Heat stress effects on the gut microbiome profile of Iberian purebred and Duroc x Iberian crossbred weaned piglets

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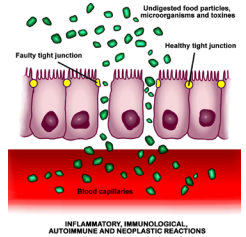
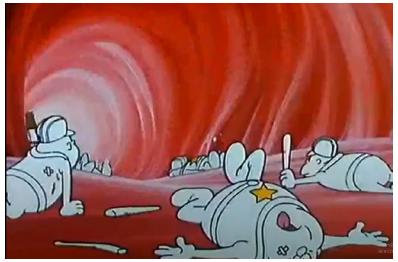
INTRODUCTION

Heat Stress (HS) → Detrimental effect on gut health

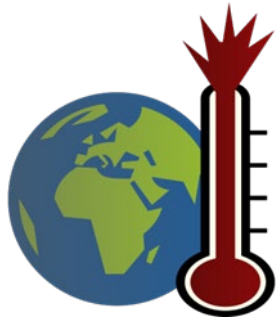
- Blood diverted to periphery to maximize **heat dissipation**. Vasoconstriction in gastrointestinal tract
- **Hipoxia**. Induce **oxidative stress** and **compromise intestinal integrity**
- Increase **intestinal permeability**. Translocation of **endotoxin** and **bacteria** to the **circulation**
- **Immune** response **altered**





INTRODUCTION

Heat Stress (HS)



- **Alter** gut **microbiome** of **livestock** animals

- Changes in microbiota show **interactions** with **host's** metabolism

- Negative consequences related with **duration** of **HS**

OBJECTIVE

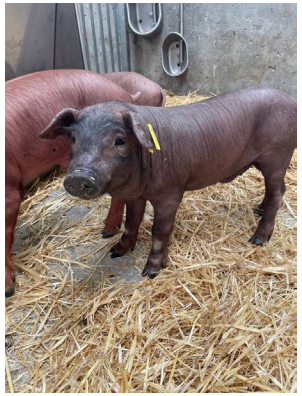
To investigate the **impact** of **HS** on the **gut microbiome** of **weaned pigs** from

Iberian purebred (IB)

Duroc x Iberian crossbred (DUIB)

MATERIAL

Iberian (n =20, 2 batches)



Duroc x Iberian (n =20, 2 batches)

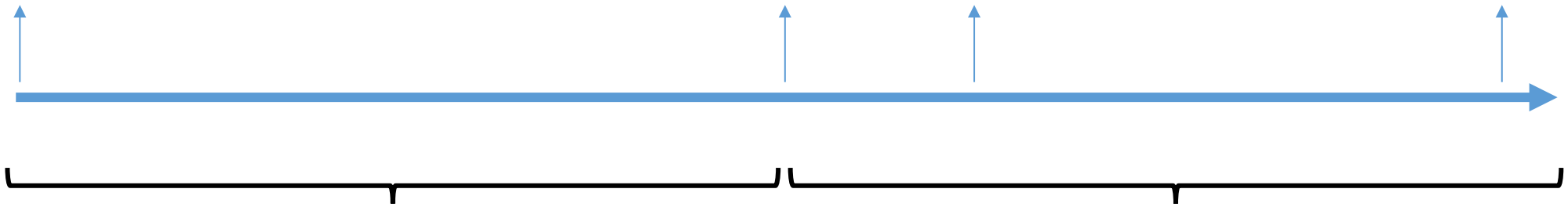
Body weight: 15.70 ± 3.26 kg
Individual pen
Individual Feed Intake

Sampling points
feaces

Day 0 (T0)

Day 2 (T2; Acute HS)

Day 7 (T7; Chronic HS)



7 days Confort Temperature (CT: 22ºC)

7 days Heat Stress (HS: 30ºC)

