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Optimal gut function in monogastric livestock

# *Role of caecal microbiota in broiler flock weight heterogeneity*

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*74th EAAP ANNUAL MEETING –LYON, France, 2023*

This project has received funding from the European Union’s Horizon 2020 research and innovation programme under the Marie Sklodowska-Curie grant agreement No 955374.

# Abstract

## ROLE OF CAECAL MICROBIOTA IN BROILER FLOCK WEIGHT HETEROGENEITY

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Despite improved genetics and farm practices, body weight (BW) heterogeneity yet exists within broiler flocks, causing operational inefficiencies and challenges in processing and marketing. Chicken performance is also linked to gut microbiota, however specific microbiota affecting BW within a flock remains elusive. This study examined caecal micbiota's role in BW differences among birds from two hatching systems (HS, HH, hatch in hatchery and HOF, hatch on-farm). The 454 male broiler chicks for each HS were raised together until day (d) 7 before being split into low (L, n=147) and high (H, n=140) BW groups, made two factorial design (HS × BW) and four groups (HH-L, HH-H, HOF-L, & HOF-H). On d 7, 14 and 38, ten birds in each group were sampled for 16s rRNA gene sequencing. Alpha diversity was analyzed by 2-way ANOVA and β diversity with 2-way PERMANOVA. Differential abundance of taxa was analyzed using LEfSe and BW-correlated genera identified by Spearman correlation analysis. Alpha diversity indices were largly influenced by BW, except for microbial evenness on d 7, which was affected by HS and higher in HOF system. Low BW chickens had higher microbial richness, evenness and diversity on d 38, while only higher richness on d 7. β diversity was solely influenced by BW, not HS, and PCoA plot had distinct BW group clusters on day 7. HS x BW had no significant effect on diversities. LEfSe analysis revealed high BW chickens enriched with SCFA-producing and health-promoting genera e.g. Unclassified Lachnospiraceae, Faecalibacterium and Clostridia family. Interestingly, Ruminococcaceae members and Lactobacillus were abundant in low BW chickens along with Escherichia-Shigella, Enterococcus, Streptococcus and Akkermansia. Correlation analysis further identified some genera positively correlated with BW e.g. Shuttleworthia, while others e.g. Fournierella were negatively correlated. Little HS-related differences on microbial composition were observed, including increased level of Escherichia-Shigella in HH system chickens. The study revealed that microbiota diversity and composition were less influenced by HS and differences in taxa abundances contributed to chicken BW differences within flock.

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# Role of caecal microbiota in broiler flock weight heterogeneity

Muhammad Zeeshan Akram

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Midterm meeting – October 11, 2023

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# Hatching systems

- **Commercial hatcheries:** feed and water deprivation and transportation (48-72 h)
- **On-farm hatching:** 18th day incubated eggs transported to broiler house from hatchery
- Immediate access to feed and water
- Early life conditions and environment could shape the gut microbiota of chickens

Jessen et al., 2021
Giersberg et al., 2021

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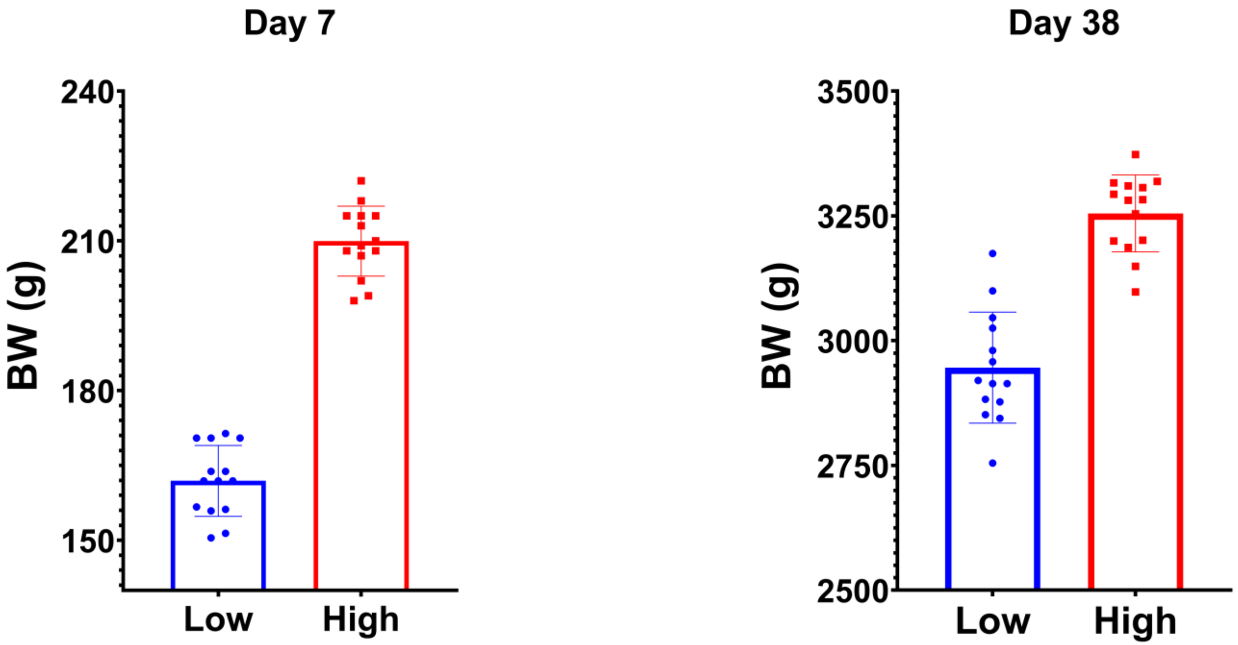
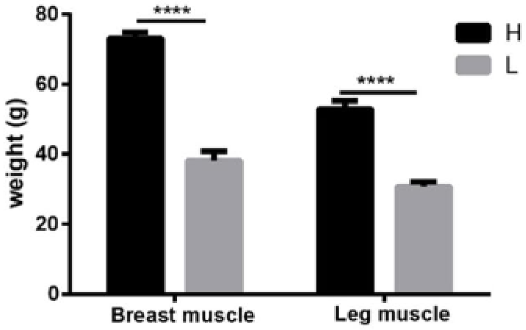
# Flock weight heterogeneity

- Broiler chickens have **different growth rates within a flock** despite being housed under same conditions and fed the same feed

- **Biological factors causing intra flock body weight (BW) variability?**





Zhang et al. Microbiome (2022)

