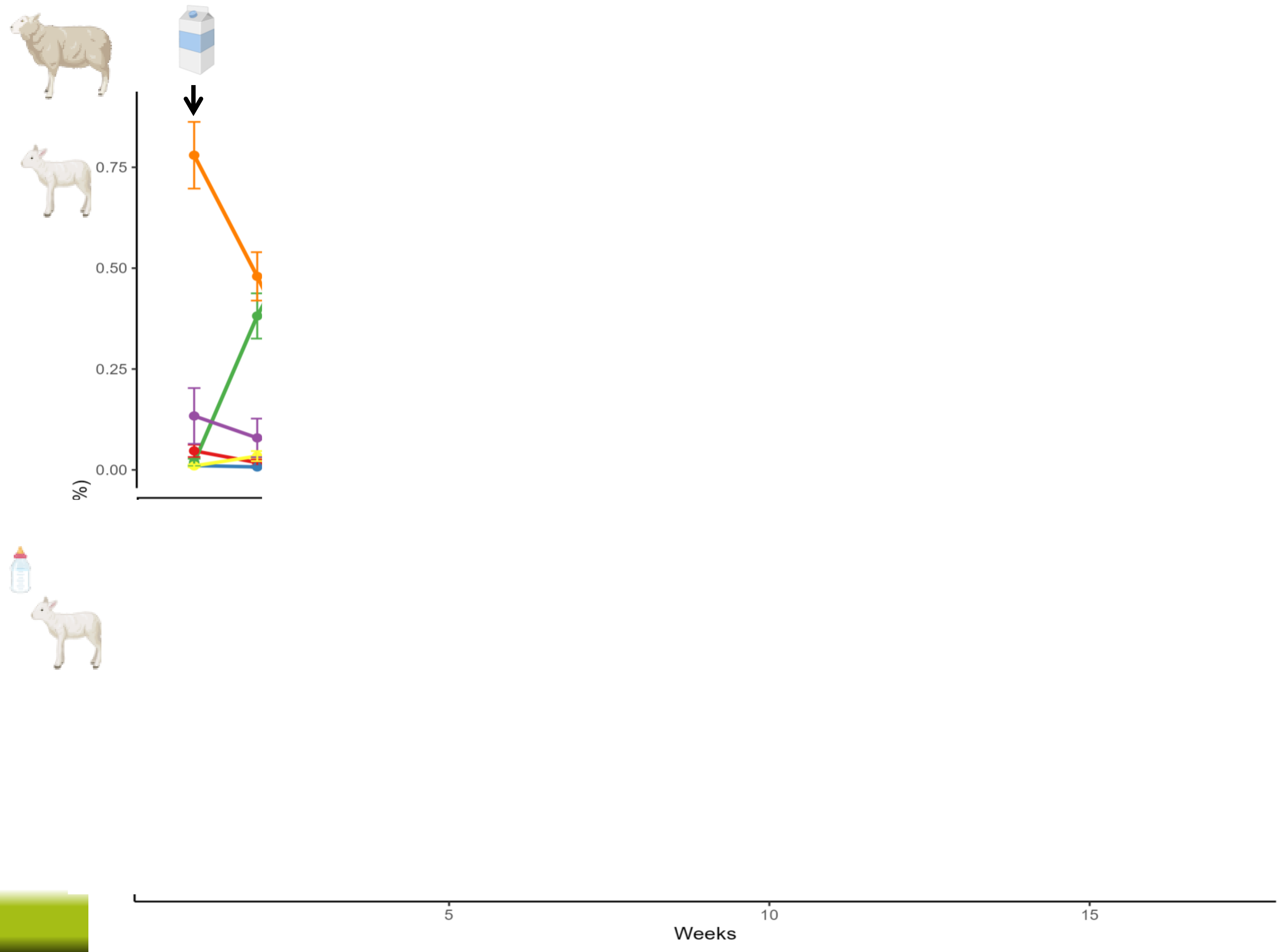