METHODS

Bacterial DNA extraction from faeces

↓

Pair-end libraries / V3 – V4 Illumina MiSeq

↓

qiime2: Reads filtering and proccesing

↓

Silva (release 138): Taxonomic assignments

↓

Phyloseq: Microbial composition and diversity analyses

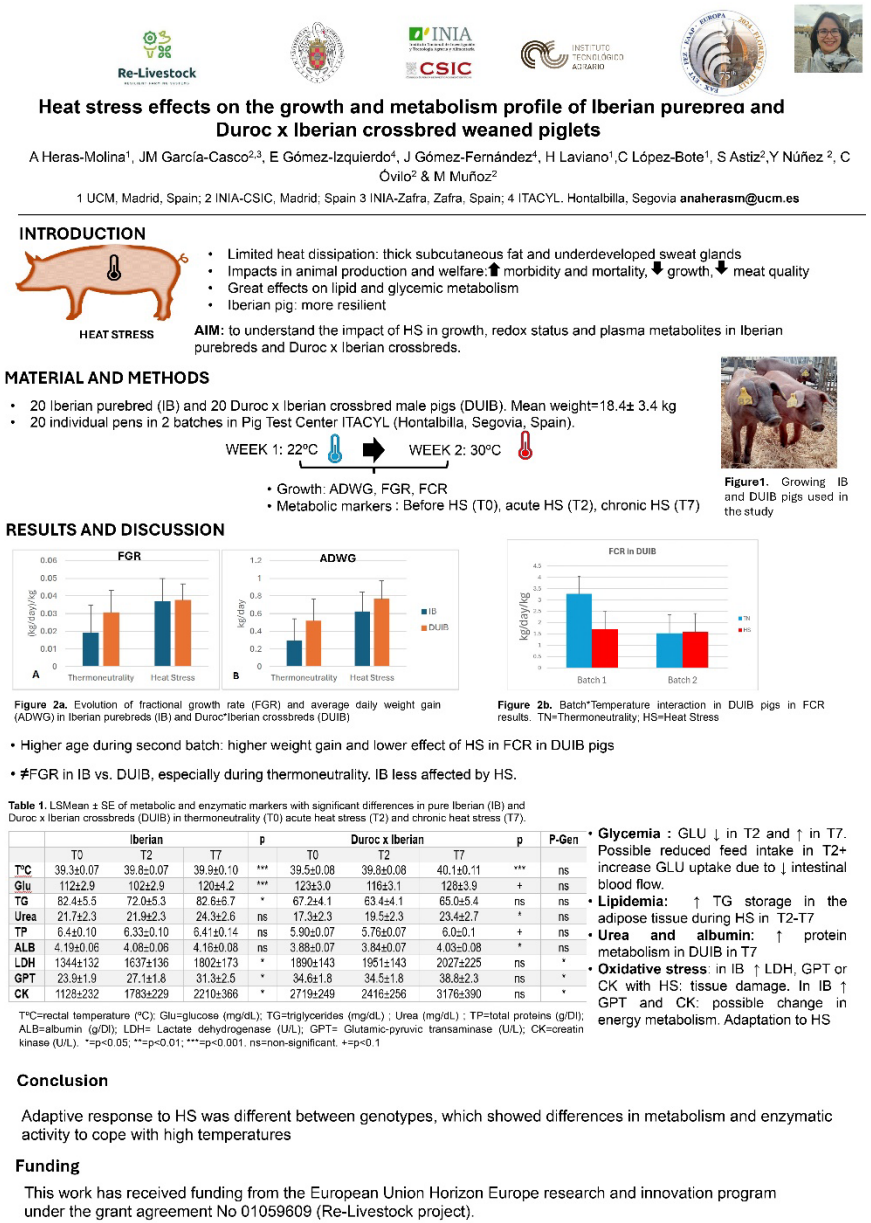
↓

zCompositions & **easyCODA**: Compositional analyses

↓

Linda: Differential Abundance Analyses

RESULTS



Heat stress effects on the growth and metabolism profile of Iberian purebred and Duroc x Iberian crossbred weaned piglets

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INTRODUCTION

HEAT STRESS

- Limited heat dissipation: thick subcutaneous fat and underdeveloped sweat glands
- Impacts in animal production and welfare: ↑ morbidity and mortality, ↓ growth, ↓ meat quality
- Great effects on lipid and glycemic metabolism
- Iberian pig: more resilient

AIM: to understand the impact of HS in growth, redox status and plasma metabolites in Iberian purebreds and Duroc x Iberian crossbreds.

MATERIAL AND METHODS

- 20 Iberian purebred (IB) and 20 Duroc x Iberian crossbred male pigs (DUIB). Mean weight=18.4± 3.4 kg
- 20 individual pens in 2 batches in Pig Test Center ITACYL (Hontalbilla, Segovia, Spain).

WEEK 1: 22ºC → WEEK 2: 30ºC

- Growth: ADWG, FGR, FCR
- Metabolic markers : Before HS (T0), acute HS (T2), chronic HS (T7)

Figure1. Growing IB and DUIB pigs used in the study

RESULTS AND DISCUSSION

Figure 2a. Evolution of fractional growth rate (FGR) and average daily weight gain (ADWG) in Iberian purebreds (IB) and Duroc*Iberian crossbreds (DUIB)

Figure 2b. Batch*Temperature interaction in DUIB pigs in FCR results. TN=Thermoneutrality; HS=Heat Stress

- Higher age during second batch: higher weight gain and lower effect of HS in FCR in DUIB pigs
- ≠FGR in IB vs. DUIB, especially during thermoneutrality. IB less affected by HS.

Table 1. LSMean ± SE of metabolic and enzymatic markers with significant differences in pure Iberian (IB) and Duroc x Iberian crossbreds (DUIB) in thermoneutrality (T0) acute heat stress (T2) and chronic heat stress (T7).