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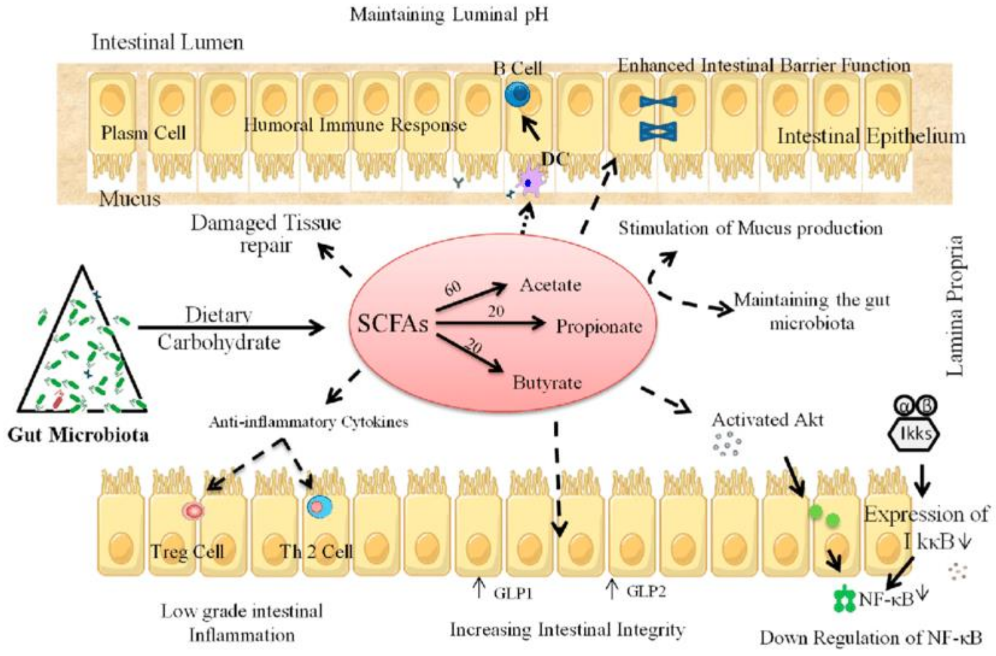
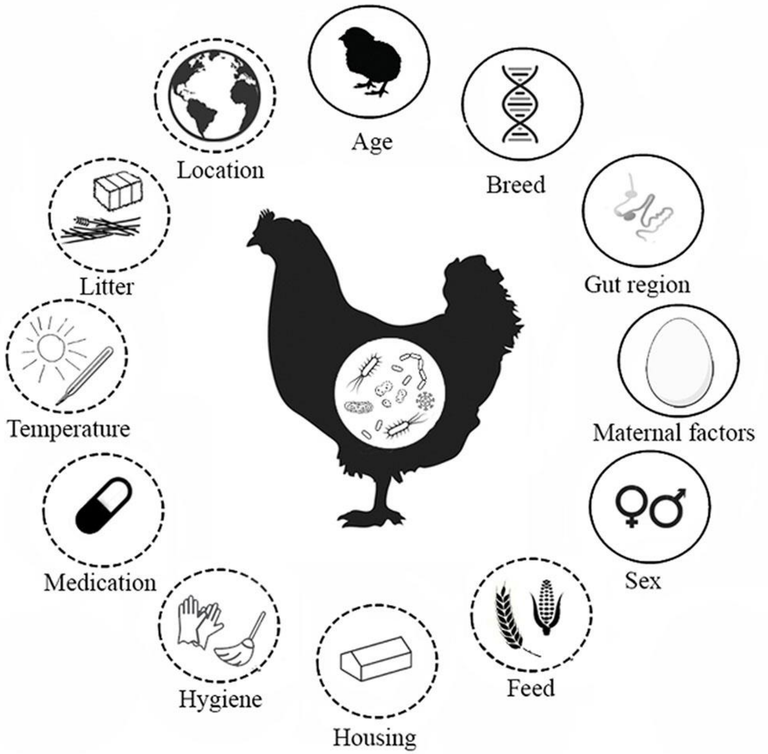
# Gut Microbiota

- Gut microbiota residing in the digestive tract, contribute to digestion, nutrient absorption, metabolism, and immune regulation

- Intestinal microbiota produces high-energy metabolites (short-chain fatty acids), that benefit the host





Front. Microbiol. 9:235. doi: 10.3389/fmicb.2018.00235

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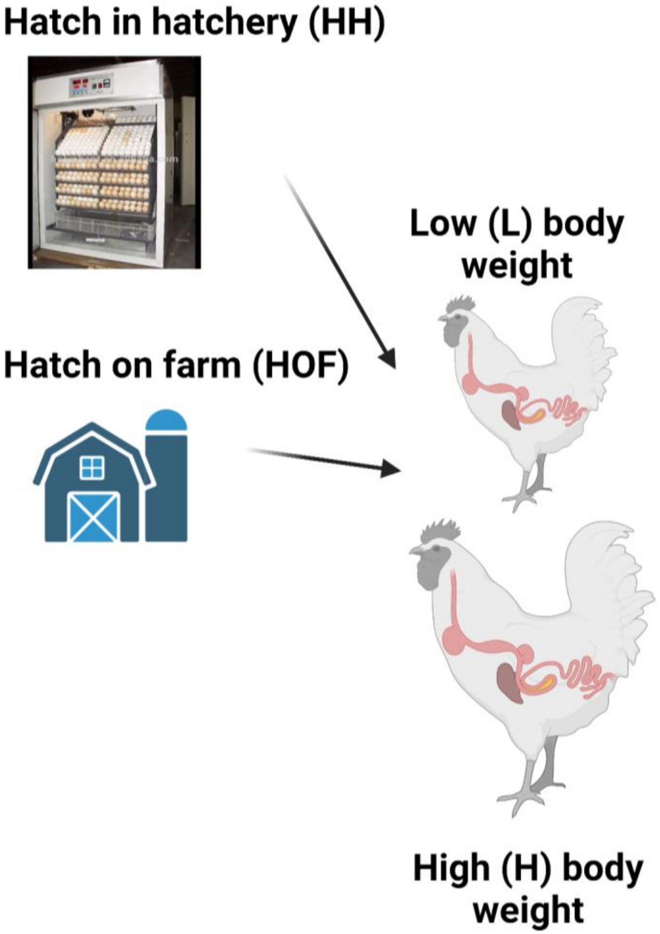
# Hypothesis and aim