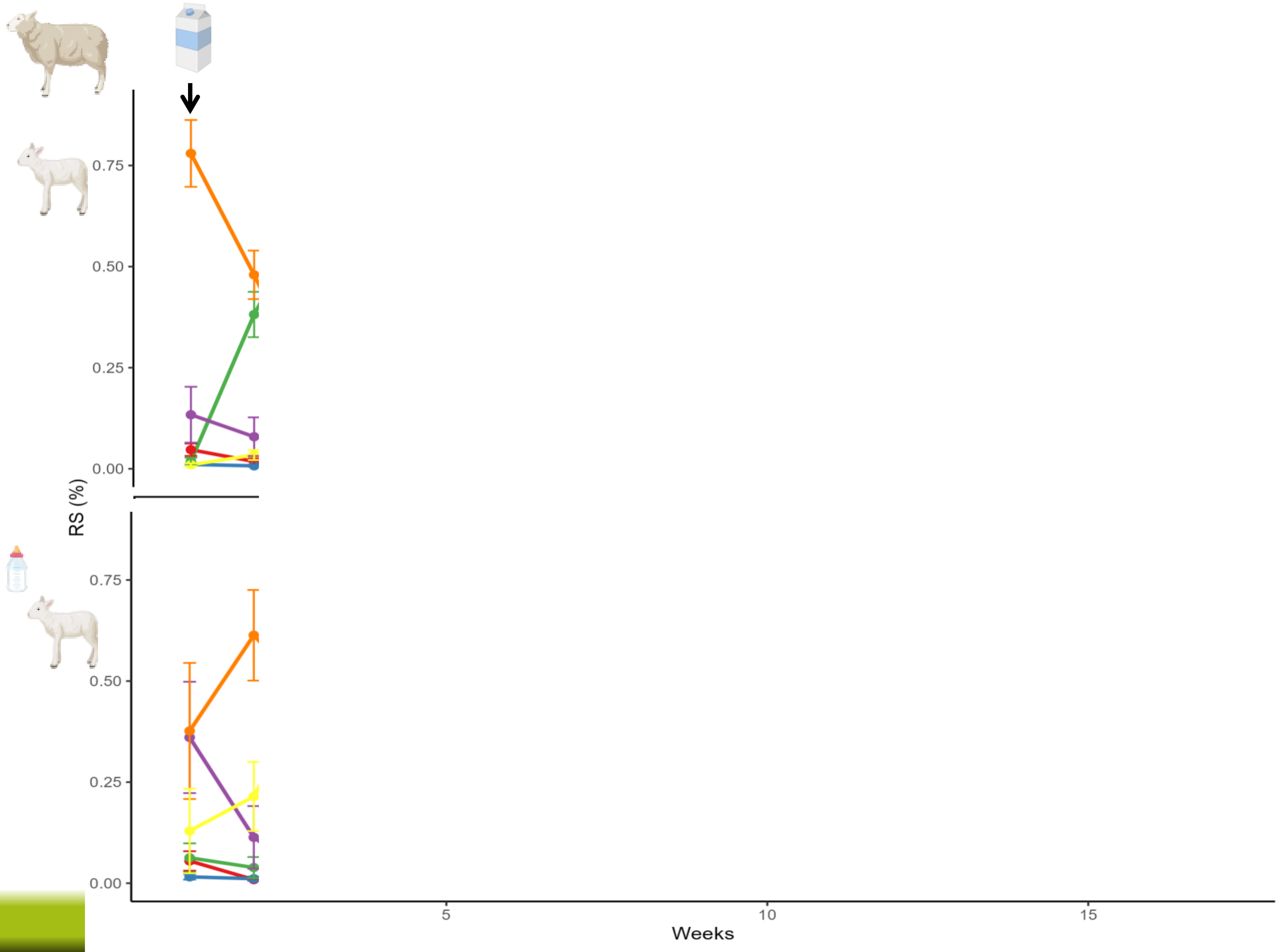
Life lambs : RS apparition