	Iberian			p	Duroc x Iberian			p	P-Gen
	T0	T2	T7		T0	T2	T7		
T°C	39.3±0.07	39.8±0.07	39.9±0.10	***	39.5±0.08	39.8±0.08	40.1±0.11	***	ns
Glu	112±2.9	102±2.9	120±4.2	***	123±3.0	116±3.1	128±3.9	+	ns
TG	82.4±5.5	72.0±5.3	82.6±6.7	*	67.2±4.1	63.4±4.1	65.0±5.4	ns	ns
Urea	21.7±2.3	21.9±2.3	24.3±2.6	ns	17.3±2.3	19.5±2.3	23.4±2.7	*	ns
TP	6.4±0.10	6.33±0.10	6.41±0.14	ns	5.90±0.07	5.76±0.07	6.0±0.1	+	ns
ALB	4.19±0.06	4.08±0.06	4.16±0.08	ns	3.88±0.07	3.84±0.07	4.03±0.08	*	ns
LDH	1344±132	1637±136	1802±173	*	1890±143	1951±143	2027±225	ns	*
GPT	23.9±1.9	27.1±1.8	31.3±2.5	*	34.6±1.8	34.5±1.8	38.8±2.3	ns	*
CK	1128±232	1783±229	2210±366	*	2719±249	2416±256	3176±390	ns	*

T°C=rectal temperature (ºC); Glu=glucose (mg/dL); TG=triglycerides (mg/dL) ; Urea (mg/dL) ; TP=total proteins (g/Dl); ALB=albumin (g/Dl); LDH= Lactate dehydrogenase (U/L); GPT= Glutamic-pyruvic transaminase (U/L); CK=creatin kinase (U/L). *=p<0.05; **=p<0.01; ***=p<0.001. ns=non-significant. +=p<0.1

- **Glycemia :** GLU ↓ in T2 and ↑ in T7. Possible reduced feed intake in T2+ increase GLU uptake due to ↓ intestinal blood flow.
- **Lipidemia:** ↑ TG storage in the adipose tissue during HS in T2-T7
- **Urea and albumin:** ↑ protein metabolism in DUIB in T7
- **Oxidative stress**: in IB ↑ LDH, GPT or CK with HS: tissue damage. In IB ↑ GPT and CK: possible change in energy metabolism. Adaptation to HS

Conclusion

Adaptive response to HS was different between genotypes, which showed differences in metabolism and enzymatic activity to cope with high temperatures

Funding

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Poster number 21.14

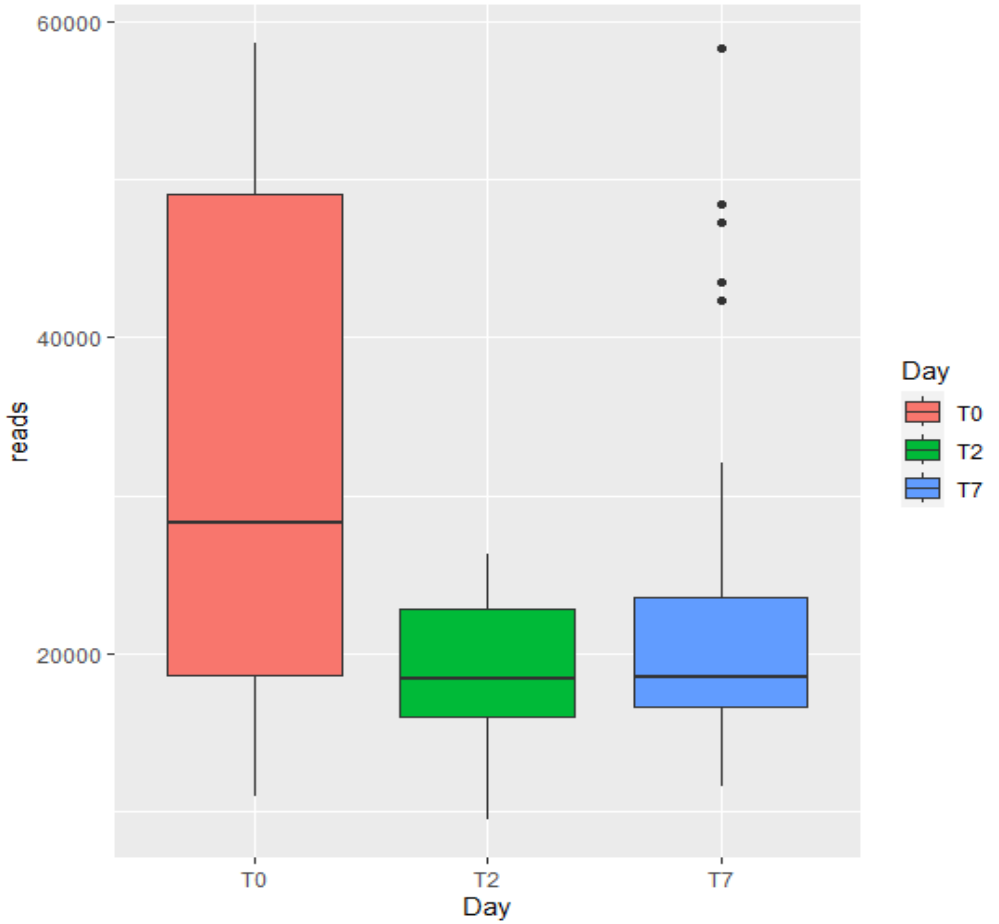
- ≠ growth rate in IB vs. DUIB. IB suffered less the HS
- Glucose: T2<T0<T7, both breeds
- Triglycerides: T2<T0<T7, Iberian bred
- ↑ Urea and albumin with HS in Iberian
- ↑ CK, LDH and GPT enzymes with HS in Iberian
- Different responses to HS between genotypes