- Broilers of different body weights could have different microbial community structure, being housed in the same barn
- Hatching system could shape the microbiota composition that influences the chicken performance and flock uniformity

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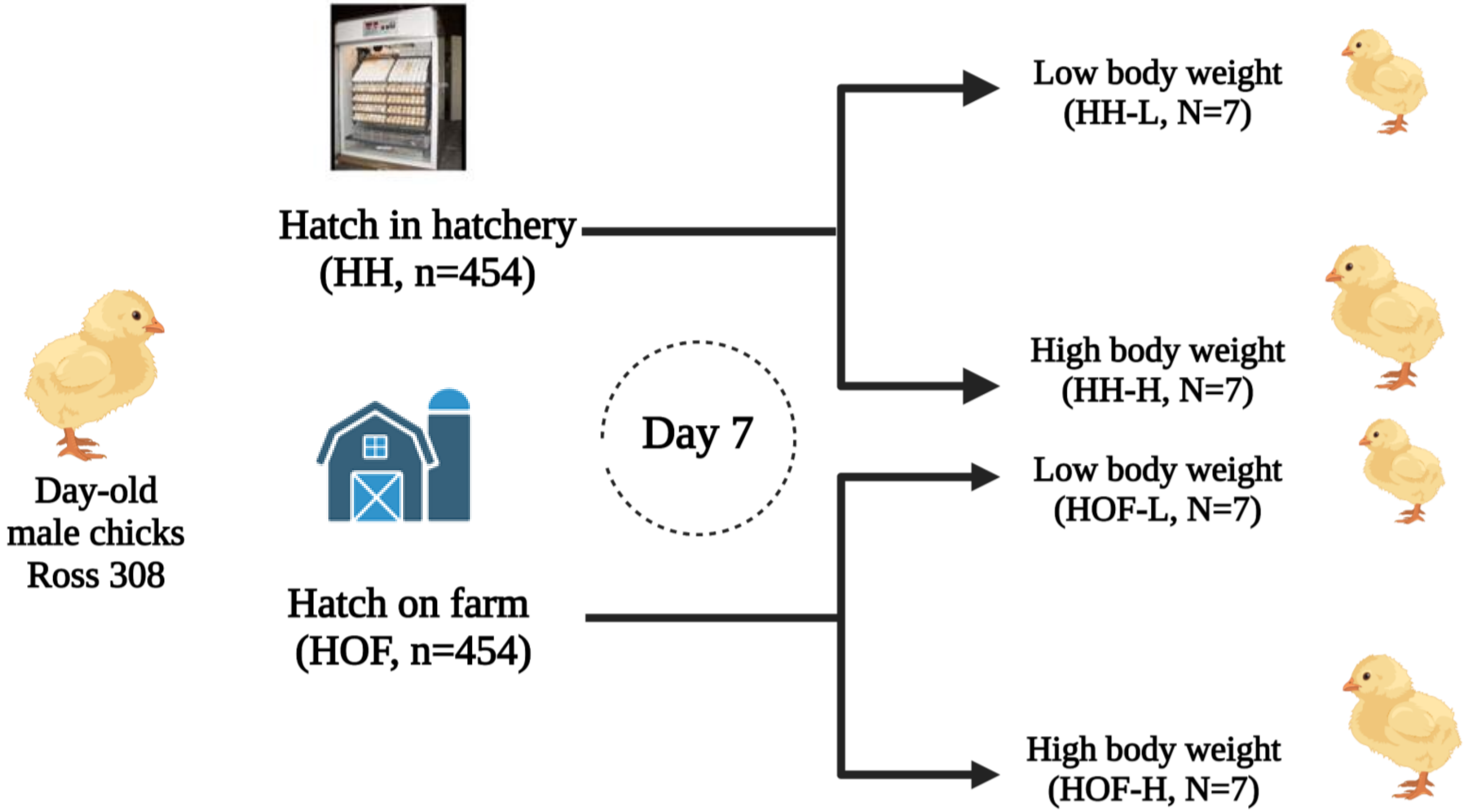
# Experimental outline



N = number of pens
n = number of animals

**Experimental design = 2x2 (Body weight x Hatching system)**

Middle body weight chickens (n=90) from both hatching systems excluded from the experiment

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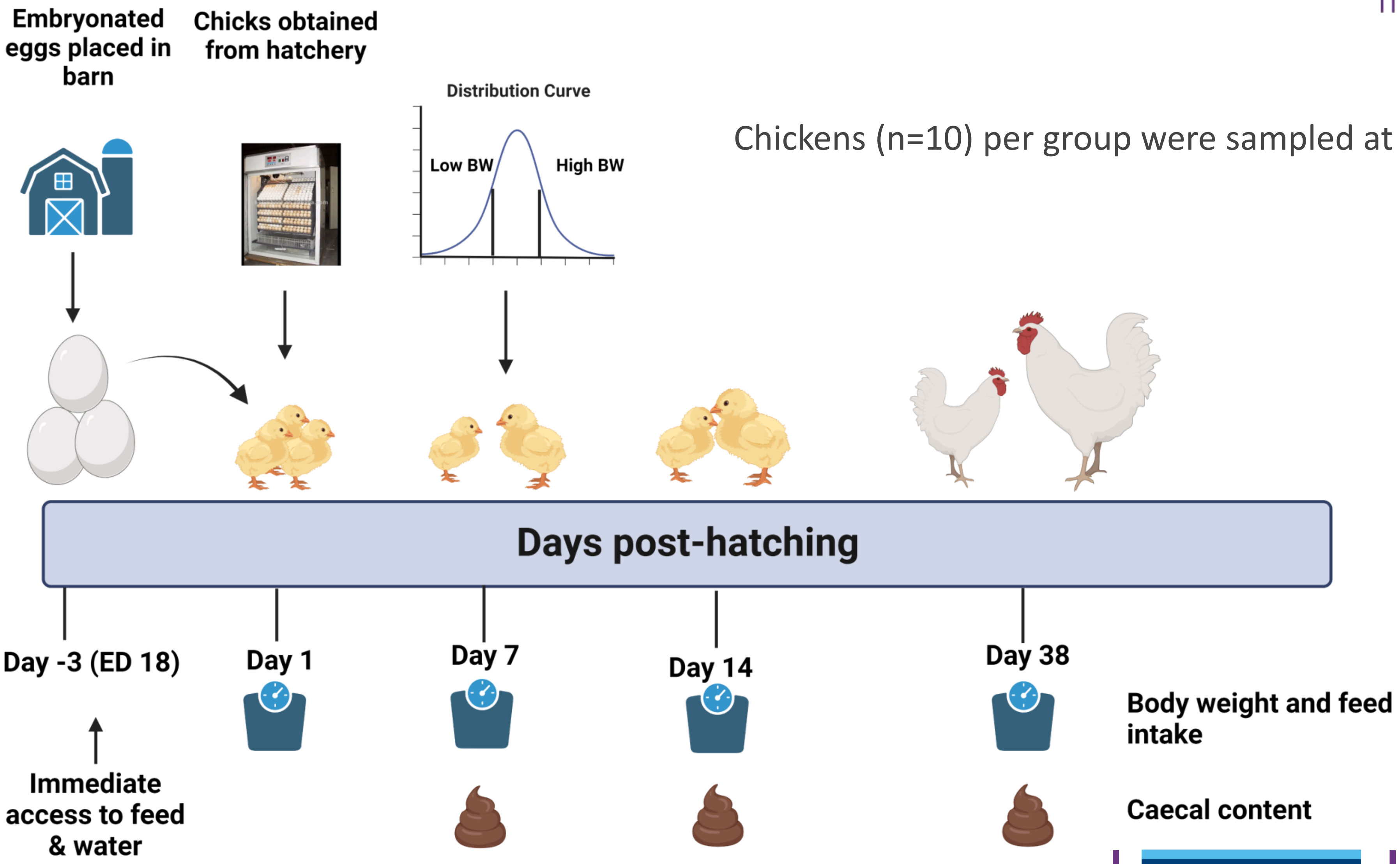
# Experimental timeline