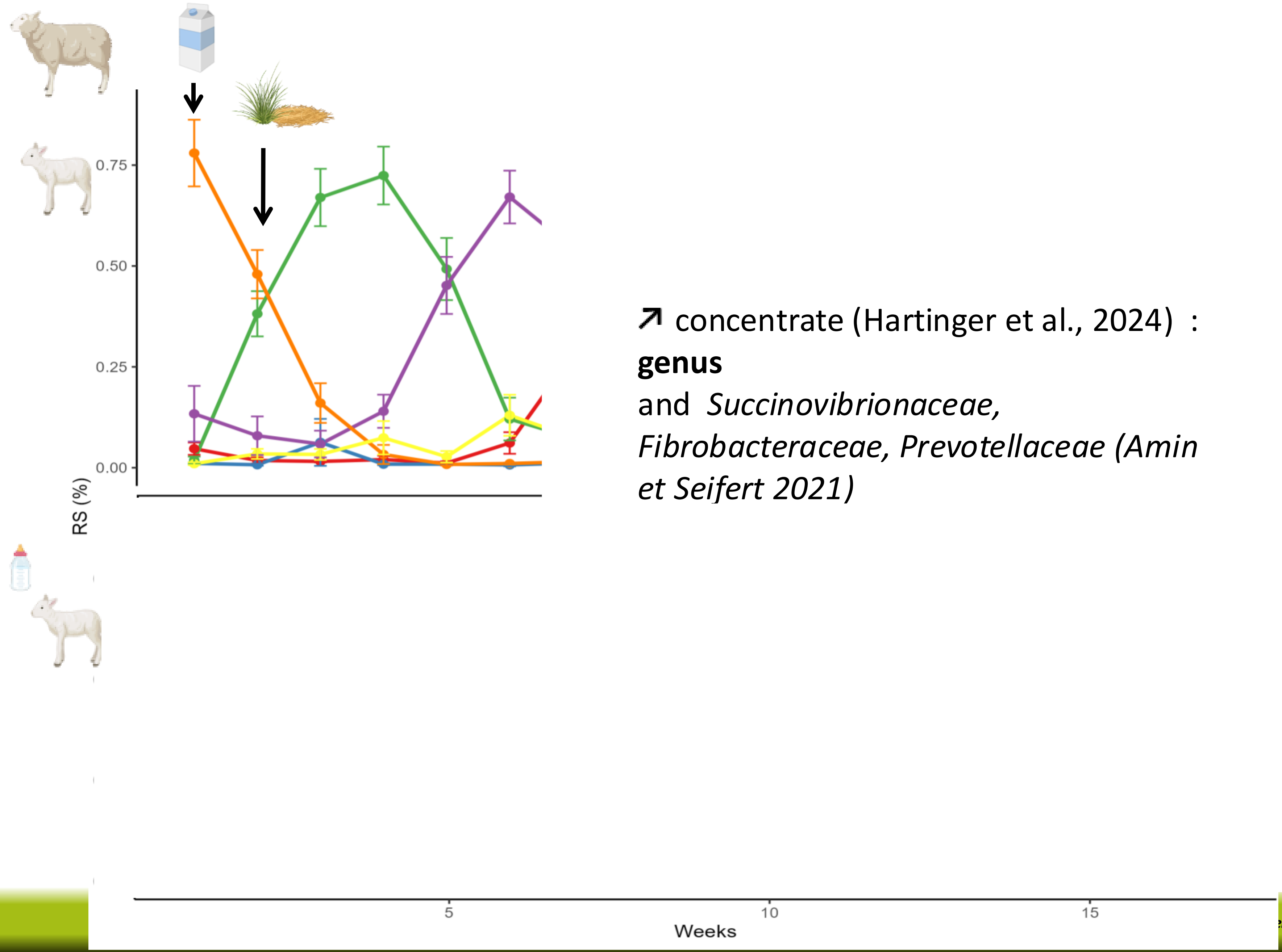
● **RS5** : *Bacteroides, Alloprevotella, Neisseriaceae :* ↑ milk-using bacteria : ***Bacteroides*** *(Amin et Seifert 2021)*