RESULTS

Filters

- Phred Quality Q > 20
- Exclusion of chimeras
- Keeping bacteria

- 7,869,590 reads
- 4,077 ASVs (removing singletons and reads with less than 0.5% of reads)

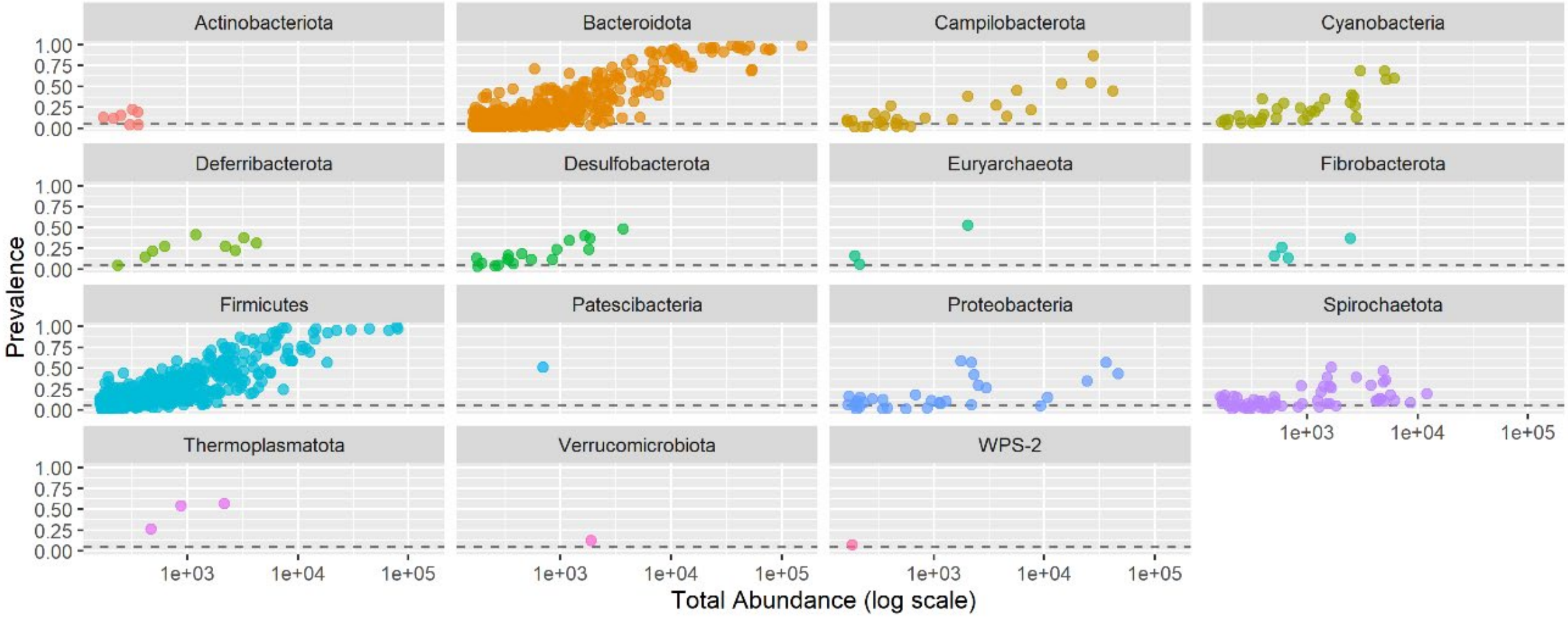


Lower number of reads with increasing temperatures