Chickens (n=10) per group were sampled at each timepoint



**Embryonated eggs placed in barn**

**Chicks obtained from hatchery**

**Days post-hatching**

**Day -3 (ED 18)**

**Day 1**

**Day 7**

**Day 14**

**Day 38**

**Immediate access to feed & water**

**Body weight and feed intake**

**Caecal content**

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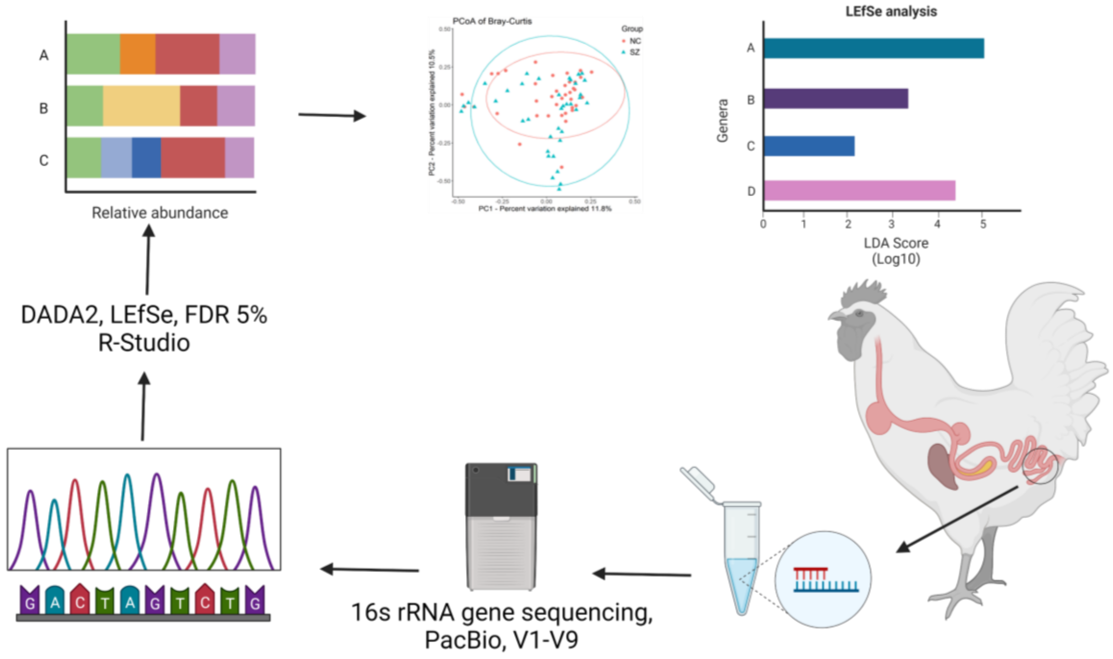
# Methodology

- Caecal microbiota analysis (16s rRNA gene sequencing, PacBio, V1-V9 region)
- Microbiota diversity metrics (Alpha diversity and Beta diversity)
- Linear discriminate analysis effect size (LEfSe) for differential abundance analysis of microbiota, LDA <2, FDR 5%

Table 1: LEfSe differential abundance models for each contrast

| Contrast | Model design |
|---|---|
| Hatching system | HH vs HOF |
| Body weight | L vs H |
| Interaction effects | HS x BW (HH-L, HH-H, HOF-L and HOF-H) |

HS: hatching system, HH: hatch in hatchery, HOF: hatch on farm, BW, body weight, L: low BW groups, H: high BW groups



DADA2, LEfSe, FDR 5%
R-Studio

16s rRNA gene sequencing,
PacBio, V1-V9

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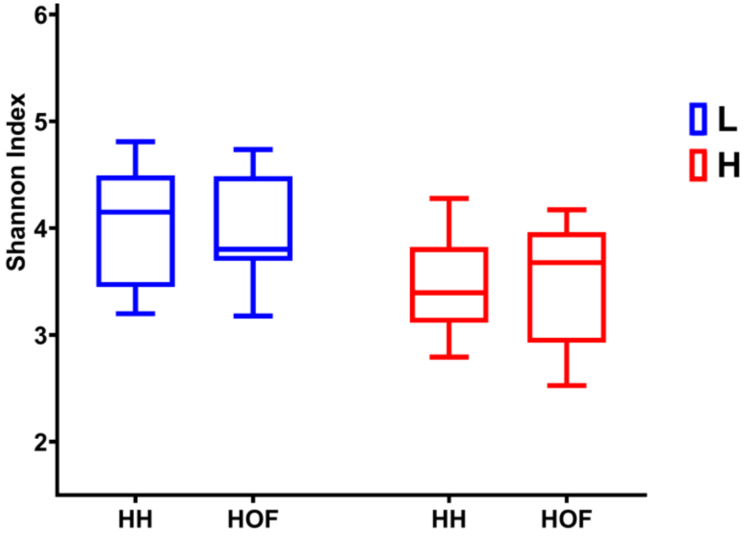
# Bacterial diversity metrics (ASV level)