Life lambs : RS apparition



● **RS5** : *Bacteroides, Alloprevotella, Neisseriaceae :* ↑ milk-using bacteria : ***Bacteroides*** *(Amin et Seifert 2021)*

Life lambs : RS apparition

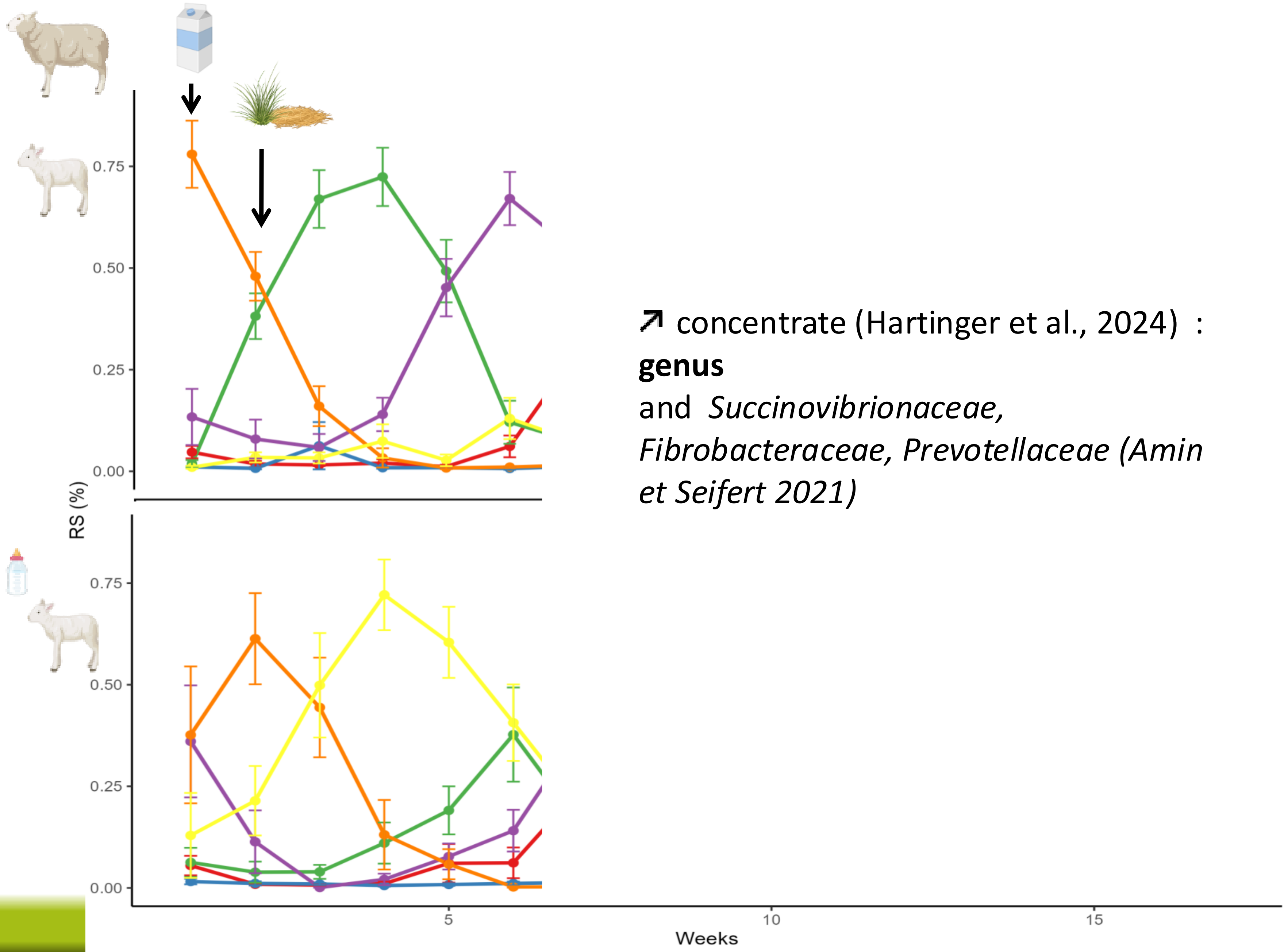


↗ concentrate (Hartinger et al., 2024) : **genus**
and *Succinovibrionaceae, Fibrobacteraceae, Prevotellaceae (Amin et Seifert 2021)*

●•**RS3** : *F082*, ***Prevotella*, *Prevotella***

● **RS4** : *Muribaculaceae*, ***Succinivibrionaceae UCG-002***, *Succiniticlasticum*