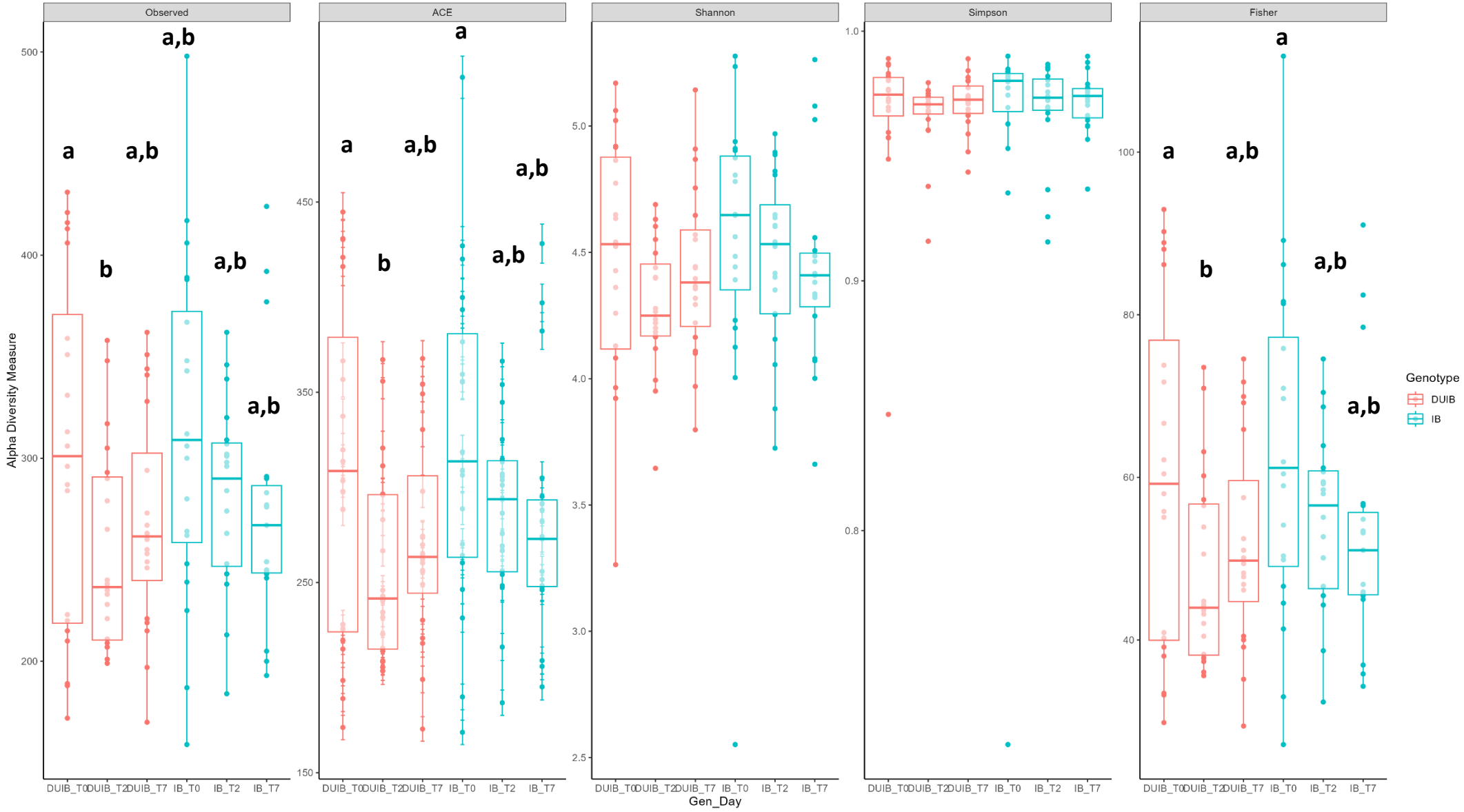
% assigned to phylogenetic levels

Phylum	Class	Order	Family	Genus	Species
100	100	99.8	81.8	68.08	32.1

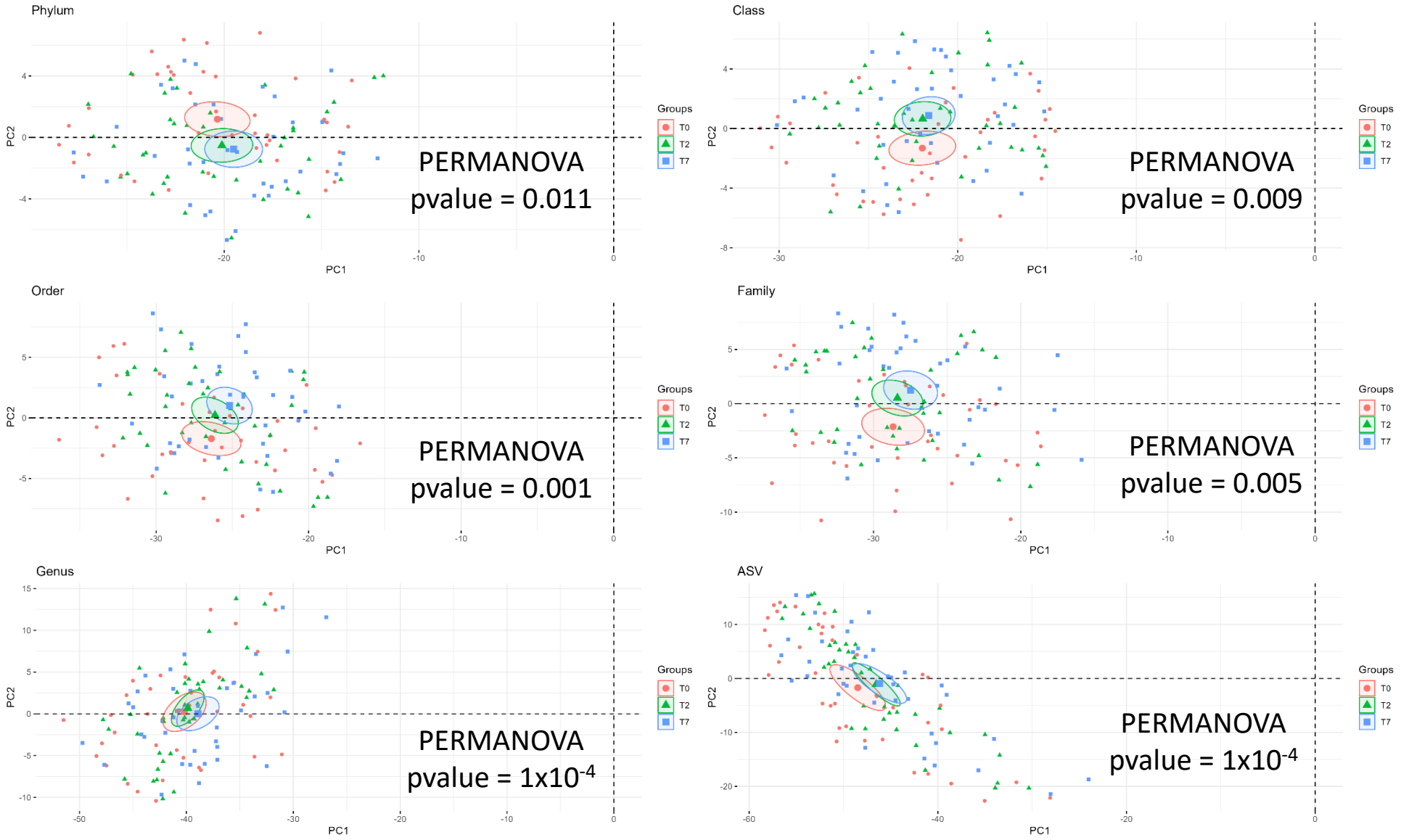
RESULTS


Actinobacteriota
Bacteroidota
Campilobacterota
Cyanobacteria
Deferribacterota
Desulfobacterota