## Day 7, Alpha-diversity

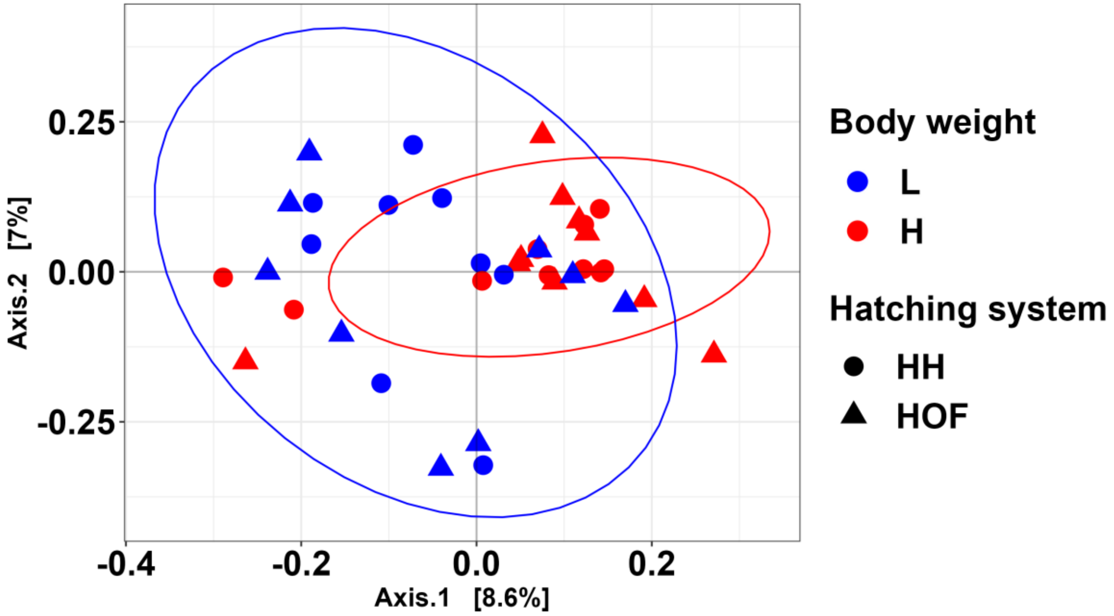
### Shannon



Hatching system: P=NS
BW: NS
Interaction effect: P= NS

## Day 7, Beta-diversity

### Bray Curtis dissimilarity, PCoA plot



Hatching system: P=NS
BW: P=0.003
Interaction effect: P= NS

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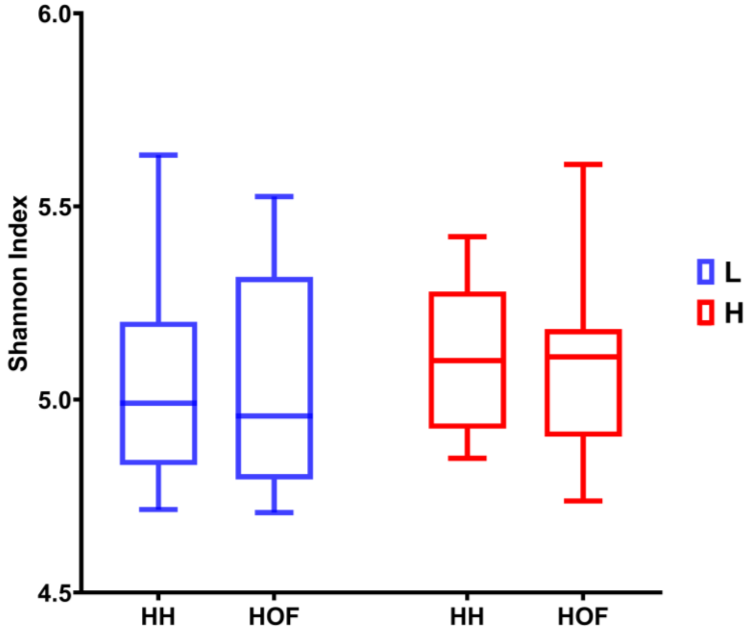
# Bacterial diversity metrics (ASV level)