Life lambs : RS apparition



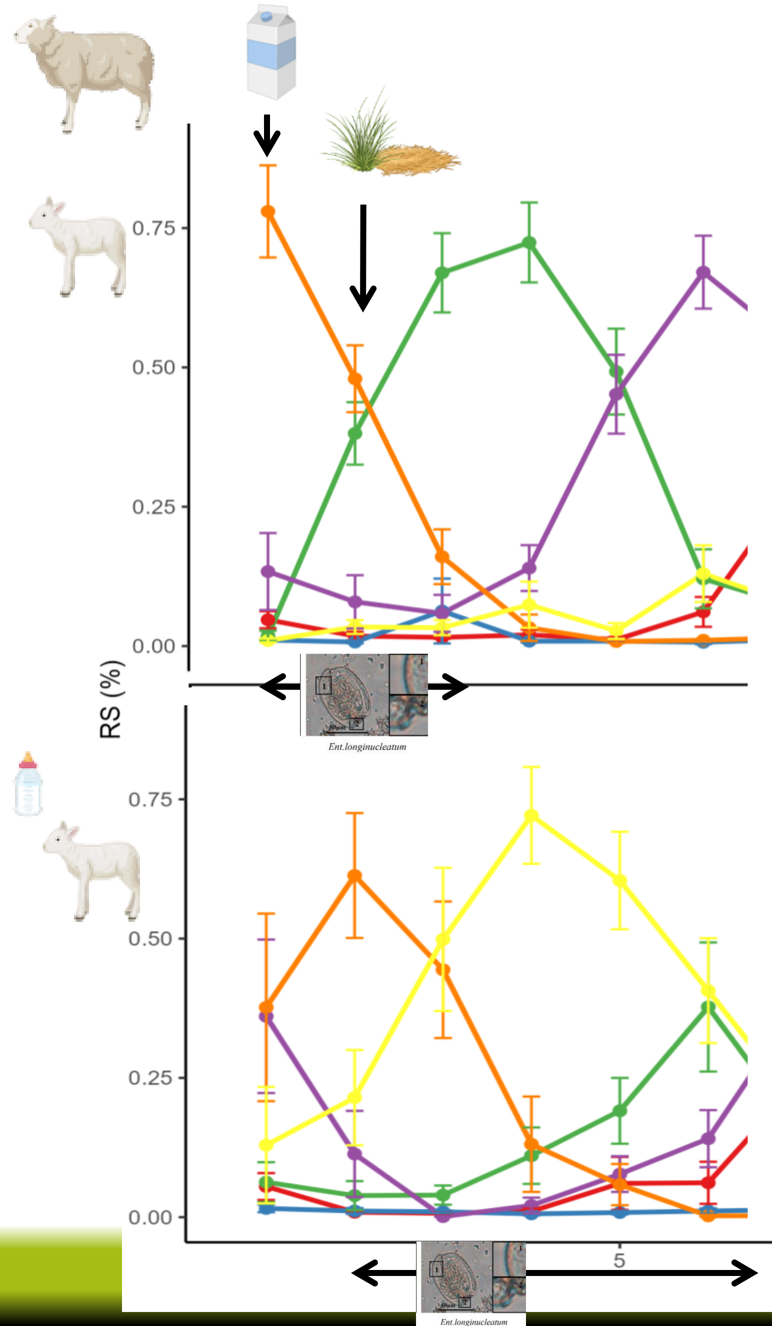
↗ concentrate (Hartinger et al., 2024) : **genus** and *Succinovibrionaceae, Fibrobacteraceae, Prevotellaceae (Amin et Seifert 2021)*

—●— •RS3 : *F082*, ***Prevotella***, ***Prevotella***

—●— RS4 : *Muribaculaceae*, ***Succinivibrionaceae UCG-002***, *Succiniticlasticum*

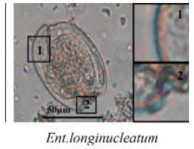
—●— •RS6 : ***Sharpea***, ***Prevotella*** *Rikenellaceae RC9 gut group*

Life lambs : RS apparition



↗ concentrate (Hartinger et al., 2024) : **genus** and *Succinovibrionaceae, Fibrobacteraceae, Prevotellaceae (Amin et Seifert 2021)*

- Protozoa implantation ?
- Late of solides feed ingestion ?