Euryarchaeota
Fibrobacterota
Firmicutes
Patescibacteria
Proteobacteria
Spirochaetota
Thermoplasmatota
Verrucomicrobiota
WPS-2
Prevalence
1.00
0.75
0.50
0.25
0.00
1e+03
1e+04
1e+05
Total Abundance (log scale)

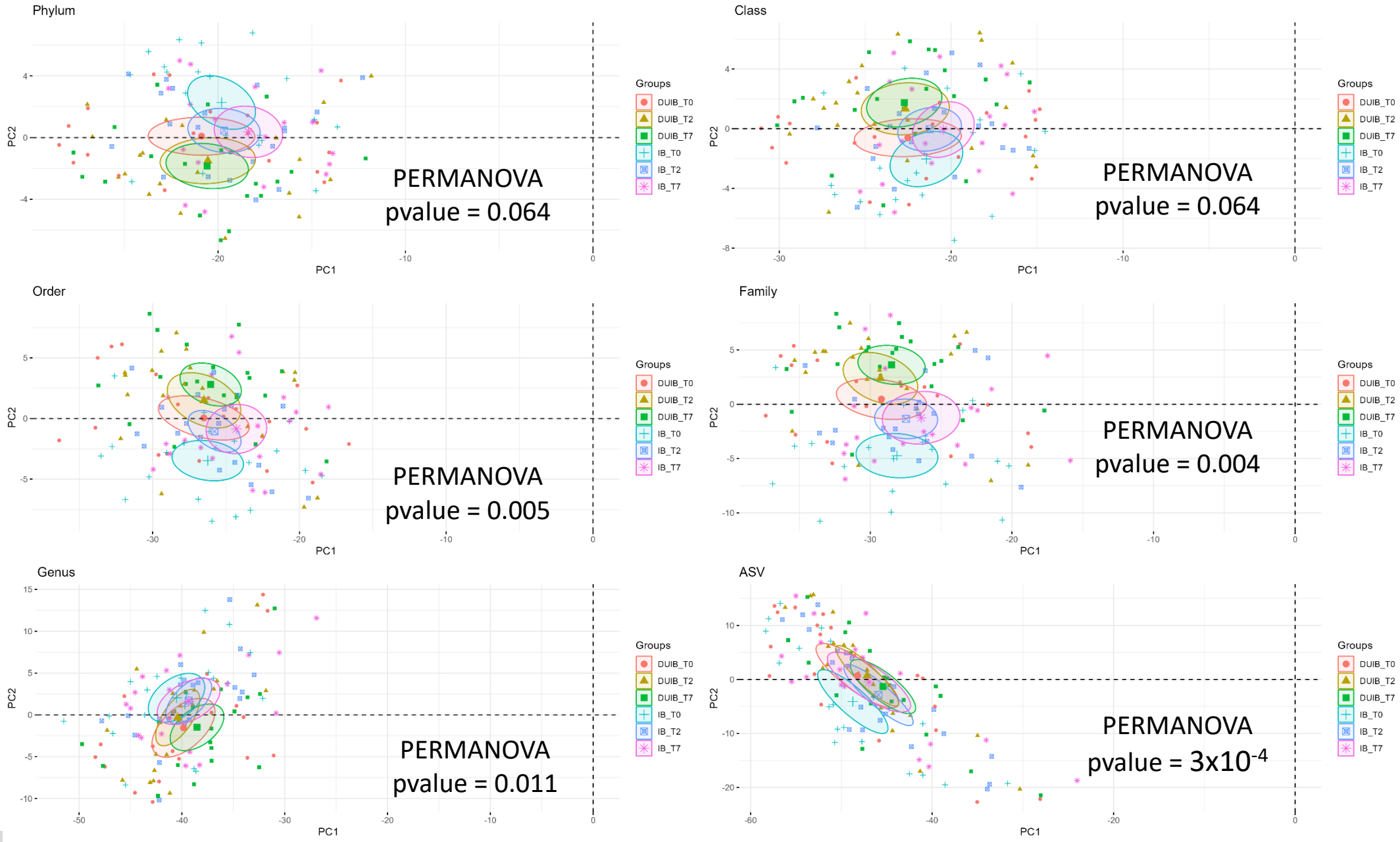
Alpha diversities (ASVs)