## Day 14, Alpha-diversity

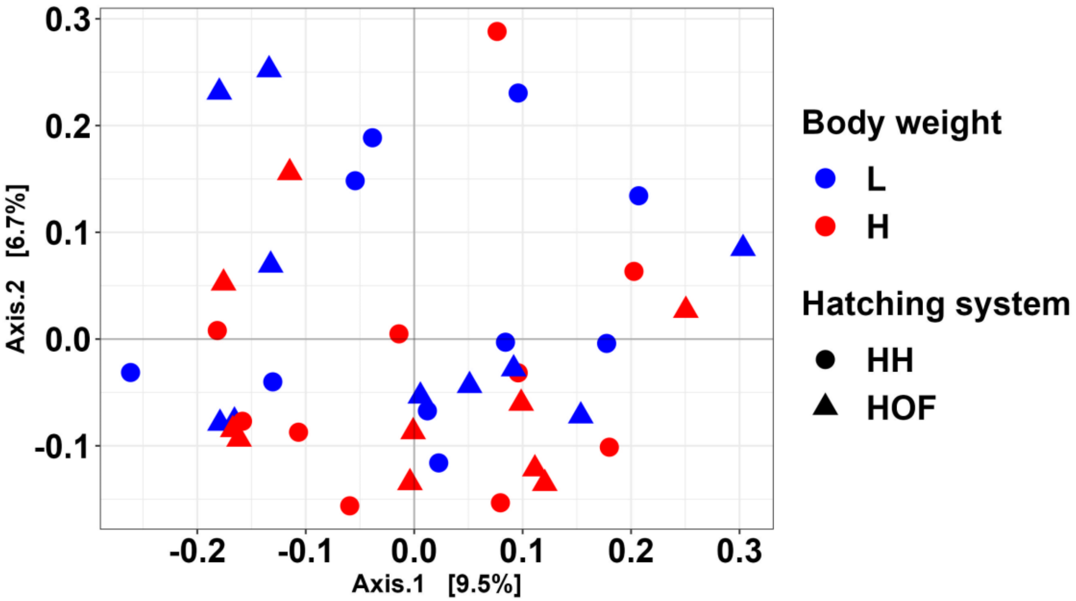
### Shannon



Hatching system: P=NS
BW: NS
Interaction effect: P= NS

## Day 14, Beta-diversity

### Bray Curtis dissimilarity, PCoA plot



Hatching system: P=NS
BW: P= NS
Interaction effect: P= NS

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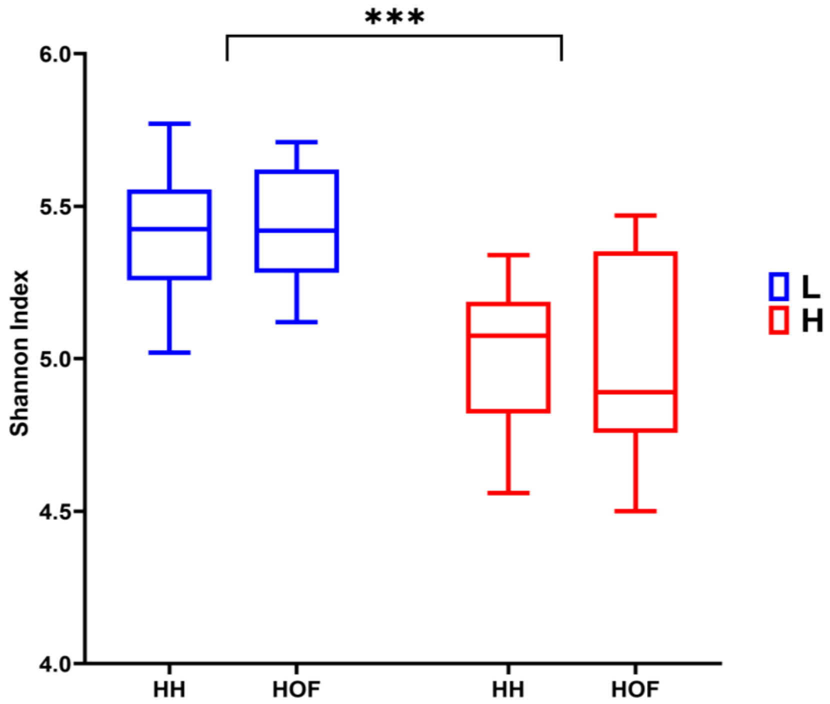
# Bacterial diversity metrics (ASV level)

## Day 38, Alpha-diversity

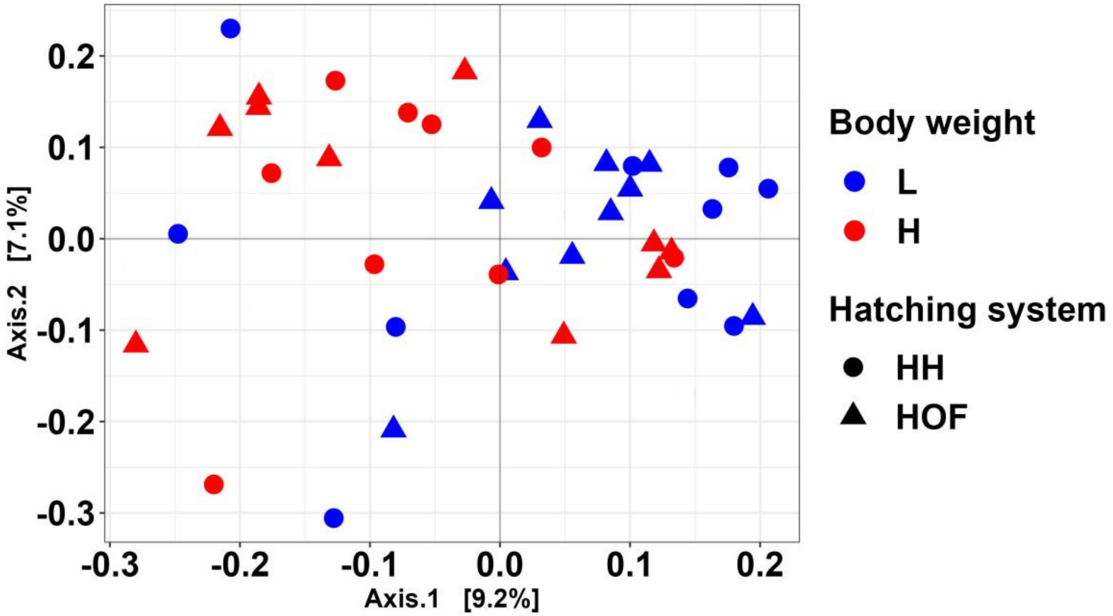
### Shannon



Hatching system: P=NS
BW: **P<0.001**
Interaction effect: P= NS

## Day 38, Beta-diversity

### Bray Curtis dissimilarity, PCoA plot



Hatching system: P=NS
BW: P=0.076
Interaction effect: P= NS

**Values with a-b superscripts differ significantly <0.05**

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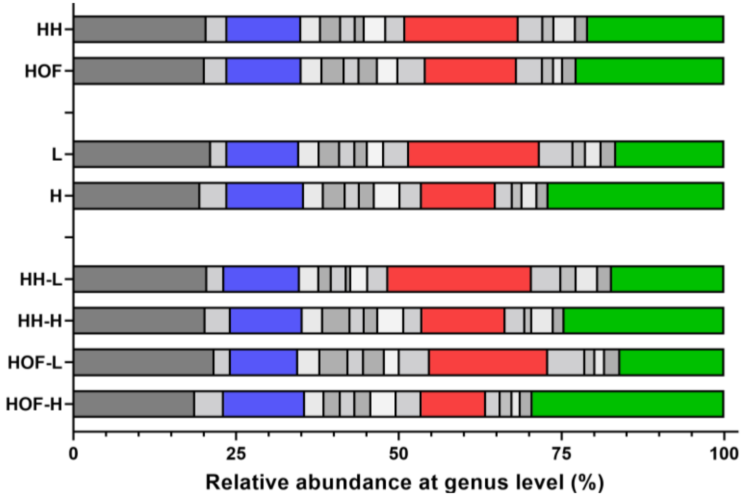
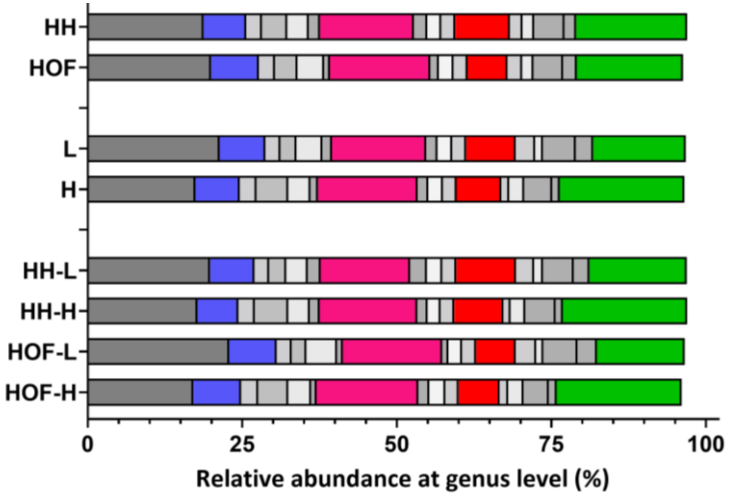
# Microbiota composition of chickens at genus level