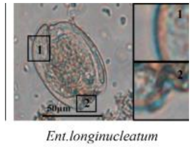
F082 *with more protozoa (Park et al. 2021)*

●

•RS3 : *F082*, ***Prevotella***, ***Prevotella***

●

RS4 : *Muribaculaceae*, ***Succinivibrionaceae UCG-002***, *Succiniticlasticum*

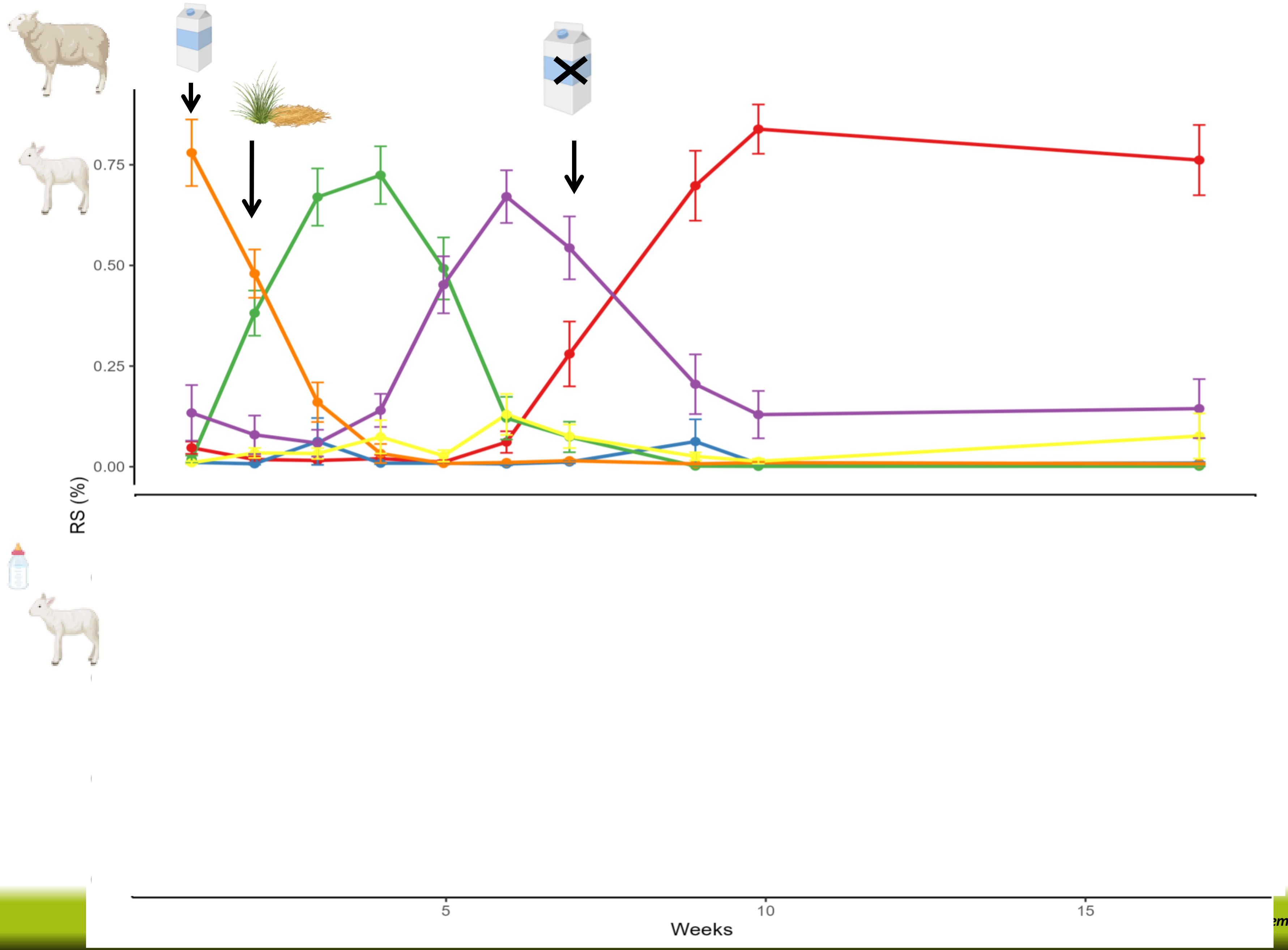


Rikenellaceae RC9 gut *with less protozoa (Park et al. 2021)*

●

•RS6 : ***Sharpea***, ***Prevotella*** *Rikenellaceae RC9 gut group*

Life lambs : RS apparition

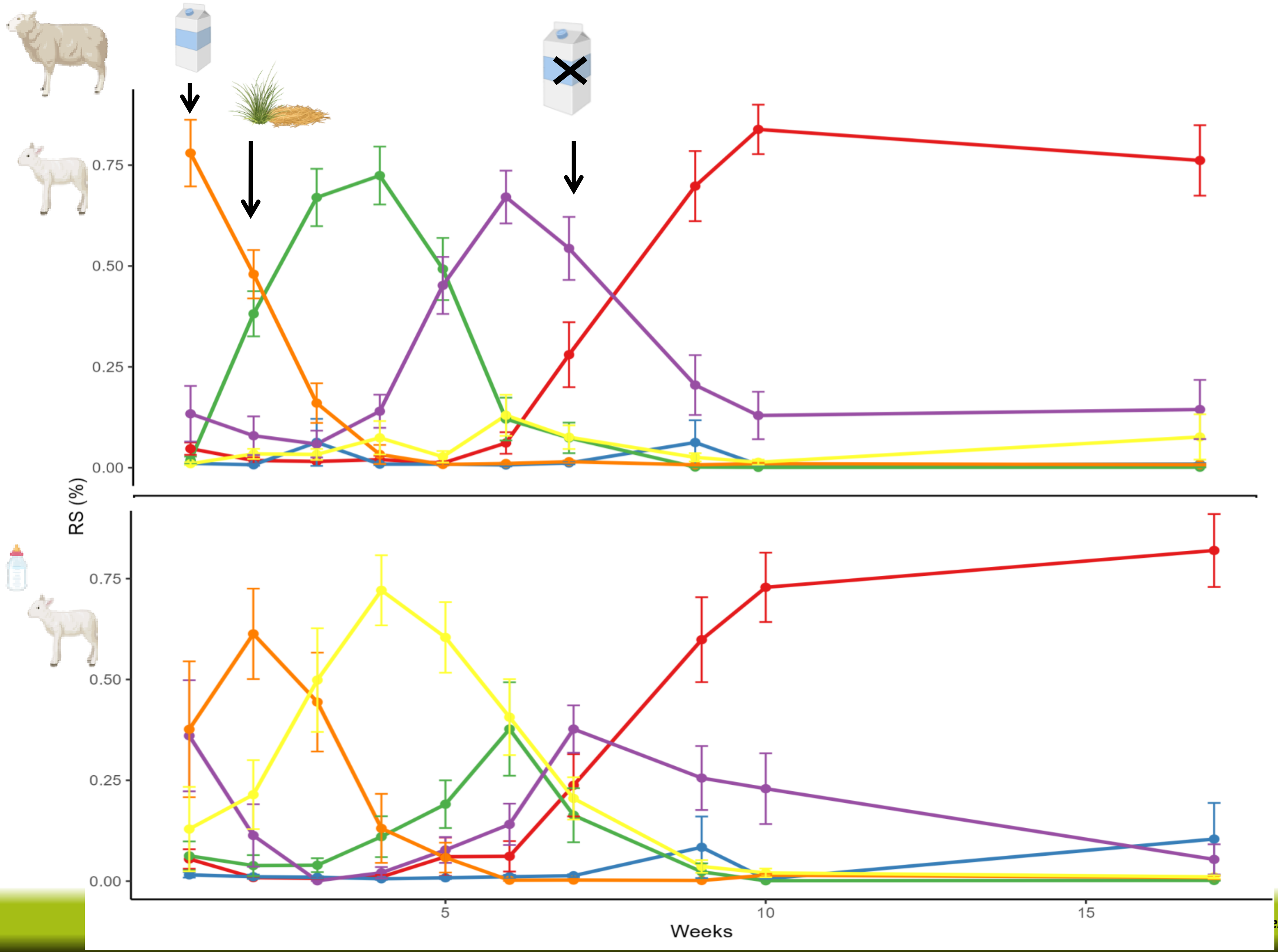


↗ concentrate (Hartinger et al., 2024) : **genus** and *Succinovibrionaceae, Fibrobacteraceae, Prevotellaceae (Amin et Seifert 2021)*

—●—

•RS1 : ***Succinivibrionaceae UCG-001***, ***Prevotella***, *Erysipelotrichaceae UCG-002*

Life lambs : RS apparition



↗ concentrate (Hartinger et al., 2024) : **genus**
and *Succinovibrionaceae, Fibrobacteraceae, Prevotellaceae (Amin et Seifert 2021)*

- **RS1** : ***Succinivibrionaceae UCG-001***, ***Prevotella***, *Erysipelotrichaceae UCG-002*