Beta-diversities. Day



Beta-diversities. Genotype_Day



Differential abundance analyses: genus

$log_2FC = |0.58|$ & p-adjusted-value<0.10

Genotype. DUIB vs IB

7 genus: *Family_XIII_AD3011_group, Lachnospiraceae_UCG-010, Mucispirillum, uncultured, Intestinimonas, T34, Helicobacter, Terrisporobacter*

Heat Stress

T0 vs T2: 0 genus
T2 vs T7: 2 genus: *Peptococcus, [Eubacterium]_coprostanoligenes_group*
T0 vs T7: 2 genus: *Clostridium_sensu_stricto_1, Peptococcus*

Within genotype

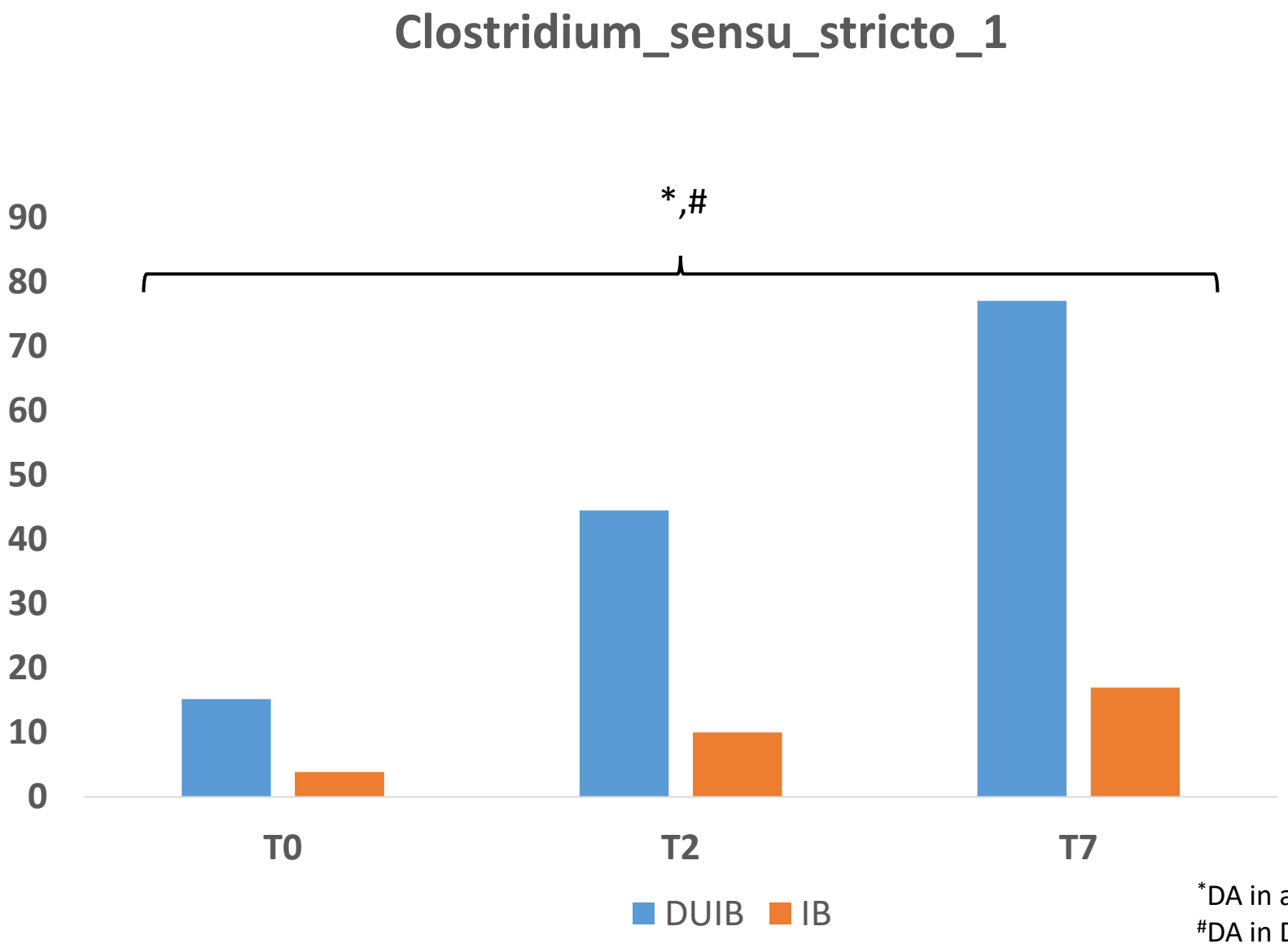
DUIB

T0 vs T2: 0
T2 vs T7: 1 genus: *Oscillibacter*
T0 vs T7 : 2 genus: *Clostridium_sensu_stricto_1, Peptococcus*