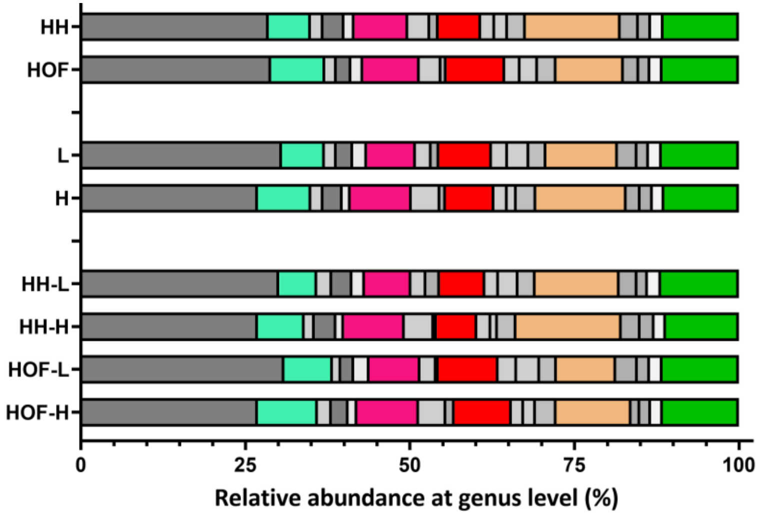
**Day 7**



**Day 14**



**Day 38**



- 🟩 Unclassified Lachnospiraceae
- 🟥 Lactobacillus
- 🟦 [Ruminococcus] torques group
- 🟪 Faecalibacterium
- 🟩 Blautia
- 🟧 Unclassified Clostridia group

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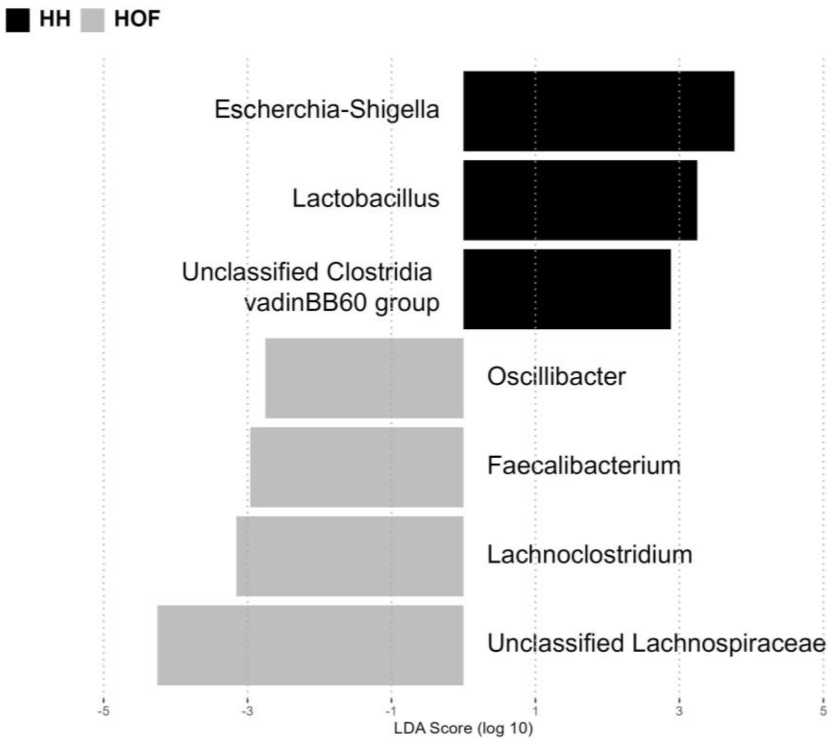
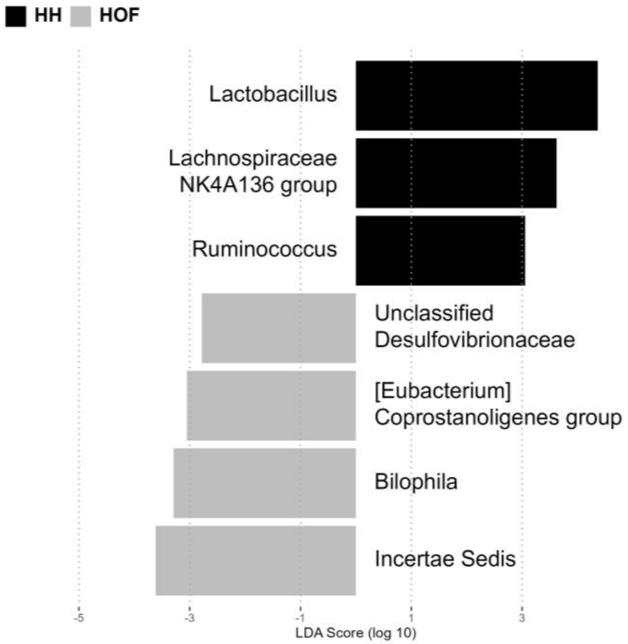
# Differential abundance analysis at genus level between hatching systems – HH vs HOF

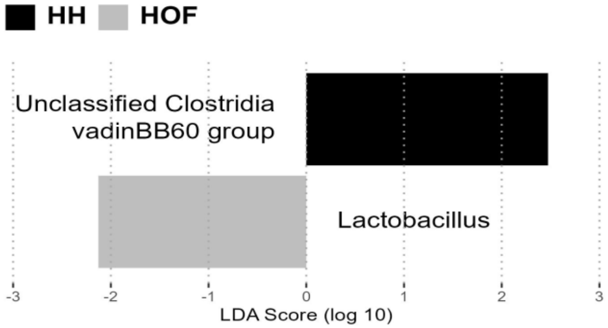
## Day 7



## Day 14



## Day 38



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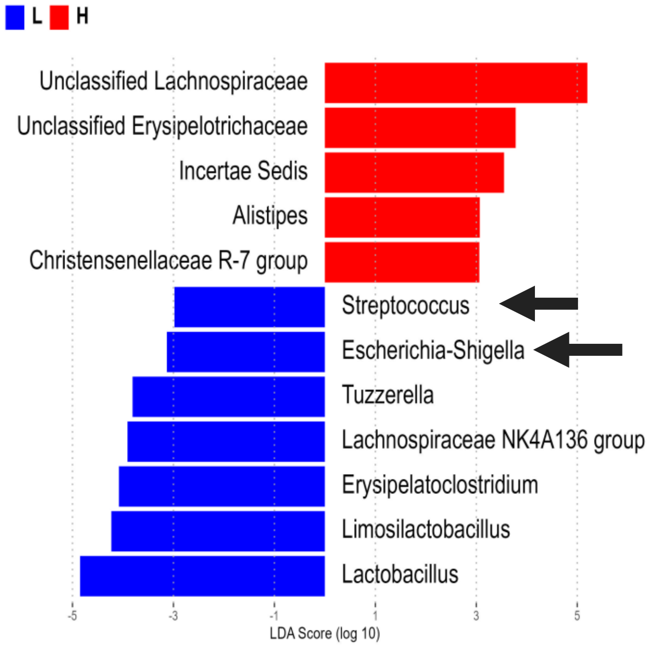
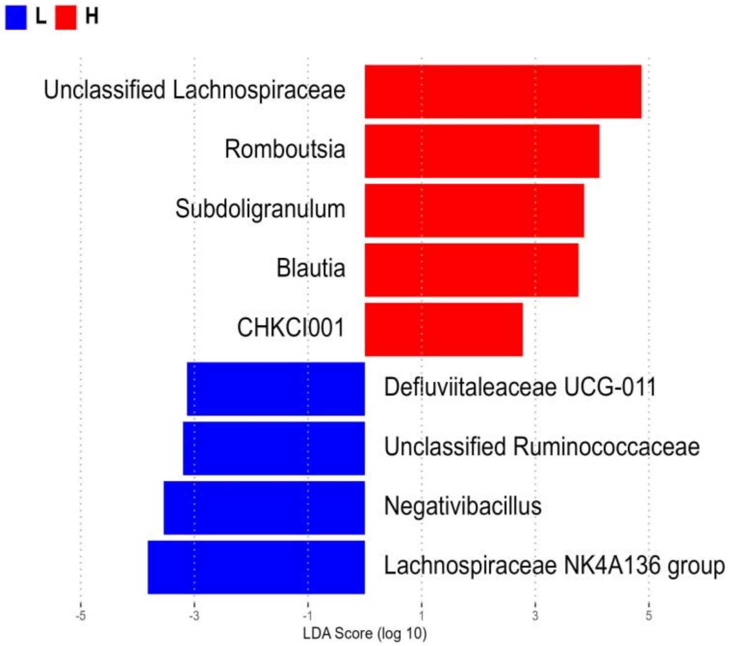
# Differential abundance analysis at genus level between BW groups – L vs H

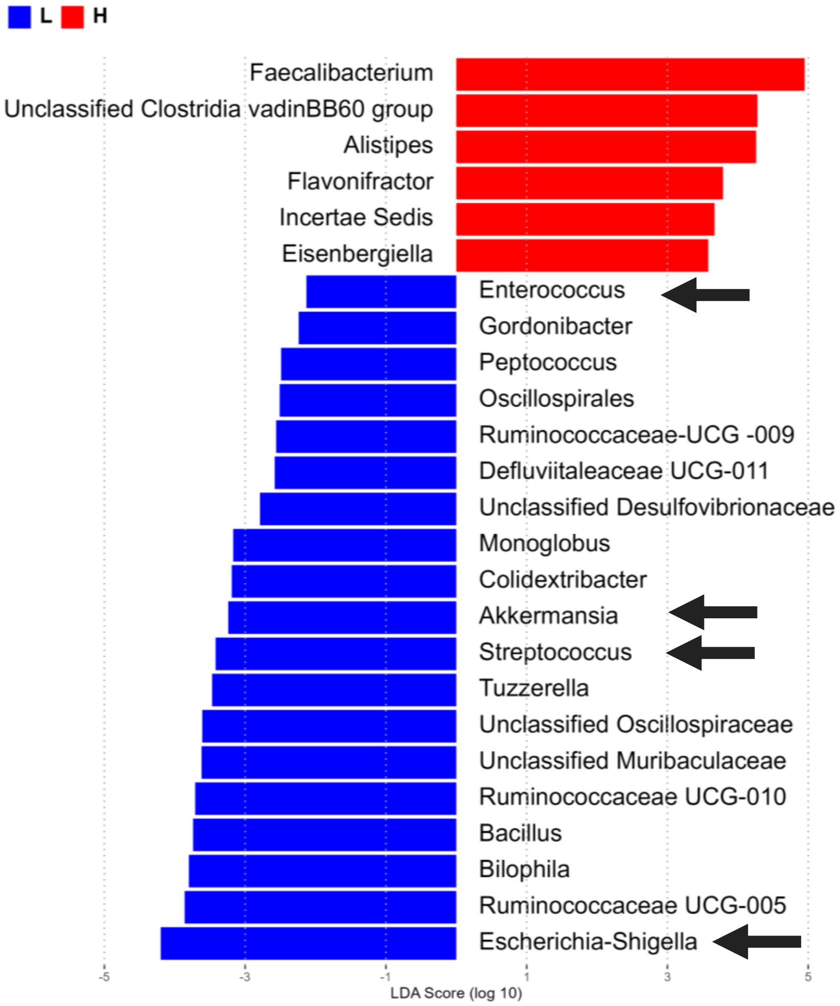
## Day 7



## Day 14



## Day 38



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# Conclusion

1. Hatching systems have minor effects on microbiota diversity and composition, and microbiota differences diminish over time.
2. High body weight chickens exhibited an increased abundance of bacteria, which are considered growth-promoting in nature.
3. Low body weight is linked to higher levels of potential pathogenic bacteria in chickens.
4. Specific bacterial taxa are associated with chicken body weight, suggesting their significant role in intra-flock body weight variation.

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# THANK YOU

## Do you have any questions?

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