IB

T0 vs T2. 3 genus: *Blautia, Alloprevotella, T34*
T2 vs T7. 0
T0 vs T7. 0

Differential abundance analyses genus

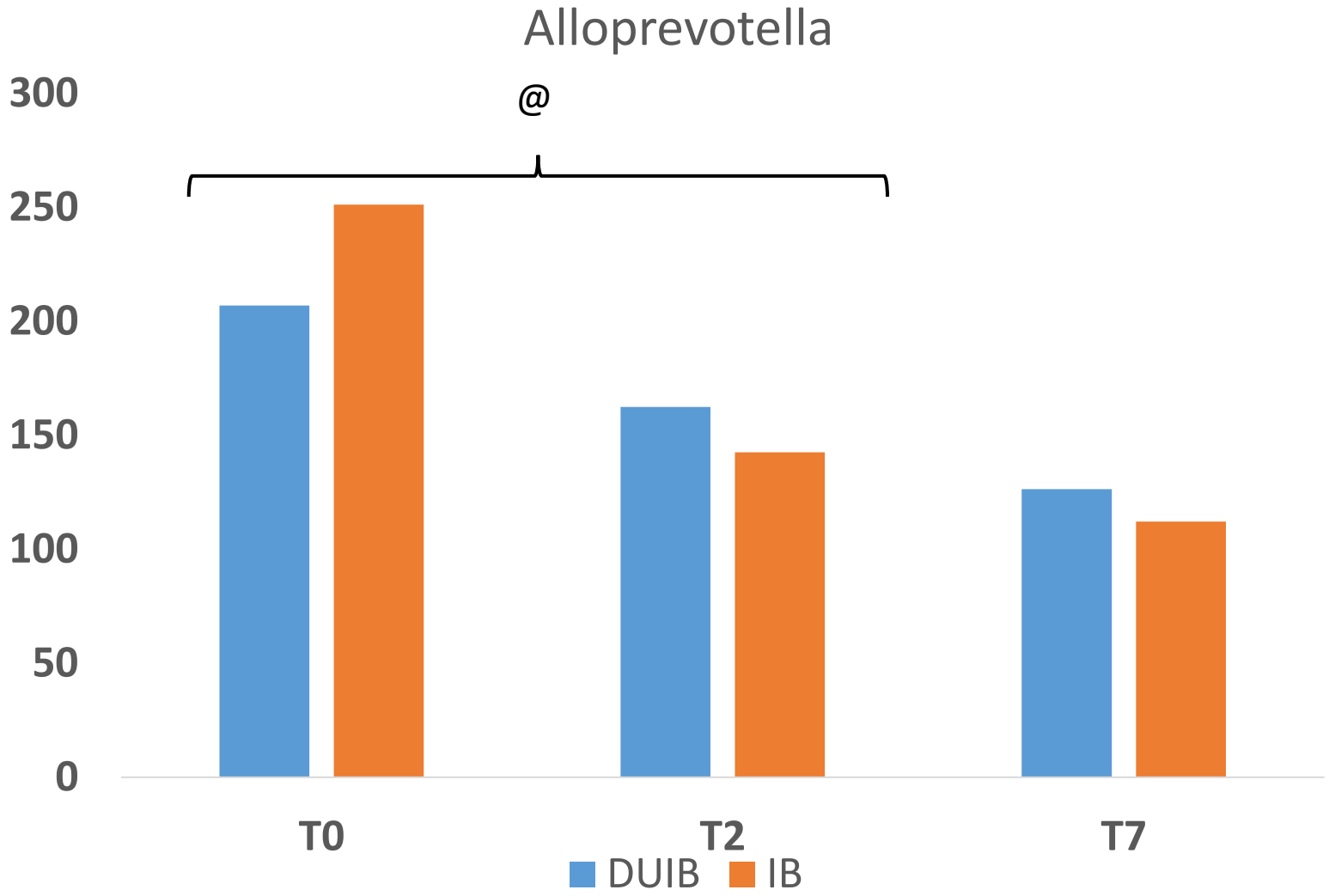


- Opportunistic pathogen
- Intestinal inflammation
- Decrease SCFAs content

No statistical difference between sampling points in Iberian

*DA in averaged breeds
#DA in DUIB

Differential abundance analyses genus



- Decrease in IB
- Trend in DUIB
- Production SCFAs

@DA in IB

Remarks

- Phyla predominant: Firmicutes & Bacteroidota

- Alpha-diversity: statistical differences in DUIB (T0vsT2) for Observed, ACE and Fisher índices.
  Lower diversity in DUIB T2

- Beta-diversity across time
  - T0 group is the one that clusters the furthest from T2 and T7
  - Differences between genotypes are larger than between sampling points within genotypes

- Low number of genus differentially abundances.
  Clostridium_sensu_stricto_1 & *Alloprevotella*: potential biomarkers to be further studied

Acknowledgments

Re-Livestock
RESILIENT FARMING SYSTEMS

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THANKS FOR YOUR ATTENTION!